**Model exploration of carbon feedbacks between microalgae and heterotrophic bacteria**

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**Project Goals: The LLNL Bioenergy SFA seeks to support sustainable and predictable bioenergy crop production through a community systems biology understanding of microbial consortia that are closely associated with bioenergy-relevant crops. We focus on host-microbial interactions in algal ponds and perennial grasses, with the goal of understanding and predicting the system-scale consequences of these interactions for biomass productivity and robustness, the balance of resources, and the functionality of surrounding microbial communities. Our approach integrates 'omics measurements with quantitative isotope tracing, characterization of metabolites and biophysical factors, genome-enabled metabolic modeling, and trait-based representations of complex multi-trophic biological communities, to characterize the microscale impacts of single cells on system scale processes.**

Eukaryotic microalgae and prokaryotic cyanobacteria (often collectively described as algae) have been proposed as a promising commercially viable feedstock for biofuels and other bioproducts. Open pond systems offer the most feasible and cost-effective way of scaling up biofuel production. However, productivity can be limited by fluctuations in environmental conditions, as well as competitive and trophic (e.g. prey-predator) interactions. While some interactions can negatively impact algal productivity, certain interactions can enhance productivity or mediate factors limiting productivity such as nutrient limitation. A major interaction between algae and bacteria is widely observed, where algae fix inorganic carbon (C) and release dissolved organic C for bacterial consumption. Under certain conditions, an exchange occurs, whereby bacteria release resources that may enhance microalgal growth including dissolved inorganic C (DIC) or nitrogen (N), phosphorus (P), iron, and vitamins. Similarly, bacteria may enhance algal growth through the reduction of inhibitory by-products such as oxygen.

In this work, *Phaeodactylum tricornutum*, a biofuel relevant microalgal species, was grown in laboratory co-cultures with and without a heterotrophic bacterium under cyclical light conditions. *P. tricornutum* grown in co-culture showed higher biomass yield than the axenic culture by 10%. Coupled with earlier experimental findings (Mouget et al., 1995; Samo et al., 2018), we proposed a positive feedback loop between algae and associated heterotrophic bacteria that enhances productivity of the system. Specifically, we hypothesize that algal exudation of fixed dissolved organic C (DOC) fuels growth of heterotrophic bacteria, leading to DOC re-mineralization and increased DIC availability for the algae, relieving limitation of carbon fixation. The strength of this interaction is dependent on environmental and biological factors such as exchange of $CO_2$ across the air/water interface, combined with algal and bacterial traits. Studies to date have yet to focus on the synergistic effects of these factors on productivity.

To test the carbon (DOC-DIC) feedback hypothesis, we developed a dynamic energy budget model of the algae and interacting heterotrophic bacteria. Model simulations were benchmarked against experimental biomass carbon data. A trait based modeling approach was then utilized to explore productivity under scenarios representing interacting gas exchange efficiencies, bacterial-algal respiration traits, and N or P uptake rates. Simulated exchange of $CO_2$ and $O_2$ across the air/water interface demonstrates how mass transfer efficiency impacts dissolved $CO_2$ (DIC) and $O_2$ (DO) concentrations. In well-mixed systems with rapid atmospheric exchange, DIC and DO concentrations are in equilibrium with atmospheric $CO_2$ and $O_2$. Under such a scenario, with no DIC and DOC accumulation, no enhanced productivity is observed due to algal-bacterial interactions. Inefficient atmospheric exchange however leads to accumulation of DIC and DOC, allowing DIC and DOC exchange between the algae and the bacteria, enhancing productivity. Model representation of the effect of $O_2$ affinity for RubisCO shows that DIC:DO ratios control the kinetics of RuBisCO and therefore, bacterial production of DIC or reduction of $O_2$ should enhance the C fixation rate, strengthening the algal-bacterial carbon feedback loop. This interaction is optimal when heterotrophic bacteria have traits that confer high respiration rates but with lower uptake or requirement for N and P than algae. Overall, our results illustrate that algal-bacterial interactions can enhance algal productivity and that that gas exchange efficiencies interact with algal and bacterial traits that can be selected to strengthen the carbon feedback loop.

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## References

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