Examining the role of physical proximity and diffusion of metabolites in algal-bacterial interactions

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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.

Symbioses are governed by the exchange of material and information, encompassing a broad diversity of biomolecules with differing rates of diffusion. We are particularly interested in the role of physical association on algal-bacterial interactions, where exchange will be governed in part by diffusive processes. Although metabolic interactions between algae and bacteria are largely driven by diffusion of metabolites, there has been no systematic approach to control the diffusion process in a culture system. Here, we present research on developing approaches to examine the consequences of spatial proximity on these interactions and community assembly of algal-associated microbiomes. We have taken a dual approach to this problem: first in designing and testing a novel platform based on a biocompatible, nanoporous and structured hydrogel that allows metabolic communication between microbial species without any physical contact (Figure 1), and second by applying metabolite imaging to map diffusion of exuded and exchanged metabolites. Using the hydrogel platform, we have cultured *Phaeodactylum tricornutum,* a model and bioenergy-relevant diatom, in the center of a set of microwells, and examined bacterial community development as a function of distance from the algae. We identified specific bacterial taxa whose relative abundances were affected by proximity to the algal cells, indicating that diffusive processes can play a role in structuring algal-associated microbial communities. Using isolates from our *P. tricornutum*-associated isolate library, we further verify that these bacterial taxa have their own growth characteristics under controlled diffusion of algal exudates. These results indicate that individual bacterial taxa within a community have unique strategies to co-exist with an algal host, allowing us to better understand these complex interactions and their role in structuring the entire community. In parallel, through a partnership with EMSL, we have begun to apply metabolite imaging to characterize metabolites exuded by *P. tricornutum* and a bacterium when grown in close spatial proximity. We have identified a broad range of algal and bacterial metabolites, including some putative plant hormones that may be involved in algal bacterial mutualism. In future research we plan to combine these two approaches to image metabolites in the hydrogel devices in order to identify and test key compounds that play a role in governing these interactions.

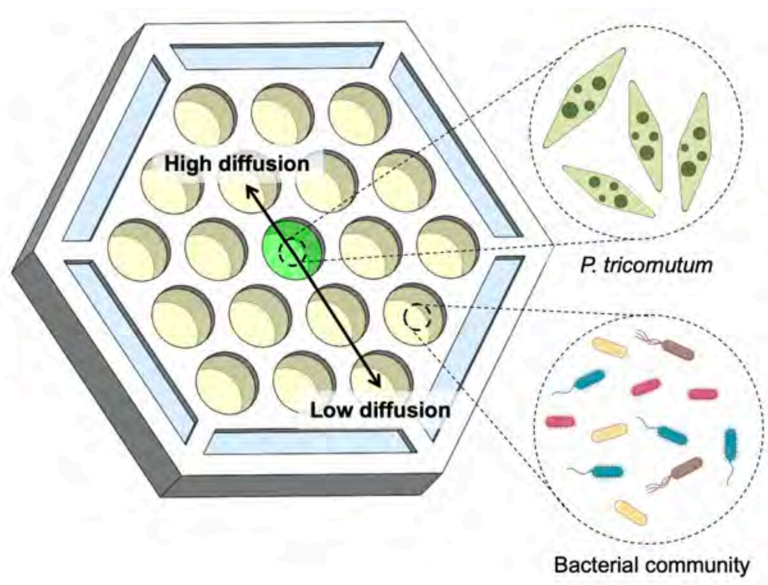

Figure 1. Schematic diagram of co-culture platform for algal and bacterial communities under controlled diffusion of metabolites.

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