Plant-Microbe Interfaces: Microbial responses to the chemical environment of the rhizosphere

Amber B. Webb[1]* (biblean@ornl.gov), Manasa R. Appidi,[2] Paul E. Abraham,[1,2] Sara Jawdy,[1] Jennifer Morrell-Falvey,[1,2] and **Mitchel J. Doktycz**[1,2]

[1]Biosciences Division, Oak Ridge National Laboratory, Oak Ridge, TN; and [2]Genome Science and Technology graduate program, University of Tennessee, Knoxville, TN

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Project Goals: The goal of the PMI SFA is to characterize and interpret the physical, molecular, and chemical interfaces between plants and microbes and determine their functional roles in biological and environmental systems. *Populus* and its associated microbial community serve as the experimental system for understanding the dynamic exchange of energy, information, and materials across this interface and its expression as functional properties at diverse spatial and temporal scales. To achieve this goal, we focus on 1) defining the bidirectional progression of molecular and cellular events involved in selecting and maintaining specific, mutualistic *Populus*-microbe interfaces, 2) defining the chemical environment and molecular signals that influence community structure and function, and 3) understanding the dynamic relationship and extrinsic stressors that shape microbiome composition and affect host performance.

Understanding the interactions among plants and bacteria within the rhizosphere and the role of the chemical environment in shaping these interactions is a central component of the PMI SFA. The ability to visualize and manipulate these interactions in real-time is an invaluable tool in developing our understanding of the mechanisms that regulate these interactions. While prior work has focused on endpoint analysis or imaging individual plants in real time at high resolution using confocal microscopy, we need to develop tools for high throughput, non-destructive imaging of plant-microbe interactions in real time. Here, we describe current efforts to develop such strategies using plants grown in Petri dishes with agar and imaging using a Cytation 5 high content imaging system. With this system, we are optimizing protocols that allow us to assess root colonization by microbes starting with a well-studied isolate, *Pantoea* YR343 labeled with GFP. We are developing these tools as a means of studying the chemical environment of the rhizosphere in the context of a microbial community. These microbial communities are made up of diverse bacterial species that exhibit a variety of sensory capabilities that allow them to respond in different ways to the chemical environment of the rhizosphere. In order to better understand how bacteria respond to the chemical environment of the rhizosphere, we utilized a proteomics approach that resulted in the identification of many different proteins that are upregulated in *Pantoea* YR343 in the presence of a plant. Currently, we are applying high-throughput imaging approaches, combined with genetic tools to develop a robust platform for analyzing the role of select proteins in plant-microbe interactions in the context of the rhizosphere and its local chemical environment.

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