

## **Plant-Microbe Interfaces: Altered root metabolome composition impacts microbiome composition in *Populus PdKOR1* RNAi plants**

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**Project Goals:** The goal of the PMI SFA is to understand the genome-dependent molecular and cellular events involved in establishing and maintaining beneficial interactions between plants and microbes. *Populus* and its associated microbial community serve as the experimental system for understanding how these molecular events manifest themselves within the spatially, structurally, and temporally complex scales of natural systems. To achieve this goal, we focus on 1) characterizing host and environmental drivers for diversity and function in the *Populus* microbiome, 2) utilizing microbial model system studies to elucidate *Populus*-microbial interactions at the molecular level and dissecting the signals and pathways responsible for initiating and maintaining microbial relationships and 3) develop metabolic and genomic modeling of these interactions to aid in interpreting the molecular mechanisms shaping the *Populus*-microbial interface.

In the present study, we examined the effect of altered carbon partitioning and allocation in the *Populus* host due to interactions with individual beneficial microbes as well as on the overall root endosphere and rhizosphere associated microbiome. The altered secondary metabolism host type used in this study are transgenic *Populus PdKOR1* RNAi plants that are downregulated in an endo- $\beta$ -1,4-glucanase gene family member. Gas chromatography-mass spectrometry profiles of *PdKOR1* plants showed a higher phenolic and salicylic acid content, and reduced lignin, sugars, shikimic acid and maleic acid content relative to non-transgenic control. Co-culture with the fungal mutualist, *Laccaria bicolor*, showed enhanced mycorrhization rate and improved biomass production in *PdKOR* plants (Kalluri et al. 2016). This suggested strong potential for impact on the broader microbial community that the plant interacts with in field settings. In contrast, the colonization rate of a previously characterized Gammaproteobacterial isolate, *Pantoea* YR343, was lower in *PdKOR1* RNAi plants.

To test whether altered root metabolome has an effect on the microbiome associated with roots under field settings, we collected root samples from independent ramets of field-grown *PdKOR1* RNAi and control plants and performed Illumina MiSeq 16S rRNA gene sequencing. Bacterial community composition, as measured by Bray-Curtis dissimilarity, differed between *PdKOR1* RNAi and control rhizospheres and roots. *Actinobacteria*, and the family *Micromonosporaceae*, were significantly more abundant, whereas *Nitrospirae* were reduced in *PdKOR1* RNAi plant

rhizosphere. These findings from single isolate co-culture experiments as well as field-based microbiome analyses show the relevance of host carbon partitioning and metabolome composition, including phenolic, sugar, amino acid and fatty-acid composition, on concomitant alterations in root-associated microbial communities. We are currently conducting metagenomics of leaf, stem and root and soil samples to capture the genetic diversity of the microbial communities (bacterial and fungal) associated with specific plant tissue type/niche, and examining the differential molecular pathways underlying the differential association of microbes via RNA-Seq analysis and microbial isolate sequencing approaches (Kalluri et al. 2018).

In conclusion, our study shows the significance of plant metabolome composition on shaping the associated microbiome and is prompting new hypotheses and experiments that will address the cascading effects of host genotype, root tissue environment, root exudate composition on interactions with soil microbiome.

### **References**

Kalluri et al. *Frontiers in Plant Sciences*. 2016. doi: 10.3389/fpls.2016.01455.

Kalluri et al. FY18 DOE Joint Genome Institute Community Sequencing Project.

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