

Plant-Microbe Interfaces: Comparative genome evolution of *Populus* root endophytes

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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.

Endophytic fungi have a strong influence on ecosystems by benefiting the health and survival of their host plants against biological and non-biological stresses. Fungal endophytes are polyphyletic and hyperdiverse, yet their common adaptations that enable an endophytic lifestyle are largely unknown. We sequenced genomes of over 40 fungal taxa representing 17 unique lineages of endophytes associated with *Populus*. We compared the 17 endophyte lineages with their closest-related free-living saprobe species to identify common genomic features associated with endophytic symbiosis lifestyle. Among the genomic features we compared, endophytic fungi were observed to consistently have significantly greater genome size and gene count than their sibling taxa, including more CAZymes and more small secreted proteins. To correct bias from the phylogenetic structure, we used phylogenetic linear regression models and phylogenetically independent contrasts to compare genome evolution of each lineage with its nearest non-symbiotic relative. Results of multiple phylogenetically-corrected analyses suggest several mechanisms by which fungi have adapted to an endophytic lifestyle. Most fungal endophytes generally have slower growth rates, lower competitive ability (as free-living fungi), while still maintaining a certain amount of saprobic ability as well as ability to communicate with their plant host. To further differentiate the exact genes contributing to endophytic lifestyle, we used an AIC model selection to identify a core set of genes discriminating endophytes from their closely-related saprobes. Six CAZymes, CE12, CBM67, GH39, CBM32, CBM38, GH1, were consistently identified as key

genes regardless of phylogenetic correction method used. These genes are likely to be the core genes that enable the endophytic lifestyle, and further investigations in other plant systems is required to verify their universality.

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