Title: Introductions of plant growth promoting bacterial strains differentially modify growth and health of *Populus* biofeedstocks

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Project Goals: The Secure Ecosystem Engineering and Design (SEED) Science Focus Area (SFA), led by Oak Ridge National Laboratory, combines unique resources and expertise in the biochemistry, genetics, and ecology of plant-microbe interactions with new approaches for analysis and manipulation of complex biological systems. The long-term objective is to develop a foundational understanding of how non-native microorganisms establish, spread, and impact ecosystems critical to U.S. Department of Energy missions. This knowledge will guide biosystems design for ecosystem engineering while providing the baseline understanding needed for risk assessment and decision-making.

Abstract text:

Biofeedstock plants, such as poplar trees (*Populus sp.*), grow in unison with soil microbes that can aid plant productivity. Commercially available biostimulants and biopesticides containing bacterial species are marketed for enhancing plant growth and pest resistance. However, the establishment and spread of beneficial bacteria in biofeedstock plants may be constrained by a variety of factors including microbe and host plant gene-gene interactions, characteristics of the invading organisms or resident community, and the environment. We are investigating how non-model bacteria drive their establishment, persistence, and the productivity of biofeedstock perennial plants.

Bacillus species are ubiquitous bacteria in soils and many field trials have demonstrated their ability to stimulate plant growth and suppress plant pathogens. Here, we conducted a greenhouse experiment to examine how single or mixed *Bacillus* strain introductions establish and differentially affect several *Populus* genotypes. *Bacillus* strains were introduced to soils planted with either *Populus deltoides* (genotype 11347 or 16842), a F1 hybrid cross between *P. deltoides* and *P. trichocarpa* (7300 or 8360D), or a no-plant control. We introduced five strains of three *Bacillus* species: *B. velezensis* (GB03 or FZB42), *B. subtilis* (3610 or RO-NN-1), *B. amyloliquefaciens* (DSM7) or a mixture of all five genotypes. Plant growth parameters – photosynthetic rate, number of leaves, stem height, SPAD greenness index, and above and belowground biomass – were measured to quantify the effect of microbial invaders on plants. Overall, we found the plant response to *Bacillus* introduction could not be generalized, and indeed varied across the different plant genotypes.

Propagule pressure is one of the most common explanatory factors that impacts success in micro- and macro-organism invasion. Therefore, we interrogated the introduction of *B. veleznesis* at four levels of propagule pressure: 0 (control group), 50% of the commercially recommended rate (3.4 x 10^9 CFU/gallon), the recommended rate (6.9 x 10^9 CFU/gallon), and double the recommended rate (13.8 x 10^9 CFU/gallon) in a field experiment. We hypothesized that (1) *P. trichocarpa* will support *B. velezensis* introductions; and (2) the establishment and rate of systemic spread will increase with increased propagule pressure. Our on-going field study is a fully factorial cross of 4 tree genotypes x 4 propagule pressure levels x 2 inoculation timing levels x 3 destructive harvest time points (n = 5, 480 plants total). We have completed two destructive harvests and one non-destructive harvest to-date. Sample processing and analyses are in progress. Preliminary findings suggest that *B. velezensis* GB03 inoculations did not alter microbial community composition in bulk soil two weeks post-planting when plants were inoculated at the time of cutting propagation. Harvests will continue non-destructively to monitor persistence of *Bacillus velezensis* GB03 and legacy effects in the microbial community over time. This is the first temporal study of biofungicide systemic spread in a biofeedstock plantation and has applications for national energy security.

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