Strain-Specific Transportomic Capacity of *Pseudomonas fluorescens* Linked to Plant Growth Promotion in Aspen Seedlings under Nutrient Stress

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Project Goals: The goal of the Environmental Sensing and Response SFA is to understand the molecular mechanisms associated with plant-microbe interaction, particularly those interactions that lead to Plant Growth Promotion (PGP) effects in marginal soils. Using a plant-bacteria co-culture, vertical plant laboratory system, we have a collection of Pseudomonad bacteria observed to possess strain- and growth condition-specific PGP effects. By linking laboratory observations with computational modeling, we identify specific molecular mechanisms by with PGP protect plant from abiotic stresses. These molecular mechanisms will provide rich opportunities for rational design of rhizosphere communities to optimize plant biomass grown in marginal lands.

Forest ecosystems are major components of the biosphere and contribute extensive ecosystem services. Trees form a significant storage sink in the global carbon (C) cycle, facilitate water fluxes in the hydrologic cycle, and provide wood and fiber for human consumption. The provision of these benefits depends upon the supply and utilization of resources (carbon dioxide, water, nutrients, and light) to and by the tree. However, photosynthesis and primary productivity are often limited by nutrient availability, which, in turn, affects the ecological roles and economic output of forests. The plant rhizosphere hosts a large and diverse community of microbes whose interactions with roots and soils influence ecosystem productivity. Diverse communities of bacteria colonize plant roots and the rhizosphere. Many of these rhizobacteria are symbionts and provide plant growth promotion (PGP) services, protecting the plant from biotic and abiotic stresses and increasing plant productivity by providing access to nutrients that would otherwise be unavailable to roots. In return, these symbiotic bacteria receive photosynthetically-derived carbon (C), in the form of sugars and organic acids, from plant root exudates. PGP activities have been characterized for a variety of forest tree species and are important in C cycling and sequestration in terrestrial ecosystems. The molecular mechanisms of these PGP activities, however, are less well known. In a previous analysis of Pseudomonas genomes, we found that the bacterial transportome, the aggregate activity of a bacteria's transmembrane transporters, was most predictive for the ecological niche of Pseudomonads in the rhizosphere.

Here, we used *Populus tremuloides* Michx. (trembling aspen) seedlings inoculated with one of three *Pseudomonas fluorescens* strains (Pf0-1, SBW25, and WH6) and one *Pseudomonas protegens* (Pf-5) as a laboratory model to further investigate the relationships between the predicted transportomic capacity of a bacterial strain and its PGP effects. Conditions of low nitrogen (N) or low phosphorus (P) availability and the corresponding replete media conditions were investigated. We measured phenotypic and biochemical parameters of *P. tremuloides* seedlings and correlated *P. fluorescens* strain-specific transportomic capacities with *P. tremuloides* seedling phenotype to predict the strain and nutrient environment-specific transporter functions that lead to PGP activities and the capacity to protect plants against nutrient stress. These predicted transportomic functions fall in three groups: (i) transport of compounds that modulate aspen seedling root architecture, (ii) transport of compounds that help to mobilize

nutrients for aspen roots, and (iii) transporters that enable bacterial acquisition of C sources from seedling root exudates. These predictions point to specific molecular mechanisms of PGP activities that can be directly tested through future, hypothesis-driven biological experiments. For example, the correlation network (Figure) links rootlet formation, an amenable phenotype, with arabinose transport. The characterization of SBW25 mutants deficient in arabinose transport (see P. Korajczyk poster) will allow us to test specifically the predicted interaction with rootlet formation.



Phenotype and *Pseudomonas* **transportome correlation network**. This is a graphical representation of the correlation network for aspen phenotypes and *Pseudomonas* transportome. Green rounded rectangles are aspen seedling phenotypes and diamonds are predicted ligands transported by *Pseudomonas* strains. Ligands are colored according to the specific *Pseudomonas* strain with the greatest (by PRTT-score) relative capacity for that ligand's transport: orange for Pf0-1, yellow for Pf-5, red for SBW25, and purple for WH6. Wavy green lines are strong correlations between aspen seedling phenotypic features. Solid straight lines are strong correlations between *Pseudomonas* transportomic capacity and aspen seedling phenotypes colored by culture condition: grey for replete media, blue for low N media, and yellow for low P media

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