Characterization of microbial community structure in pine bark substrates©

J. Altland[a], F.C. Michel, Jr. and S. Valles

USDA Agriculture Research Service, Wooster, Ohio 44691, USA.

A large body of research has addressed the biological community in soilless substrates. Most of this research pertains to specific sets of pathogens or plant growth promoting microbes. Very little is known about the overall microbial community in terms of species range, diversity and relative population density. The objectives of this research were to analyze microbial community structure in a typical pine bark substrate used for nursery crop production and determine the impacts of compost amendment and plant growth on these communities. Three substrates (v/v); 80:20:0, 80:10:10, and 80:0:20, pine bark:sphagnum peat:leaf compost were prepared. The substrates were filled into 20 L nursery containers and half were planted with a single birch liner (*Betula nigra* 'Cully', Heritage® river birch) from a 50-cell flat while the other half remained fallow. Containers were fertilized with 73 g controlled release fertilizer. There were six single-pot replications per treatment. The microbial consortia in the potting media were characterized using high-throughput ribosomal RNA gene and intergenic spacer region sequencing. Representative samples (500 g) were taken from each container starting on April 12 and monthly thereafter throughout the growing season (4 months) and stored at -22°C until analyzed. DNA was extracted and purified using DNeasy PowerSoil Kit components. The product size was verified by gel eletrophoresis. A 25 µL aliquot at a 5 mg mL^{-1} concentration was used for PCR amplification. Universal as well as population-specific bacterial and fungal primers were used to identify and quantify tens of thousands of individual ribotypes within each sample by comparison of the amplified sequences to 16S gene and ITS databases. The data was processed using an open-source bioinformatics pipeline (QIIME). Bacterial communities of the substrates immediately after potting differed in composition. The compost amended substrate (80:0:20) was dominated by proteobacteria (37.4%), actinobacteria (35.6%) and acidobacteria (23.0%). The peat amended substrate was initially dominated by proteobacteria but also had relatively large percentages of chloroflexi and bacteroidetes. Over time, bacteroidetes increased while actinomycetes and acidobacteria decreased in all of the mixes. While there were initially differences in microbial communities between the substrate types, after 2 months the communities in all substrates were similar. Planting trees or adding compost to the media did not have a strong impact on bacterial community composition after 2 months.

[a]E-mail: James.altland@ars.usda.gov