Bacterial and Fungal Microbiome Analysis of Alfalfa Rhizosphere Soils

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Soil microbial communities are increasingly being recognized for their critical roles in agriculture. While microbiome studies enabled by next generation sequencing platforms reveal soils to be some of the most diverse environments known, certain taxa may have disproportionate influence in their functional roles affecting plant health, productivity, access to nutrients, or related ecosystem services. Organisms that are found to co-associate across multiple soil environments are of interest for their putative interactive roles that may be generalized for different sites or management practices. We examined the composition of bacterial and fungal communities associated with the rhizosphere soils of alfalfa from three different regions in Minnesota with distinct soil characteristics. Three alfalfa germplasms exhibiting different root morphological characteristics underwent fertilization treatments with phosphorus and potassium. The microbial communities inhabiting root-associated soils were determined by sequencing the 16S rRNA genes and ITS1 loci of bacteria and fungi, respectively, from replicated DNA extractions of each experimental treatment. The Illumina MiSeq sequencing platform generated means of over 450,000 16S and 340,000 ITS sequences per sample. Overall, bacterial community composition was represented by a large proportion of Alphaproteobacteria (23% of total bacterial taxa), including *Sinorhizobium* spp., which are known to fix nitrogen in alfalfa root nodules. Other highly represented taxa at all sites included the Actinobacteria (21% of total), Acidobacteria (9%), and Gammaproteobacteria (9%). Given that some taxa are present in multiple sites in this study, understanding their distribution in additional locations would contribute to elucidating the composition of a broader 'core' alfalfa rhizosphere microbiome. Differences in microbial communities were observed between sites, suggesting soil properties may be the primary factor impacting community composition. Co-occurrence association networks were constructed to determine which microbial community members most significantly occurred together in multiple samples, which supports the development of future hypotheses to explore whether these groups of organisms interact near plant roots. This work identifies subsets of these communities which may be prioritized in future screens for potential roles in nutrient cycling and in facilitating plant-microbe interactions.