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New Applications of Proteomics Informs Microbial Functions in Soils

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Within terrestrial systems, microorganisms play fundamental roles as drivers of terrestrial carbon fluxes via either the release of carbon to the atmosphere or sequestration of the carbon in biologically inactive forms. These microbes do not live in isolation, and as such understanding the function of the microbe with in the context of the community is critical to the characterization of the community as a whole. While the genome sequence describes the potential functions resident in the microbial community, it is the protein expression profiles reveal the actual functional potential of any biological system, especially those in soils. However, the proteomic characterization of soil microbial communities can be challenging for a number of reasons. First, soil chemical heterogeneity can potentially interfere with protein extraction thus biasing results toward those microbes that are either not well attached or easily lysed and toward those proteins that do not have strong interactions with the soil matrix. Second, proteomic characterization requires the use of a genome sequence. However, many times a genome sequence of the exact community is not available and a surrogate must be used. If a genome sequence is available, the microbial strain variation leads to a consensus genome sequence and may not exactly correspond to the peptide sequences. Additionally, sequence data that is too comprehensive may be difficult to interpret. Last, the determination nutrient flux through soil systems usually requires the use of Stable isotope Labeling strategies which have not been well formulated for soil systems.

This presentation will discuss methods and approaches for addressing these challenges in the proteomic characterization of soil microbial communities. The variety of protein extraction techniques from soil each are optimal for different soil types, and we will discuss a series of these different extraction methods for different types of soils that effectively allow for the characterization the functional potential of the resident microbes in the soil. Identification of proteins in a complex mixture is based on the comparison of mass spectrometry data to a known genome sequence, however, the availability and quality of these sequences if variable for any given microbial community. For soils collected from Hopland CA, we have determined how three metagenomic databases provide complementary information and demonstrate how database selection can influence the quality and quantity of peptide/protein identifications. Finally, stable isotope probing is a method that identifies and targets active components in a complex system. We examined soils that have been grown with plants exposed to 13-C labeled CO_2 and using advanced protein SIP technology determined the overall proteomic profile of the microbes as well as tracked the label into those proteins that were synthesized in response to the carbon input. Along with the establishment of these methods in soils, we are poised to provide fundamental knowledge on the structure and function of the community with in these soils.