38. Cross-system analysis of carbon assimilation dynamics in soil microbial communities: Documenting the function of non-cultivated microorganisms in terrestrial ecosystems

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Project Goals: This research program will reveal fundamental aspects of soil C-cycling and provide ecological and metabolic insights on diverse non-cultivated soil microorganisms that play major roles in the global C-cycle. Specific goals include: 1) Map the C assimilation dynamics for thousands of non-cultivated microorganisms in soil by harnessing a full cycle microbial food web mapping approach that employs an array of ^{13}C- labeled molecules; 2) Map the C assimilation dynamics of soil microorganisms across soil systems as a function of soil C content and pH, master variables that impact soil community structure and function; and 3) Evaluate ecological and seasonal patterns of activity and abundance for discrete microbial taxa across gradients of soil C content and pH and as a function of their C-assimilation dynamics. These goals will be achieved by employing a newly developed microbial food web mapping approach, enabled by advances in ^{13}C-stable isotope probing of nucleic acids and next generation sequencing.

Global changes in atmospheric CO_2, temperature, precipitation, and ecosystem N inputs, are expected to impact primary production and carbon inputs to soils, but it remains difficult to predict the response of soil processes to anthropogenic change. Models that predict soil C- cycling as a function of ecosystem properties do not explain well variation in soil processes. Our difficulty in predicting the response of soil processes to environmental change suggests a need for a greater understanding of the biotic mechanisms that govern the soil C-cycle. Changes in microbial community structure and function have been proposed to impact soil C-cycling both qualitatively and quantitatively. However, our ability to predict the impacts of these changes on terrestrial ecosystems is constrained by our limited understanding of mechanisms that drive microbial processes in soil systems.

A fundamental limitation in understanding microbial C-cycling in soils is that we have a glaring lack of information about which microorganisms actually mediate critical soil processes and whether their activities vary across ecosystems. This research project will address this fundamental problem by using a suite of experiments to map C-assimilation dynamics for thousands of discrete microbial taxa across dimensions of both seasonal and edaphic variation.

First, we plan to deploy a full cycle food web mapping experiment in contrasting soils to simultaneously track the assimilation of eleven ^{13}C-labeled compounds into soil microorganisms. These compounds have been selected to represent dominant components of plant biomass (cellulose, hemicellulose, lignin) and intermediate products of plant biomass decomposition (glucose, xylose, benzoic acid, glycerol, palmitate, amino acids, lactate, and oxalate). By tracking the assimilation of these isotopically labeled compounds in parallel and in relation to appropriate controls it will be possible to map routes of plant biomass C metabolism by soil communities and identify the specific contribution of thousands of microorganisms to this process. This approach will be employed in contrasting ecosystems including a cultivated agricultural site and a primary forest site. These sites are located in the same geographic region with soils of the same soil order but have very different soil C content representative of their ecosystem types. This effort will reveal the contribution of thousands of uncultivated microorganisms in the soil C-cycle. We will identify the metabolic

capabilities of non-cultivated microorganisms in soil and explore how their activity varies across contrasting ecosystems. Furthermore, we will explore differences in how microbial communities process plant biomass C across these ecosystem types.

Second, we plan to deploy a series of streamlined food web mapping experiments to track cellulose and xylose (the major product of hemicellulose degradation) assimilation by soil communities as a function of soil C content and pH. While both soil C and pH are major drivers of soil microbial community structure it remains unknown whether and how these factors alter soil microbial C-cycling. We will evaluate how these factors impact the C-assimilation dynamics of soil communities across a series of sites. Sites will include conventionally managed cultivated fields, fields managed organically for 10 or 20 years, non-cultivated pasture, and secondary and primary forest sites. These 6 sites represent a gradient of C content and represent the range of soil C commonly found in much of the Northeastern USA. In addition, the impact of pH will be assessed in each of three sites (acidic, slightly acid, neutral to alkaline soils) representing two types of systems: meadow and primary forest. We will determine how these critical soil parameters alter the route that C takes through the soil community and we will identify microorganisms whose activity varies with respect to soil C status, pH, and ecosystem type.

Finally, we will characterize the seasonal dynamics of soil microbial communities for replicate plots at all field sites. SSU rRNA (RNA) and SSU rRNA gene (DNA) sequencing will be used to assess changes in community composition over time and across sites. Food web mapping experiments from objectives 1 and 2 will yield information on C metabolism for discrete taxa across different soils and ecosystem types, but this approach is not amenable to the sampling density required to analyze microbial communities over time. Hence, C assimilation dynamics of discrete taxa identified in objectives 1 and 2 will be evaluated as a function their SSU rRNA and SSU rRNA gene relative abundance over space and time for all field sites. This combined approach will make it possible to build and test hypotheses relating C-assimilation by discrete microbial taxa to their ecological distribution in space and time. We will test species-time relationships for microbial taxa as a function of correspondence in their C-assimilation profiles.

The genetic capacity of microbial communities can be studied through ‘omic approaches but it remains difficult to make direct links between the genetic capacity of microorganisms and their function in the soil C-cycle. Microbial Food Web Mapping through next generation sequencing enabled stable isotope probing (NGS-SIP) makes it possible to link gene sequences to soil C- cycle processes as they occur in soil. This approach allows us to characterize the activity of non- cultivated microorganisms in a range of terrestrial systems. This data will be used to build a base of information about the role of non-cultivated organisms in critical C-cycle processes in terrestrial ecosystems and will provide insight on the manner in which soil communities metabolize soil organic matter. This research will yield fundamentally new insights into the role of soil communities as a driver of soil C-cycling in soils, revealing information about how individual microbial taxa contribute to variation in community function.