

Title: Phylogenetic signal in soil fungal and bacterial communities in response to experimental nitrogen addition

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Project Goals: Anthropogenic N deposition is a chronic and increasing condition in temperate regions that may strongly influence C cycling dynamics. Large increases in N addition have been seen in eastern forests, which have the potential to act as either a source of C or as a major terrestrial C sink. One major theme of our Science Focus Area is to determine the influence of chronic N deposition on microbial C cycling processes in temperate regions, particularly mesic forests. In temperate forest ecosystems, fungal and bacterial biomass is concentrated in shallow surface soil strata where C and N cycling are major processes. The goals of this project are to correlate the resident fungal and bacterial communities, enzyme activities, and local geochemistry across strongly stratified forest soils, determine the impacts of chronic N amendment on soil bacterial and fungal communities across soil layers within a pine forest, and use phylogenetic methods to quantify the effect of N on patterns of soil microbial community assembly. Achieving these goals will provide an understanding of the responsive members of the microbial community within forest soils, and provide insight into whether community shifts and responses vary with environmental conditions, as well as how those changes may impact C cycling. This poster focuses on our recent results from a long-term N deposition experiment conducted in a temperate pine forest in North Carolina.

We used high throughput sequencing methods to target the soil bacterial and fungal communities, which together are major players in N and C cycles, and are applying multivariate statistical approaches to examine shifts in the phylogenetic structure of these communities, as well as the phylogenetic signal in N response among community members. To conduct this study, we designed and validated a new fungal PCR primer (LR22R) to target an approximately 300–400 bp region of the D2 hypervariable region of the fungal LSU for use with the Illumina MiSeq platform. Both *in silico* and empirical analyses showed that the LR22R–LR3 pair captured a broad range of fungal taxonomic groups with a small fraction of non-fungal groups. Based on analysis of known and environmental communities, the new primer had broad utility for constructing accurate phylogenetic trees for fungi, which allowed us to utilize phylogenetic metrics to quantify shifts in both the bacterial and fungal communities. Using highly replicated rDNA surveys we found strong phylogenetic signal in the taxonomic response to N deposition for both the bacterial and fungal communities, and the response was stronger for the bacterial community. Strong shifts in bacterial communities, with relatively minor response in the fungal community suggest that nutrient cycling will be increasingly fungal-dominated in response to N additions. By combining sequence-based community analyses with soil chemistry and enzyme activity measures, we aim to identify key responsive community members with relevance as indicators of community change and with utility for modeling soil processes.

References

Mueller RC, LV Gallegos-Graves, CR Kuske (2015) A new *fungus large subunit ribosomal RNA primer for high-throughput sequencing surveys*. FEMS Microbiology Ecology
<http://dx.doi.org/10.1093/femsec/fiv153>

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