**Anthropogenic N deposition slows decay by favoring bacterial metabolism: Insights from metagenomic analyses**

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**Project Goals:** With support from DoE BER, we have been able to study the molecular and microbial mechanisms by which experimental N deposition has decreased plant litter decay and increased the accumulation of organic matter in a northern hardwood forest ecosystem located in Michigan, USA. Experimental plots have been exposed of increased NO₃ deposition for 20 years at a rate expected by 2050 across some portions of eastern North America. Using biogeochemical and molecular analyses, we have been able to test our hypotheses that the ecosystem response to experimental N deposition (i.e., greater soil C storage) is governed by the environmental regulation of microbial gene expression. Here, we present findings that suggest saprotrophic bacteria, which degrade organic matter less efficiently than their fungal counterparts, may be favored by future rates of N deposition.

Litter decomposition is an enzymatically-complex process that is mediated by a diverse assemblage of saprophytic microorganisms and can be suppressed by anthropogenic N deposition. In a northern hardwood forest ecosystem located in Michigan, USA, 20 years of experimentally increased atmospheric N deposition has reduced forest floor decay and increased soil C storage. Here, we paired extracellular enzyme assays with shotgun metagenomics to assess if chronic N deposition has altered the functional capacity of microbial communities inhabiting decaying forest floor. Experimental N deposition significantly reduced the activity of extracellular enzymes mediating plant cell wall decay, which occurred concurrently with changes in the relative abundance of metagenomic functional gene pathways. Experimental N deposition altered the relative abundance of 60 functional gene pathways mediating the metabolism of carbohydrates, aromatic compounds, as well as respiration. Moreover, experimental N deposition increased the relative abundance of 50 of the 60 gene pathways, the majority of which were associated with saprotrophic bacteria. Conversely, the relative abundance and composition of fungal genes mediating the metabolism of plant litter was not affected by chronic N deposition. Future rates of atmospheric N deposition have favored saprotrophic soil bacteria, whereas the metabolic potential of saprotrophic fungi appears resilient to this agent of environmental change.

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