In order to detect possible differences in N fertilization response between mycorrhizal types, we established a plot network of 6 AM and 6 ECM dominated (>65% diameter at breast height; Table S1) 10 x 10 m plots in the lower elevation hardwood zone of both the reference and fertilized watershed in May 2016 (N=24 plots). Tree species were similar between watersheds with AM trees represented by ﻿red maple (*Acer rubrum*) and sugar maple (*Acer saccharum*) and ECM trees represented by American beech (*Fagus grandifolia*), grey birch (*Betula populifolia*), and yellow birch (*Betula* *alleghaniensis*).

To capture variability across the growing season, we sampled soils in each plot in May, July, and September of 2016. In each plot we extracted three 10 x 10 cm organic horizon layers and homogenized them into a single sample defining this as the OH soil fraction. Next, we sampled four 5 cm diameter mineral soil cores to a depth of 15 cm beneath the OH layer and homogenized these by plot. All samples were kept on ice and transported to West Virginia University for further processing within 48-72 h. Upon return to the lab, we separated rhizosphere soil from mineral soil samples via the soil-adhesion method wherein the rhizosphere soil fraction was operationally defined as soil that remained clung to roots after modest shaking (Phillips & Fahey, 2005). After removal of roots, all soils were passed through a 2 mm sieve and stored at -80°C until further analysis.

To determine the extent to which N fertilization impacts microbial allocation to extracellular enzymes, we assayed the potential activity of hydrolytic enzymes that release N (﻿N-acetylglucosaminidase; NAG), phosphorus (acid phosphatase; AP), and simple carbon (ß-glucosidase; BG). In addition, we measured microbial allocation to complex C degrading oxidative enzymes phenol oxidase and peroxidase. Briefly, 1g of thawed soil was homogenized in 50mM sodium acetate buffer (pH 5.0). Next, hydrolytic activities were determined ﻿using a fluorometric microplate assay (Gemini XPS, Molecular Devices, USA). with methylumbelliferone-linked substrates and oxidative enzymes using a colorimetric microplate assay (Tecan Infinite 200 Pro, Switzerland) with L-3,4-dihydroxyphenylalanine linked substrates according to Saiya-Cork *et al.*, 2002.

Saiya-Cork KR, Sinsabaugh RL, Zak DR (2002) The effects of long term nitrogen deposition on extracellular enzyme activity in an Acer saccharum forest soil. *Soil Biology and Biochemistry*, 34, 1309–1315. DOI: 10.1016/S0038-0717(02)00074-3.

See protocol: https://allison.bio.uci.edu/protocols/fluorimetricenzymeprotocol.pdf