ESF Summer 2015 Internship Proposal

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*A study on the effect of nutrient manipulations on N mineralization over time*

**Background**

The Multiple Element Limitation in Northern Hardwood Ecosystems (MELNHE) project studies nitrogen (N) and phosphorus (P) acquisition and limitation by manipulating nutrients in northern hardwood forests. This research utilizes the resource optimization hypothesis, which suggests that due to resource competition, species should maintain a balanced allocation pattern for uptake of nutrients (Nutrient co-limitation in young and mature northern hardwood forests, n.d.). If plants adjust their internal metabolic requirements to changes in resource availability, ecosystem productivity will increase. The Multi-Element Limitation (MEL) model borrows ideas from this hypothesis, and predicts that resources are often optimized by ecosystems and are thus co-limiting (Nutrient co-limitation in young and mature northern hardwood forests, n.d.). In order to test this theory, researchers are performing nutrient manipulations by applying a series of amendments, including N, P, N+P, and Ca, to tree stands of varying ages.

Study sites include the Bartlett Experimental Forest, Hubbard Brook Experimental Forest, and Jeffers Brook in the White Mountain National Forest in New Hampshire, which represent a gradient in soil fertility. Jeffers Brook is the most fertile and Bartlett is the least fertile. In each site, researchers analyze stem diameter, leaf area, sap flow, foliar chemistry, leaf litter production and chemistry, foliar nutrient resorption, root biomass and production, mycorrhizal associations, soil respiration, heterotrophic respiration, N and P availability, N mineralization, soil phosphatase activity, soil carbon and nitrogen, nutrient uptake capacity of roots, and mineral weathering. These ongoing experiments will contribute to knowledge regarding hardwood forests.

**Introduction**

The MELNHE project performs nutrient manipulations in order to observe variations in ecosystem productivity. The addition of nutrients has unique effects on tree stands. Every few years, N mineralization is tested at each site during the summer field season after nutrient manipulations have been performed. From these experiments, researchers have observed that N mineralization has increased in the N plots, there has been no response in P plots, and N mineralization has substantially decreased in N+P plots in mineral soil (Fig. 1, 2, 3). It is not yet understood if testing N mineralization at different times throughout the growing season will show similar results. Studies have shown that there is a seasonal pattern to N mineralization, where N min was found to be highest in the middle of the growing season (July) (Ross et al, 2004). Alternatively, Fisk and Fahey found higher N min earlier in the growing season, and a lower fraction of gross N min that as immobilized, which is consistent with higher N availability (Fisk and Fahey, 2001). We aim to test whether or not there is a seasonal response of N mineralization to added N and P.

Nitrogen is commonly thought to be the limiting nutrient in forest ecosystems. When N is limited, productivity and carbon sequestration decrease (Chapman et al, 2013). Factors affecting the loss of available N depend on rates of N transformations and quantities of mobile NO3 (Fisk et al, 2002). In addition, forest stand age can impact N availability. Specifically, mature forests show decreased N leaching as a result of the immobilization of N, which can likely be attributed to greater microbial activity (Fisk et al, 2002). Similarly, forests that have been clearfelled are likely to experience N leaching as a result of higher soil temperatures and moisture, which are ideal conditions for enhanced soil microbial activity (Fisk and Fahey, 1990). A major component in the N cycle is N mineralization. N mineralization is the process by which organic N is decomposed by microbes in the soil into plant-available ammonium (Johnson et al, 2005). Mineralization is affected by soil temperature and moisture. As the mineralization increases, so does productivity. The addition of nutrients alters this process.

I don’t know the motivation for the study.

In this study, we will measure potential net N mineralization in C7, C8, and C9 in the Bartlett Experimental Forest in Bartlett, New Hampshire throughout the growing season. I will collect 48 samples in each stand pre-fertilization and post-fertilization. We hypothesize that if nutrient manipulations are added then treatments responses will show similar results over time. We expect that treatment responses will be similar to what has been observed in the past, but N mineralization will fluctuate throughout the summer.

**Research Questions**

*How will nutrient manipulations impact N mineralization over time? Do we see the same treatment response throughout the growing season?*

**Hypothesis**

If N mineralization is tested throughout the growing season, then treatments will show similar but fluctuating responses to past results.

* Rationale: In past experiments, soils were sampled 21 days after fertilization and run for N mineralization. N mineralization increased in N plots, showed no response in P plots, and decreased in N+P plots. As soil temperature increases throughout the season, a fluctuation in N mineralization will result.

**Methods**

*Site Description*

 We will collect soil samples in three sites in the Bartlett Experimental Forests. We chose plots C7, C8, and C9 as they are the mature plots in Bartlett. In each of the 3 plots, there are 4 stands. The stands have varying nutrient manipulations: a control with no nutrients added, N, N+P, and P. Each stand has 4 fenced-off substands where we will collect our samples.

*Sample collection*

 In each of the substands, we will collect 3 cores. Cores will be collected by using a split core, which will be hammered into the ground with a mallet and then removed. Split cores are 4 cm in diameter and 30 cm in length. Once cores are collected, we will split each of the horizons and composite all of the horizons in each substand. We will split horizons into Oe, Oa, and mineral soil. We will collect a total of 36 composited cores for all stands before fertilization in early June and again in July and August.

*Identifying horizon layers*

 We will separate our samples into the Oe, Oa, and mineral soil horizons. This will be done either in the field or the lab. We will identify breaks in the soil horizons and use a kitchen knife to separate them and composite similar horizons from the same substand. Oe can be identified by separating the less decomposed, spongy layer off the top. Oa is the highly decomposed, dark, and greasy layer. The mineral layer is easier to identify, and is silty and lighter in color.

*Moisture content for dry mass conversion*

5-10 g of soil will be weighed in a weigh boat and the weight of the boat and boat+soil will be recorded. Weights for each sample will be about 18 g, 25 g, and 40 g for the Oe, Oa, and mineral horizons, respectively. The soil will be dried in an oven at 60’C to constant mass (~4-5 days). After this time, samples will be reweighed and the weight of the soil+boat will be recorded. The dry weight conversion factor is:

((dry+boat)-boat)/((fresh+boat)-boat)

The fresh soil mass will then be multiplied by the dry weight conversion to get the dry weight.

*Potential net N mineralization*

* Time 0 (t=0) extractions
* Varying masses of fresh soil will be placed into a tared centrifuge tube and the masses will be recorded. Weights for each sample will be about 18 g, 25 g, and 40 g for the Oe, Oa, and mineral horizons, respectively. 80 mL of 2M KCl will be added to the tube. Tubes will be shaken by hand for 60 seconds. Tubes will be allowed to settle for 12-24 hours after shaking. The solution will be filtered through a Whatman #1 paper. Extracts will be stored in the fridge and sent to Miami University within 2 weeks of extraction to analyze for NH4 and NO3.
* Incubations and t=n (final) extractions
* Fresh soil will be added to a tared Ball jar and weighed. Weights for each sample will be about 18 g, 25 g, and 40 g for the Oe, Oa, and mineral horizons, respectively . The mass will be recorded. Jars will be capped tightly so soil does not dry. Every few days during the incubation period, fresh air will be let into the jars. After the incubation period, samples will be extracted using the same procedure used above.
* N analysis and N mineralization calculation
* KCl extracts will be analyzed for NH4 and NO3 in Miami. Samples will be transformed from ug N/mL into ug N/total soil extracted by multiplying by the number of mL KCl used to extract soil. The result will be expressed as ug N/g soil extracted. The N mineralization per day is: [(NH4i + NO3i)-(NH4f + NO3f)] / number of days incubated, where NH4 and NO3 are ug N/g soil.

**Predictions**

* N mineralization will fluctuate throughout the growing season, but there will be similar responses to treatments in each site (Fig. 1, 2, and 3).

Fig. 1 shows the compiled data for net N mineralization in the Oe horizons. These results are expected.

Fig. 2 shows the compiled data for net N mineralization in the Oa horizons. Similar results are expected for this project.

Fig. 3 shows the compiled data for net N mineralization in the mineral horizons. Similar results are expected for this project.

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