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**Microbial Biomass in North Eastern Hardwood Forests**

**Introduction**

Microbial biomass is an important indicator of the health of the soil, the soil organic matter, and the nutrients available in the soil. Microbes are crucial for breaking down organic plant matter through the release of extracellular enzymes, which in turn acts as a fuel to produce more of these decomposing enzymes, such as phosphatase, and to produce greater biomass. It has been shown that microbial biomass is limited by the resources taken up by the microbes and the ratio between the major nutrients of carbon, nitrogen, and phosphorus. (Mooshammer 2014). This idea of co-limitation supports the theory that organisms avoid limitation by individual resources by using plentiful resources to obtain resources that are more sparse. (Ratliff 2012). The microbes will uptake available substrates and regulate the efficiency they use the nutrients by keeping the balance of these substrates by releasing ones in excess and keeping ones in more limited supply. This homeostasis in the microbes causes the limiting resource to be stored in the microbes and added to the microbial biomass. (Mooshammer 2014). The Growth Rate Hypothesis is one hypothesis that has been made on how microbes maintain their stoichiometric homeostatis. It shows a relationship between the phosphorus concentrations and the cellular growth rate where growing cells need phosphorus to produce protein.This could suggest a sensitivity for microbial metabolism to available phosphorus, so the microbial biomass is limited by phosphurus in the microbial metabolism. Likewise, nitrogen rich proteins are used in microbes for structure and therefore the microbial biomass is limited in growth by nitrogen. (Hartman. 2013).

The Multiple Element Limitation in Northern Hardwood Ecosystems (MELNHE) is a project designed to test the limiting factors in ecosystems in the north eastern hardwood forests. Since the microbes recycle nutrients, they provide organic nutrients for plant uptake; therefore, the limitation of microbial biomass results in limitation of nutrients put back into the soil from the microbes. Thus, this causes a starved soil for nutrients for plant uptake, decreasing the overall health of the forest. Previous studies have been done on the microbial biomass in this site, but since it was done shortly after the nutrient additions began, it was too soon for changes to be seen and no conclusion could be made on the limiting resources to the microbial biomass. The effects of an unhealthy forest ecosystem could affect areas of human life such as the economy in the logging industry. With an understanding of limitations in the microbial biomass of a forest ecosystem, these limitations can be corrected to optimize the microbial biomass and boost the recycling of nutrients in the soil. This could also increase the aboveground biomass, benefiting the industries reliant on aboveground forests..

**Question**

Is the microbial biomass in the soil of the northern hardwood forests limited by nitrogen, phosphorus, or co-limited by both nutrients?

**Hypothesis**

The addition of limiting nutrients into an ecosystem will result in an increase in the microbial biomass.

**Methods**

Field: The MELNHE project contains 13 experiment stand, each with four 50m x 50m plots, a control plot, and three nutrient treated plots fertilized with nitrogen, phosphorus, and nitrogen and phosphorus. Within each plot there are four subplots in which three soil cores will be taken from. The cores from the subplots will be divided by horizon (Oa, Oe, and B). The divided cores will be homogenized with the other samples of the same horizon from the same overall plot to give three samples per plot, one sample for each horizon.

Lab: To measure the microbial nitrogen and phosphorus, the *Brookes et al.* method of fumigating the sample with chloroform will be used. For microbial N, two identically weighed subsamples will be taken from the samples per plot. One subsample will be used immediately for the time zero. 0.5M K2SO4 extract will be added to the soil and then shaken vigorously and filtered. The filtrate will be sent for analysis for N. The other sample will be fumigated in chloroform for three days and then extracted in the same method as the unfumigated time zero sample. The difference between the fumigated and unfumigated sample gives the microbial N. Two more identically sized subsamples will be used for testing by incubation. The time zero subsample will be immediately extracted with 2M KCl and filtered and analyzed for inorganic forms of N. The other subsample will be incubated for 21 days and then tested in the same method. This test gives the available nitrogen that was in the soil sample. The Microbial biomass P will be calculated in the same method at the fumigation method for microbial biomass N, but the extract used will be 0.5M NaHCO3, and found to be the difference between unfumigated subsample and the fumigated subsample.

**Expected Results**

It is expected that co-limitation will be found within the plots. The N and P treated plots will have the greatest microbial N and microbial P because the increased nitrogen will allow the microbes to grow and take up more phosphorus. If more phosphorus is available for uptake then the microbes will have greater phosphorus levels within them. In the plots solely treated with nitrogen, it is expected to have a slightly higher microbial N and P than plots only treated with phosphorus because the nitrogen will allow more growth and nutrient uptake, but since there was no phosphorus addition to this plot, there will be less phosphorus for the microbes to take in. The plots treated with phosphorus only will not be enhanced in growth because the lack of nitrogen addition and therefore will not have enhanced growth and the ability to take in the extra available phosphorus. Although the phosphorus levels are still expected to be higher than the control plot because there will not be limited phosphorus levels with the treated plot. The control will have the least amount of microbial N and P because the lack of nutrient enhancement.

Work Cited

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