# LONG-TERM WARMING AND NITROGEN DEPOSITION DRIVE CHANGES IN ENZYME ACTIVITY


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## ABSTRACT


Warming and nitrogen deposition change the earth's ecosystems drastically. Carbon (C) and Nitrogen (N) are two prominent regulators of these ecosystems. Warming causes carbon to be released into the atmosphere as carbon dioxide, a potent greenhouse gas. It also results in increased deposition of N, the most limiting nutrient for terrestrial ecosystems, in soils. Microbial extracellular enzyme activity (EEA) controls how carbon and nitrogen flow through the soil ecosystem and recent work has shown that enzymes are sensitive to environmental change. Enzymes catalyze decomposition in the environment and such changes may alter soil carbon cycles. This study is a small-scale simulation of what the earth's climate could be like 50 years from now. All four enzymes we tested varied differently when subject to environmental factors such as warming, nitrogen, and warming and nitrogen combined. Separately, nitrogen deposition and warming caused activity in some enzymes to increase and suppressed activity in others, but together nitrogen and warming had an antagonistic effect. Our data will be used to expand our knowledge on the effects of long-term warming on EEA in soil.


## INTRODUCTION

***Anthropogenic activities disrupt the natural environment:***

- projected warming between 2 °C and 6 °C (5.)
- release greenhouse gases: $CO_2$, $CH_4$, $NO_2$, $H_2O$, and $O_3$
- increase in atmospheric temperature increases soil temperature

***Carbon:***

- gives soils structure
- increases productivity by creating diverse habitat for microbes

***Soil Organic Matter: (7.)***

- Healthy SOM is paramount to healthy plant growth
- Maintained through decomposition

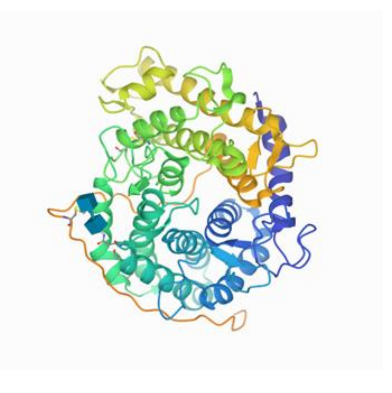
***Microbes decompose Soil Organic Matter***

- soil carbon cycle
- makes nutrients readily available for plant use

***Enzymes***

- catalyze reactions in the soil
- provide energy for microbes

**Figure I: Enzyme**



Castro, J. (2014). *Enzyme* [Clip art]. Live Science. https://www.livescience.com/45145-how-do-enzymes-work.html

## HYPOTHESIS

***Soil microbial enzyme activity decreases as a result of both soil warming and increased nitrogen deposition***

## METHODOLOGY

***Site Description:*** Prospect Hill Tract of the Harvard Forest Long Term Ecological Research Site in Massachusetts, USA

- even-aged, mixed hardwoods

**Figure II: Plot Locations**

***Soil type***: fine, loamy and mixed mesic

***Climate***

- Annual temperature: 7 °C (3.)
- Summer: high of 32 °C
- Winter: low of -25 °C
- Average total precipitation: 1100mm (Boose et al. 2002)

**Figure III: Soil Plot**


***Buried heating cables***

- 10 cm depth below the soil surface
- spaced 20 cm
- Nitrogen added to the plots as liquid ammonium nitrate

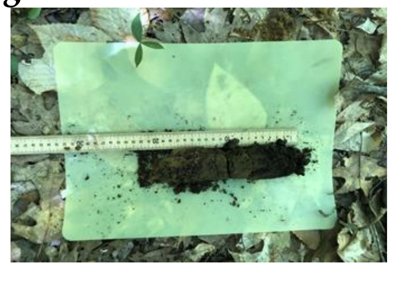
***Factorial treatment structure:*** (3.)

- above ambient soil conditions for 14 years
- +5 °C warming, +50 kg N $ha^{-1}$ $year^{-1}$

**SWaN experiment**: 23 plots

- 6 controls
- 5 heated
- 6 nitrogen addition
- 6 heated with nitrogen

**Figure IV: Soil measurements**



***Collection:***

- organic horizon sampled in a 10 x 10 cm area
- mineral horizon sampled to a depth of 20 cm
- samples were stored at 4°C
- samples sieved:
  - rocks, roots, and other organic debris > 2 mm removed
  - stored at -80 °C until soil enzyme assay was performed

***Enzymes:***

- **Cellobiohydrolase (CBH)**: degrades cellulose
- **β-D- glucosidase (BG)**: Degrades glucose bonds
- **N-acetyl-β-D-glucosidase (NAG)**: degrades chitin
- **Acid Phosphatase (PHOS)**: extracts phosphates from soil proteins

Fluorometric enzyme assay used to measure microbial enzyme activity. These units were then used to calculate final activity in µmols/g*hr.

**Figure V: Soil samples**

## RESULTS

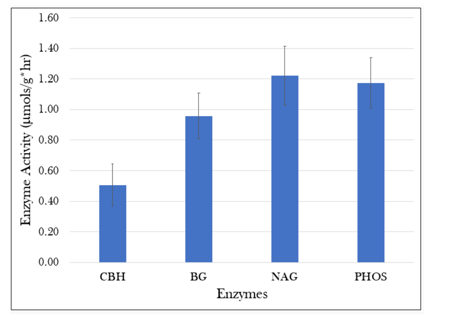
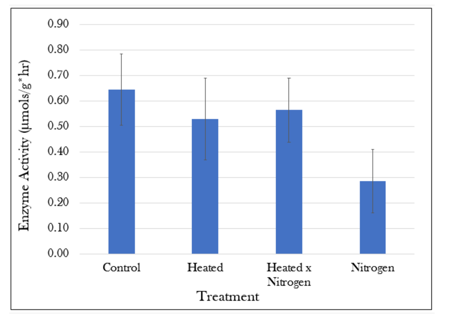


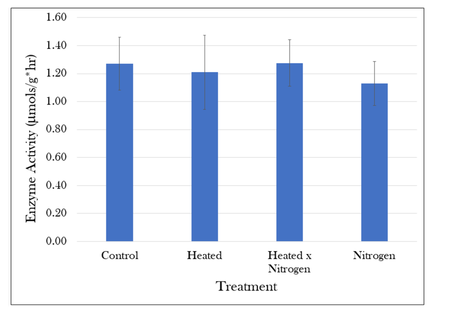

**Figure VI: Average of the activity of all four enzymes in µmols/g*hr**

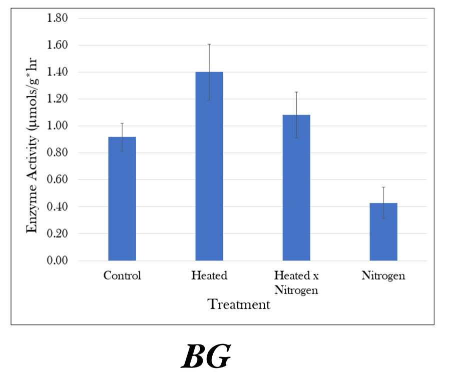
**Figure VII: Enzyme Activity for all four enzymes in µmols/g*hr**

***CBH***
Least activity overall

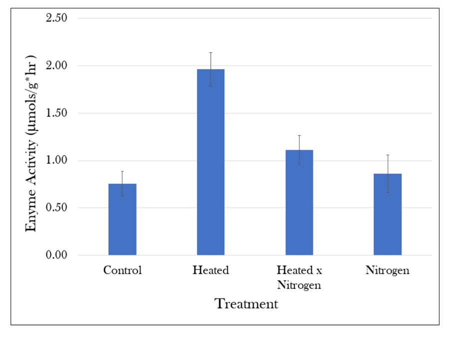



***NAG***
Greatest activity overall with minimal variation




***BG***
Responded most to all three treatments




***PHOS***
Increased with warming and decreased with N deposition

## DISCUSSION

EEA response to treatment varied amongst enzymes

***Each enzyme plays a different role in nutrient cycling in the broader ecosystem***

- soil C, P, and the pH may influence variation in enzyme activity
- NAG degrades some of the most persistent compounds in the environment (2.)
- low PHOS activity with nitrogen deposition
- increase of PHOS with warming
- increase of BG with warming

Root and mycelial presence could also have an effect on enzyme activity

***Mycorrhizal colonization rates in the same soils:***

- decline under warming and nitrogen addition alone (1.)
- significantly increase when soils are warmed and fertilized simultaneously

## CONCLUSION

***Hypothesis***

- Supported:
  - nitrogen deposition seemed to reduce enzyme activity to the largest degree for CBH, NAG, and PHOS
  - NAG and CBH decreased with warming
- Not supported:
  - BG and PHOS increased with warming

The combination of warming and Nitrogen had an antagonistic effect on EEA

***Future Work***

- Assess soil conditions as a factor
- Study environmental science in college and hopefully join a research team.

## ACKNOWLEDGMENTS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