

# The Effects of Extreme Rainfall Events on Saprotrophic Fungal Activity in Michigan Forests

Richard Cheung

University of Michigan Biological Station  
EEB 381 General Ecology  
August 15, 2018  
Dr. Brendan O'Neill

## Abstract

In northern Michigan, a large part of climate change's impact will be the continual increase in the severity of rainfall events. As it stands, much of the rainfall accumulated in Michigan occurs in the 10 days of highest precipitation. This has implications on the terrestrial environment, and transitively the rate of decomposition of carbon. In forests, saprotrophic fungi mediate a portion of decomposition through the use of extracellular enzymes, such as phenol oxidase. Using phenol oxidase activity as a proxy for fungal activity, we tested the effects an extreme rainfall event (1 in/hr) on three different Michigan landscapes with very different soil chemistry and composition. We determined that this large increase in water had an inverse relationship with enzyme activity in both the dune site and the moraine. However, in the wetland, the rainfall simulation did not have much of an effect on the soil moisture and thus did not impact the phenol oxidase activity significantly. Water did not appear to be a limiting factor in any of the sites.

*I grant the Regents of the University of Michigan the non-exclusive right to retain, reproduce, and distribute my paper, titled in electronic formats and at no cost throughout the world.*

*The University of Michigan may make and keep more than one copy of the Paper for purposes of security, backup, preservation and access, and may migrate the Paper to any medium or format for the purpose of preservation and access in the future.*

Signed,

A large, stylized handwritten signature in black ink, appearing to read 'Richard Cheung', is written over the 'Signed,' text.

Richard Cheung

Research Paper

Ecology 381

The Effects of Extreme Rainfall Events on Saprotrophic Fungal Activity in Michigan Forests

**Abstract:**

In northern Michigan, a large part of climate change's impact will be the continual increase in the severity of rainfall events. As it stands, much of the rainfall accumulated in Michigan occurs in the 10 days of highest precipitation. This has implications on the terrestrial environment, and transitively the rate of decomposition of carbon. In forests, saprotrophic fungi mediate a portion of decomposition through the use of extracellular enzymes, such as phenol oxidase. Using phenol oxidase activity as a proxy for fungal activity, we tested the effects an extreme rainfall event (1in/hr) on three different Michigan landscapes with very different soil chemistry and composition. We determined that this large increase in water had an inverse relationship with enzyme activity in both the dune site and the moraine. However, in the wetland, the rainfall simulation did not have much of an effect on the soil moisture and thus did not impact the phenol oxidase activity significantly. Water did not appear to be a limiting factor in any of the sites.

**Introduction:**

Over the course of the next hundred years, global warming will impact the world greatly. Climate change has implications on whole ecosystems and global weather patterns (Thomas et al., 2004). In the U.S. Midwest, the occurrence and severity of rainfall has been increasing since

the late 1930s, and the past 2-3 decades have been the wettest decades on record for the region (Andreson et al., 2012). Intense precipitation events account for much of the rain received in the Midwest; approximately 30% of annual rainfall occurs on the 10 days of highest precipitation (Andreson et al., 2012). Extreme precipitation events will continue to increase in severity and have lasting implications on Michigan's terrestrial environments by effecting processes such as decomposition. Here we focus on how severe rainfall events may effect indicators of decomposition in northern Michigan forests.

The landscapes of Michigan were formed by the movement of glaciers during the last ice age. 21,000 years ago, northern Michigan experienced advances and retreats of the Laurentide ice sheet, resulting in features such as moraines. Moraines and outwash plains are distinguished by how they were formed with moraines formed at the edge of the glacier, while an outwash plain is formed through the deposition of meltwater sediments (Karowe, 2018). These differences result in very different soil types, including topsoil, clay, and sand varieties (MDEQ, 2015). A dune, on the other hand, is formed through saltation, which is a mechanism by which wind moves sand grains (Herrmann, 2007). Unlike moraines, dunes, and outwash plains, a wetland is characterized by the by permanent or periodic inundation in water (Tiner 2017). It is a more generalized term to describe the moisture content of the environment.

These differences in soil type correspond to differences in organic matter content. Certain landscapes are able to accumulate differing amounts of organic matter. Additionally, the ability of the soil to retain moisture has implications on the aboveground plant communities, which leads to distinct litter. Conifer forest litter may have a higher level of phenolic content and lower pH, for instance. Depending on the habitat, decomposition may relate to the total amount of accumulated soil carbon and/or the quality of the carbon entering the soil through plant litter.

The breakdown of organic carbon into inorganic materials is mediated by decomposers. In terrestrial ecosystems, saprotrophic fungi are essential in the breakdown of organic matter for nutrient cycling (Crowther, 2012). Saprotrophs function through the extracellular release of specific enzymes that aid in the breakdown of organic material. The breakdown of lignin is accomplished, in large part, by the activity of enzymes such as phenol oxidase and peroxidase, which work to depolymerize phenolic compounds like lignin (Sinsabaugh, 2010). Active fungi release these enzymes, so more activity may be determined by increased enzyme presence/activity.

With saprotrophic fungi being a vital part of northern Michigan's terrestrial environment, we wanted to understand how global climate change would impact saprotrophic productivity in different Michigan terrestrial landscapes. We predict that in environments limited in water, an extreme precipitation event would increase fungal activity. Additionally, in environments where soil water is at an optimum or excess, an extreme precipitation event would decrease fungal activity by decreasing organic carbon availability. Dunes and outwash plains are composed of sandier soil, which allows water to leach through more easily (Karowe, 2018). Because of this, we predict water to be the limiting factor in fungal productivity, measured through enzyme activity, in the dune site. Therefore, we predict more activity to occur in the dune. The wetland is defined by its saturation, and we do not think that adding water will have that large of an effect on the phenol oxidase activity. In the moraine, we predict that water will not be a limiting factor due to its presence in relatively high amounts in the soil, and therefore the large amount of water will overwhelm and negatively impact saprotrophic productivity.

We hypothesize that phenol oxidase activity will differ between sites (dune, wetland, moraine) based on available carbon and carbon quality. Additionally, we hypothesize that the

effect of an extreme rainfall event would have a distinct effect on these sites, with drier sites having an increased response of enzyme activity after moisture addition.

### **Methods:**

Our contrasting sites included a dune, a moraine, and a wetland near the University of Michigan Biological Station in Pellston, Michigan. The dune was considered to be so due to its glacial history. It was once a dune at the edge of a glacial lake and still maintains some of those characteristics. The wetland is on an outwash plain, but it has built up a large amount of organic material due to its high moisture. For each site, we had a total of three plots including a control. Each plot was created on a flat plane on the soil and spaced approximately 10 meters apart to prevent transfer of water. We used a quadrat to measure the one square meter plots, which were marked by flags and string. On July 27<sup>th</sup>, 2018, we simulated an extreme rainfall event at all three different sites on the two experimental plots.

Using statistics on Michigan annual rainfall, we determined that 1 inch/hour would be considered an extreme rainfall event (“Data Tools: Daily Weather Records”). We used a sprinkler system to deliver deionized water at that rate to the plots for 30 minutes.

We collected soil samples using a soil core 10 cm down from the soil, brushing away the o-layer to expose the soil. We collected the samples before we simulated rainfall, 12 hours, 24 hours, and 72 hours post rainfall event, including on the control plot. Each time we collected samples, we took five soil cores. The samples were collected in Ziploc bags, homogenized, and stored in an icebox. We sieved the soil using a 2 mm sieve and soil was stored in a refrigerator to preserve fungal enzymes. To obtain soil moisture percent, we weighed 20 g of soil, placed in a 60 degree Fahrenheit oven and reweighed it once it was completely devoid of moisture.

Using the soil samples, we utilized analytical chemistry to obtain information on the soil. This included percent carbon using loss on ignition, phenol composition determined using high performance liquid chromatography (HPLC), ergosterol concentration determined through gas chromatography mass spectrometry (GCMS), soil pH, and phenol oxidase activity determined fluorometrically after incubation over time. Differences between the sites were analyzed using ANOVA. The differences in phenol oxidase activity over time was analyzed using repeated measures ANOVA. We set the cutoff for significance at  $p=.05$ . Statistics were run in Excel and R.

### Results:

The mean percent carbon was .679, 7.006, and 1.510 for the dune, wetland, and moraine, respectively (Fig. 1). The wetland control (W3) was removed due to the plot being very different from the others, and thus deemed not suitable as a site replicate. These differences were statistically significant ( $F=86.775$ ,  $p=.0001$ ). The differences between the soil pH's were also statistically significant ( $F=13.098$ ,  $p=.006$ ) (Fig. 2).

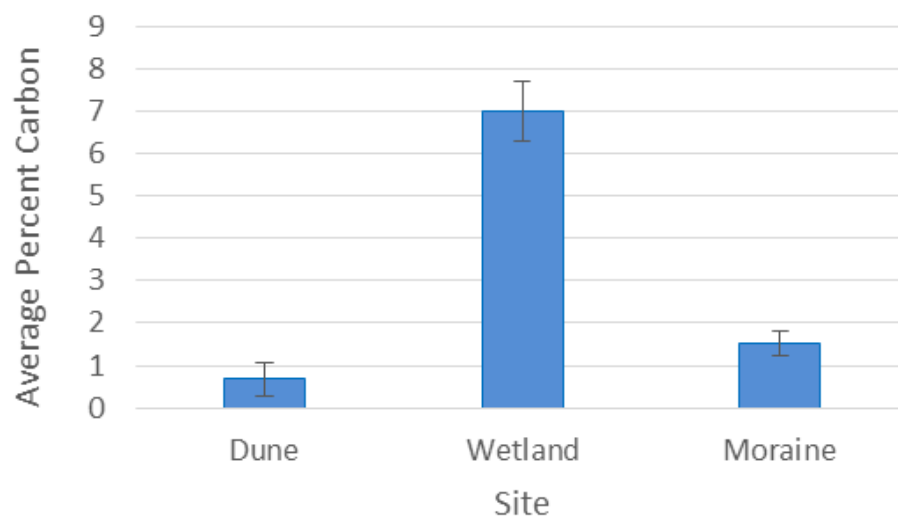


Figure 1. Average carbon content (%) in each site.

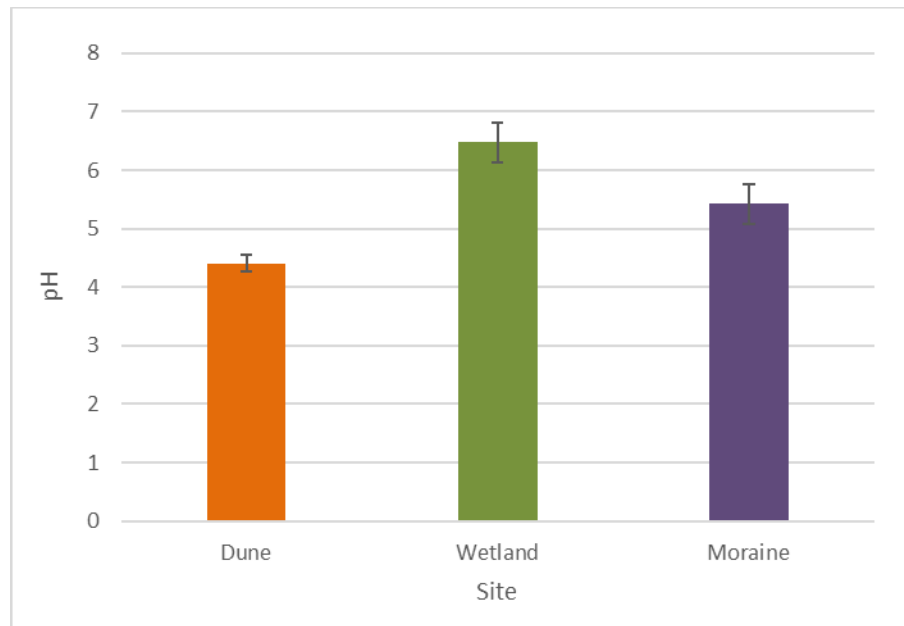


Figure 2. Average pH of the soil at each site.

The average phenolic concentrations (mg/gram of soil) over carbon (%) were .136, .095, and .180 for the dune, wetland, and moraine, respectively (Fig. 3). The wetland control sample (W3) was again excluded from this data due to the carbon percent being much higher than the other plots. These numbers indicate the proportion of phenolic content per total soil carbon. The differences were almost marginally significant ( $F=5.498$ ,  $p=.107$ ). However, the difference between the content in the moraine and wetland were seen to be statistically significant ( $t=4.315$ ,  $p=.0229$ ).

Phenolic concentrations increases with carbon concentration. In the wetland, the phenol concentration is higher, but relative to the total carbon, it is small. In the moraine, this trend is the opposite. The phenol concentration is low, but when compared to the carbon in the moraine, there is a relatively high amount of phenol (Fig. 3, 4).

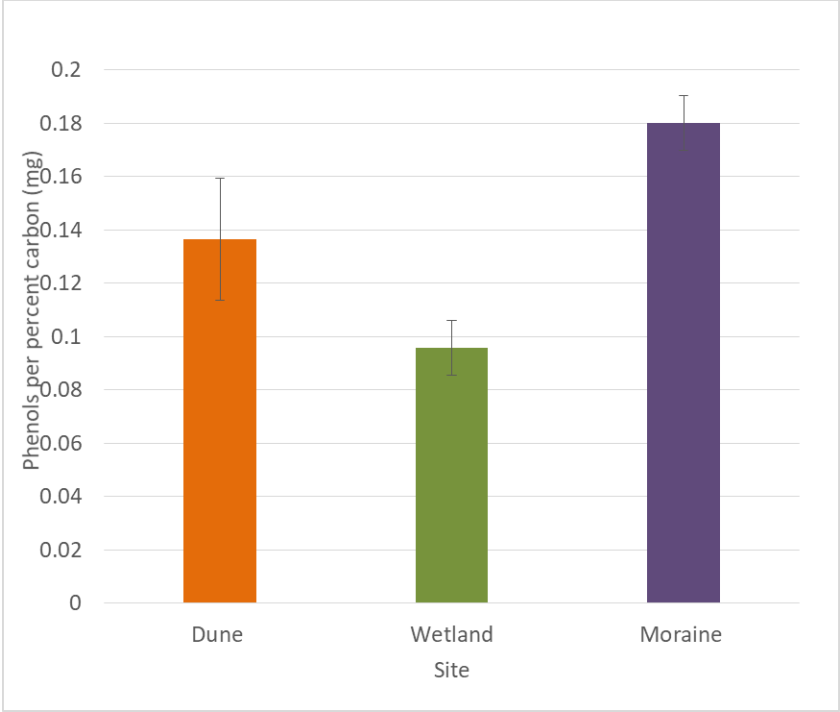


Figure 3. Phenol content of each site, relativized by their carbon content (%).

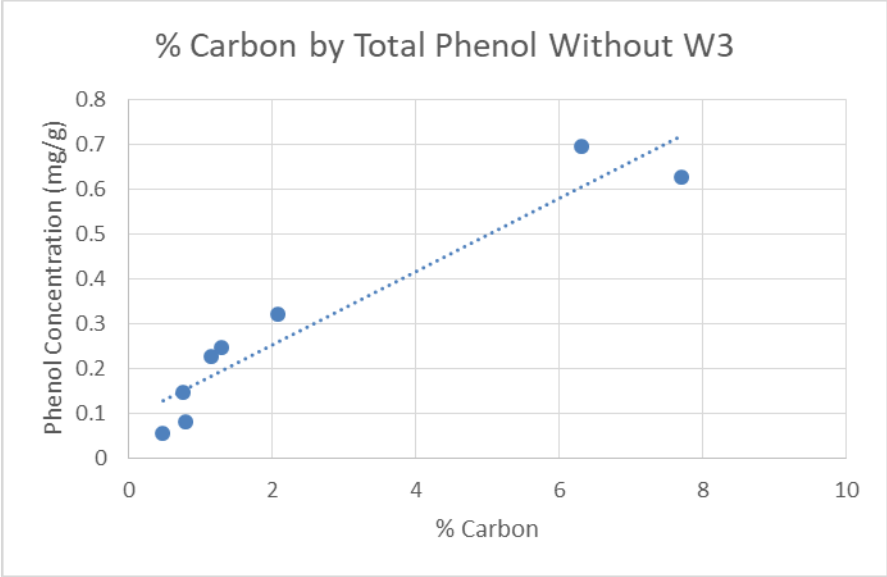


Figure 4. Correlation graph of carbon (%) by total phenol sans W3 data (correlation (r)=.954 without W3, r=.544 including W3).



The average ergosterol concentration relativized for carbon concentration demonstrates that there is a high amount of ergosterol, a proxy for fungal content, relative to the amount of carbon in the dune. In the moraine, the opposite trend is shown. There is not a lot of fungi for the concentration carbon (Fig. 5). These differences in relative ergosterol concentration were marginally statistically significant ( $F=4.69$ ,  $p=.071$ ).

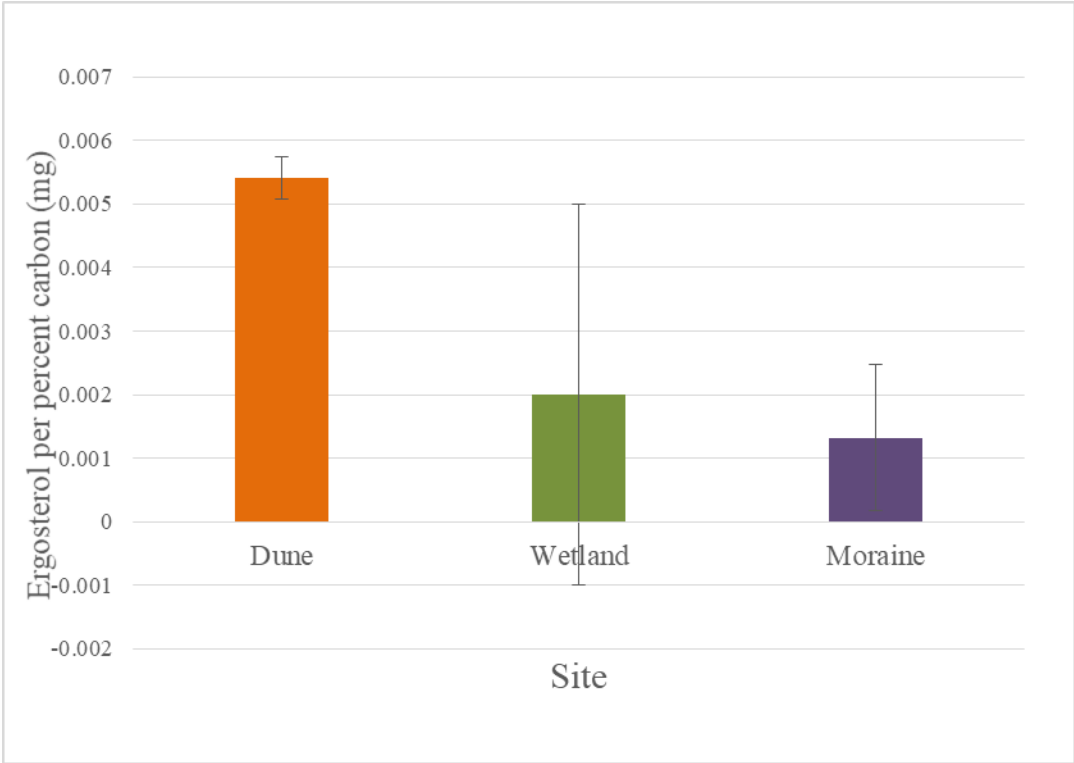


Figure 5. Mean ergosterol (mg/mL) over carbon concentration (%) at each site.

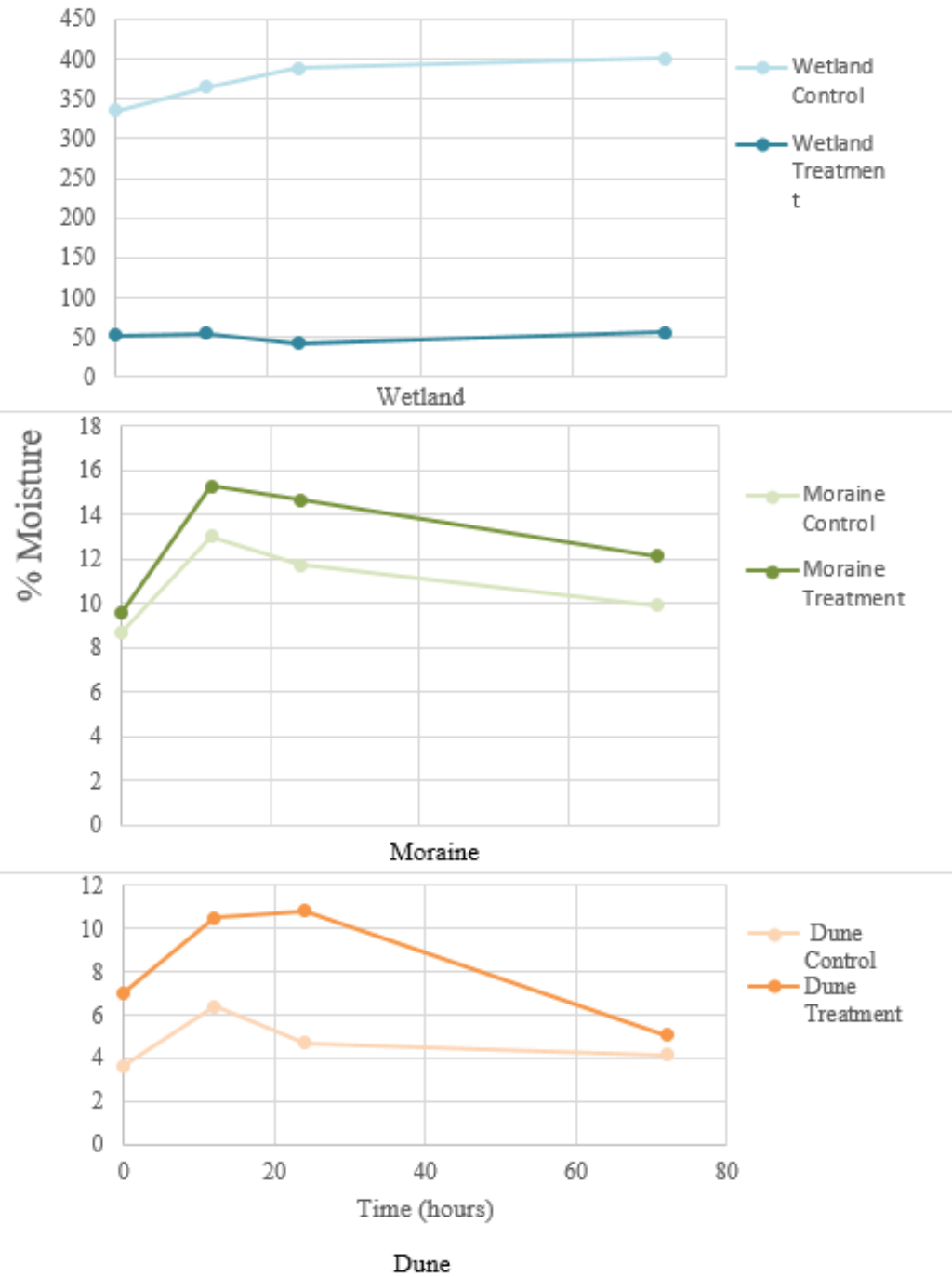


Figure 6. Soil moisture in the three different plots through time. We excluded wetland control because it had a much higher gravimetric weight than the rest of the data.

Enzymatic activity displayed different trends in each of the sites (Fig. 7). In each of the treatment sites, activity dropped at 12 hours post-water treatment. After that, the dune enzyme activity increased, moraine activity continued to increase then decrease, and wetland continued to decrease, then increase. For the dune and moraine, these results were almost marginally statistically significant ( $F=3.4088$ ,  $p=.107$ ). The wetland's enzymatic activity did not demonstrate statistical significance.

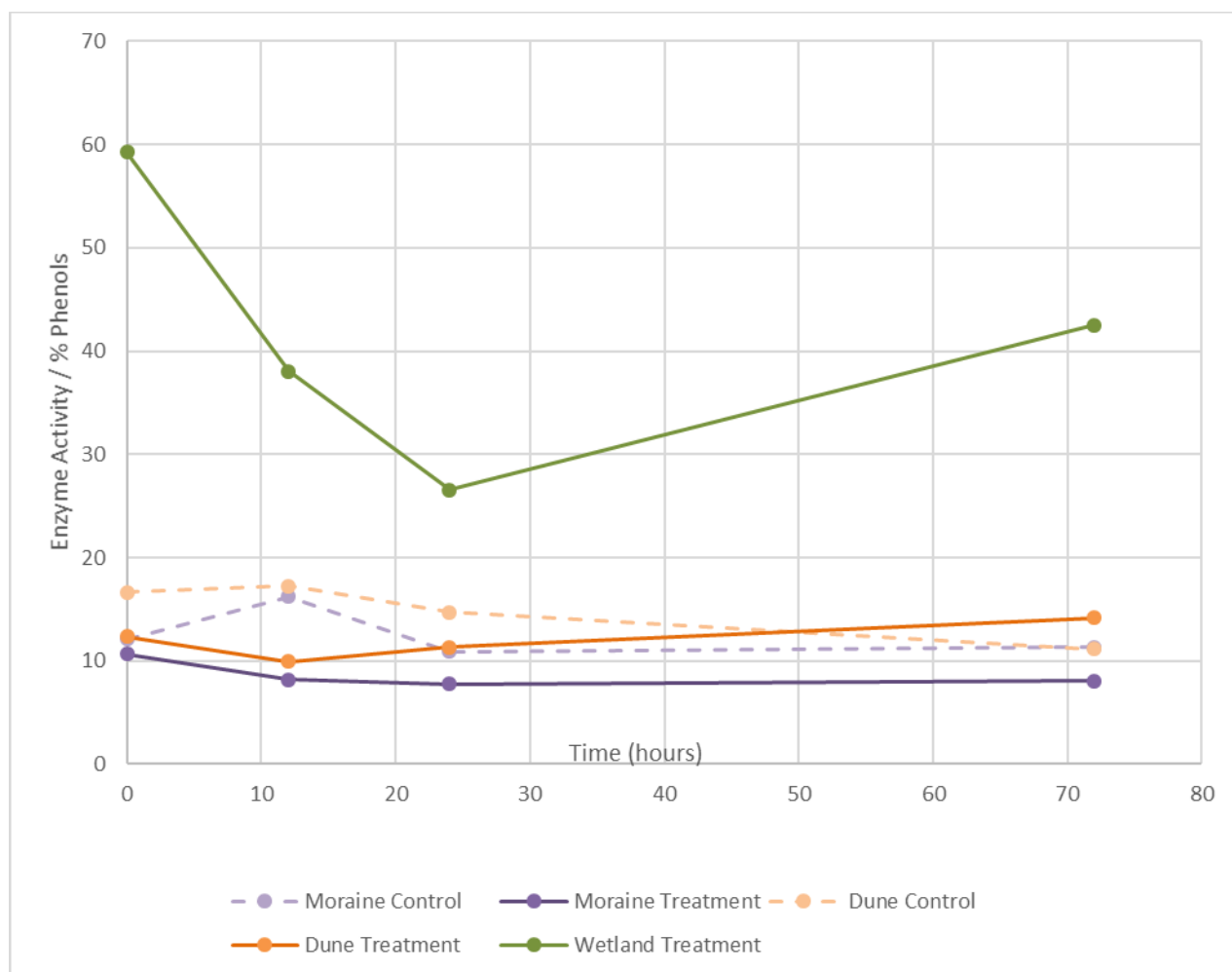


Figure 7. Enzyme activity over phenolic composition in the three different plots through time. The controls for the dune and moraine are seen as dotted lines. Wetland control was excluded due to how much higher the values were.

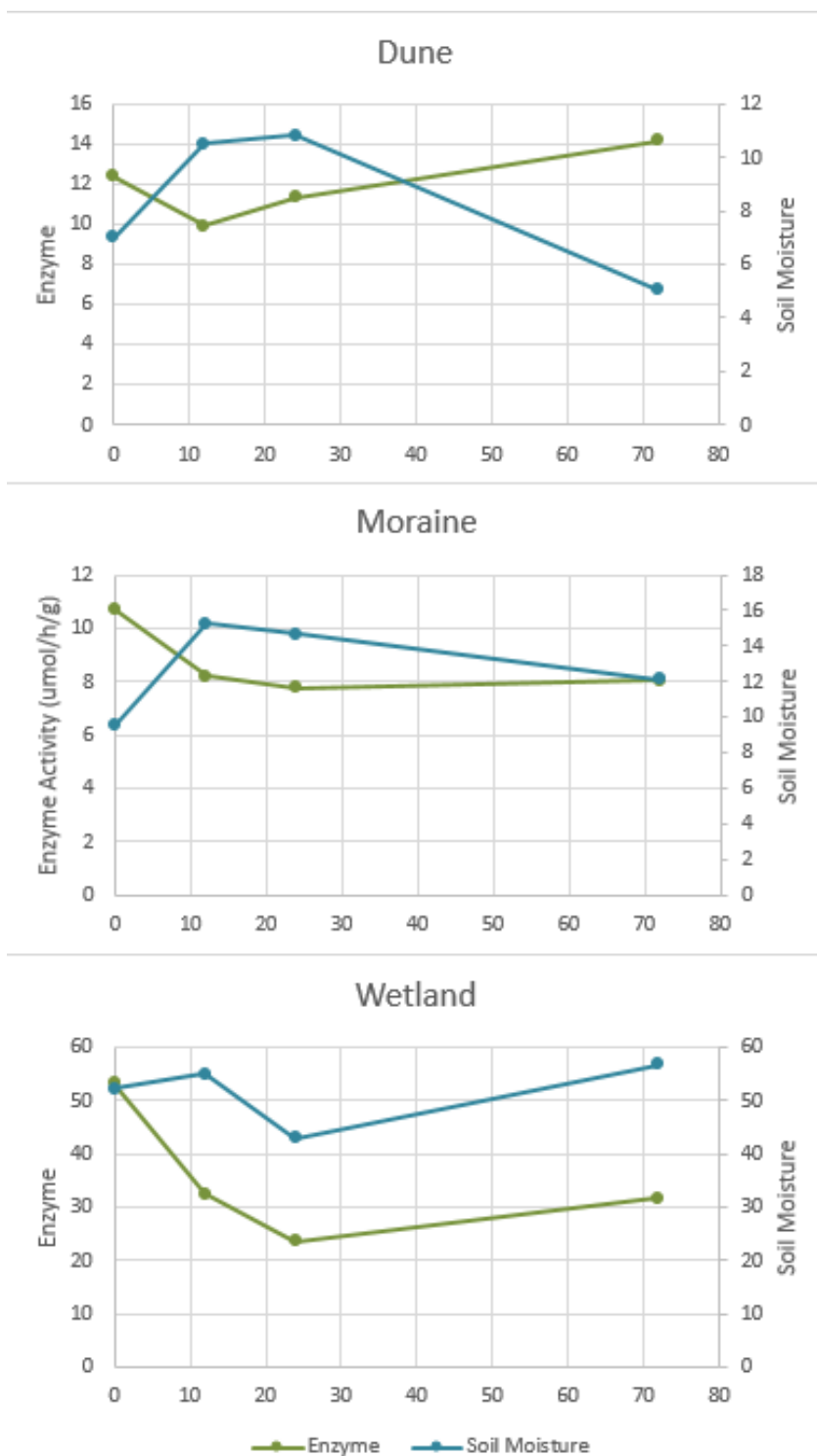


Figure 8. Enzyme activity (relativized for phenolic concentration) and soil moisture in the different plots through time.

**Discussion:**

Each of the sites was very different from one another in a number of ways. The site differences were large, especially for carbon (%) and phenolic content relative to carbon (%) (Fig. 1, 3). Another site difference that demonstrated marginal significance was levels of ergosterol relativized to carbon concentration (Fig. 5). This supported our hypothesis that differing sites would show strong differences in both carbon and fungal content. The simulated intense rainfall had a mixed effect on enzyme activity. As we predicted, this effect was not as strong on the wetland site and was what we expected in the moraine site, but the trend in activity was the reverse of what we expected on the dune site.

We were able to determine the overarching, long term effects of differing soil types on fungal communities and activity. For example, in the dune, there was a low amount of carbon and a high amount of ergosterol per carbon (Fig. 1, 5). This could be indicative of a fungi-dependent environment. This contrasts the moraine, which had a mid-ranged carbon content and a low ergosterol to carbon ratio (Fig. 1, 5). Additionally, the moraine had a high phenol to carbon ratio (Fig 3). The moraine may depend more on other sources of decomposition, such as animals and bacteria. This environment with low fungal presence is further reinforced by the high presence of phenols, which saprotrophic fungi break down. The wetland demonstrated high carbon concentration, a relatively low ergosterol to carbon ratio, and a low phenol to carbon ratio (Fig. 1, 3, 5). This could indicate that perhaps a high proportion of wetland fungi are productive saprotrophs that are efficient at breaking down phenolic bonds, as it had a relatively low concentration of phenol in the soil.

To understand the fungal responses to the intense rainfall event, the degree to which soil moisture was retained in the sites must be assessed. Initially in each of the sites, the soil moisture

increased, but it varied after 12 hours. The moisture content also increased in the control plots, but we are unsure of the variability in the soil and are therefore unable to make conclusions on the controls. Based on the soil types, the soil moisture trends that we expected to see, occurred. In the sandy dune, the moisture increased greatly at first, then quickly dropped due to the sandy soil, which had more difficulty retaining water (Fig. 6). In the wetland, the moisture increased temporarily and quickly decreased, then increased back to the normal amount (Fig. 6). Due to the variability in the wetland data and the high baseline levels of water, we believed the overall moisture to have not been greatly affected by our rainfall simulation. In the broader spectrum of soil types of the moraine, moisture increased and then slowly decreased through the 72 hours (Fig. 6). It held water better than the porous dune and the saturated wetland.

In both the dune and the moraine, the enzymatic activity was suppressed, to some extent, with the addition of water (Figures 7, 8). As is demonstrated in Figure 8, soil moisture and enzyme activity displayed inverse patterns – as the soil moisture increased, the phenol oxidase activity decreased (Fig. 8). The treatment did not appear to have an effect on the wetland, at least not to the extent that we can conclusively determine. Soil moisture in this site did not change much, and as a result we cannot conclude how our simulation affected the enzyme activity. This demonstrates that water did not appear to be a limiting factor in any of the environments, and perhaps the increased soil moisture impacted the ability of fungi to obtain organic carbon.

Our results agree with a similar study done on fungal abundance and soil carbon in California by Hawkes et al. (2010). Their study was performed over the course of four years and tested the effects of ambient rainfall additions on different plots of land. They concluded that fungal communities (measured through ergosterol and Bradford reactive soil proteins) were more abundant and diverse under low rainfall, and that fungal communities were less abundant and

diverse under high rainfall. Additionally, they concluded that fungal responses may be based on the availability of water for metabolic activity; only in limited water conditions will fungi abundance respond positively to additional rainfall (Hawkes et al. 2010).

Our results for the phenol oxidase activity were marginally statistically significant, but they were not significant enough to draw strong conclusions. This may have been a result of many sources of error. For instance, in the analytical chemistry portion of the lab, we may have made errors in collecting our spectrophotometer data, as there were a lot of free floating soil particles in our “background”, or fluid without L-dopa, readings. For future directions, it is advisable to make sure the plots at each site are as uniform as possible. Our wetland control was much different from the treatment sites, and thus much of the data was unusable.

We saw a short term response to the addition of water. However, there needs to be more research done to conclusively understand the long term effects of increased rainfall on terrestrial ecosystems and northern Michigan’s landscapes in the future. An understanding of how saprotrophic communities are affected by heavy precipitation is key in predicting the response of the soil, and therefore carbon cycling, to future climate change.

*Works cited*

Crowther, T. W., Boddy, L., & Hefin Jones, T. (2012). Functional and ecological consequences of saprotrophic fungus–grazer interactions. *The ISME Journal*, 6(11), 1992–2001.

“Data Tools: Daily Weather Records.” National Climatic Data Center, NOAA,

[www.ncdc.noaa.gov/cdo-web/datatools/records](http://www.ncdc.noaa.gov/cdo-web/datatools/records).

Hawkes, C. V., Kivlin, S. N., Rocca, J. D., Hudget, V. , Thomsen, M. A. and Suttle, K. B.

(2011). Fungal community responses to precipitation. *Global Change Biology*, 17: 1637-1645.

Herrmann H.J. (2007) Dune Formation. In: Schadschneider A., Pöschel T., Kühne R.,

Schreckenberg M., Wolf D.E. (eds) *Traffic and Granular Flow'05*. Springer, Berlin, Heidelberg.

Karowe, D. 2018. Effect of Glacial History on Current Plant Communities. *General Ecology at UMBS*. 1-3.

MDEQ (Michigan Department of Environmental Quality). (2015). Michigan Background Soil Survey 2005 (Updated 2015). [https://www.michigan.gov/documents/deq/deq-rrd-MichiganBackgroundSoilSurvey\\_495685\\_7.pdf](https://www.michigan.gov/documents/deq/deq-rrd-MichiganBackgroundSoilSurvey_495685_7.pdf). Accessed August 9, 2018.

Sinsabaugh, R. L. (2010). Phenol oxidase, peroxidase and organic matter dynamics of soil. *Soil Biology and Biochemistry*, 42(3), 391-404.

Thomas, C.D., Cameron, A., Green, R.E. et al. (16 more authors). (2004). Extinction risk from



climate change. *Nature*, 427 (6970), 145-148.

Tiner, Ralph W. Wetland Indicators: a Guide to Wetland Identification, Delineation,

Classification and Mapping. 2nd ed., CRC Press/Taylor & Francis Group, 2017.