

Short- Term Survey of mycorrhizal Fungal Biodiversity and Species Richness in UMBS Burn Plots

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Fungi form a tight symbiosis with plants through a mutualism known as arbuscular mycorrhizae, benefiting both the fungi and the plant. These symbioses are very specific and can be accounted for by the fruiting bodies, usually appearing under the plant species in which they form a relationship with. At the University of Michigan Biological Station in Pellston, Michigan a one week survey was conducted at plots burned at different times to observe how succession influences fungi biodiversity and species richness. A correlation was found between the plots and the species richness, the older plots having more species present. The data for biodiversity had some promising suggestions when it comes to species richness, but a further study needs to be completed to say anything of significance about biodiversity and regression.

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INTRODUCTION

Variation of life forms within a given ecosystem is the result of 3.5 billion years of evolution. Biodiversity provides many ecosystem services that are often not readily visible. It plays a part in regulating the chemistry of our atmosphere and water supply. Biodiversity is also a sign of health, directly involved in water purification, recycling nutrients and providing fertile soils.

Fire is used as a management tool in a variety of ecosystems in an effort to increase biodiversity and promote ecosystem sustainability (Frost 1985). At University of Michigan Biological Station in Pellston, Michigan U.S.A., controlled burns are used to observe succession. Ecological succession is the progressive replacement of the species in a community after a disturbance has occurred in the community (Chandler et al. 1983, Krebs 1994). Forest fires can cause destruction of soil structure and soil microbial communities on the top few centimeters of soil (Dumontet *et al.* 1996; Neary *et al.* 1999), increase susceptibility to soil erosion (Neary *et al.* 1999), allow certain seed species to germinate (Clark & Wilson 1994), and increase opportunity for the colonization of exotic plant species (Allen 1991). The effects of fire on soil microbes have received less attention than above-ground processes, but the soil microbial community has a significant impact on the plant community composition. Mycorrhizal fungi form a symbiosis with plants that is critical to the establishment and survival of many plant species.

Symbiosis is defined broadly as “two or more organisms living together” and in most cases both partners benefit (Lewis 1985). Symbiosis occurs frequently in nature, in fact the chloroplast found in plants comes from a symbiotic cyanobacteria (citation). A common symbiotic relationship is between fungi and the roots of trees; fungi aid the nutrient uptake of roots and receive a small amount of sucrose for their service. This symbiotic relationship is known as mycorrhizae, fungi colonizes roots forming a biological link between the root and the soil.

The mycorrhizal mycelia of fungi are much smaller in diameter than the smallest root, this results in a larger surface area to volume ratio so more nutrients can be absorbed into the plants (Kirk 2001). Also, plants supported by mycorrhizae are more vigorous and better adapted to growth in adverse conditions, such as distressed soils due to drought, high salinity, or under biotic stress. Furthermore the recovery from stress conditions is faster in mycorrhized plants. For example, plants growing in soils with a basic pH may be incapable of taking up phosphate ions but the mycelium of the mycorrhizal fungi associated with them can access the otherwise unavailable nutrient sources (Li et al., 2006). In Michigan, soils are noted for their low clay composition rendering a low cation exchange capacity where the soil cannot easily retain nutrients (Spurr and Barnes, 1980) so mycorrhizal associations are needed for successful growth of plants.

It is estimated that more than 90% of the plant species form a mycorrhizal symbiosis. This relationship begins when a spore germinates and infects a root. It can take a long time for the fungus to spread to a significant portion of the root system, therefore young forests do not have as intensive mycorrhizae as older ones. When mycorrhizae diversity increases in a given

area, plant species diversity also has shown to increase. In addition, plants that have formed symbiosis with mycorrhizae are able to outcompete plant species that are unable to form such symbiosis (Erikson 2001).

There are two types of mycorrhizae, ectomycorrhiza (EM) and endomycorrhiza. The ectomycorrhiza is an association that takes place right on the surface of the roots. EM fungi can reproduce in the absence of a plant host and form a facultative mutualism. Arbuscular mycorrhizae (AM) are endomycorrhizal by nature. They penetrate into the root cortex and form arbuscules within the root cells. They only can reproduce themselves when in presence of a host plant and are "obligate symbionts". There is a saying among fungophiles that states, "Know the tree, know the mushroom." AM fungal species are very specific for certain types of trees. Among the species not forming endomycorrhizal symbiosis are the brassicaceae and conifer families. (Daniels 2006).

To test how mycorrhizae are shaped by succession and biodiversity, we surveyed plots of land going through different stages of succession. Although succession is not a rigid or finite process, it can be described in terms of unidirectional stages of increasing complexity. After a primary or secondary disturbance, a forest enters a pioneer stage. Next there are intermediate stages, and lastly a forest will enter a climax stage where succession stops and equilibrium occurs.

We will observe plots of land at different stages of succession to see how the fungal diversity differs. We hypothesize that the plots at the pioneer stage will have lower mycorrhizae, and therefore lower diversity, than plots in intermediate or late stages of succession. It takes time for mycorrhizae to colonize plants so older plots will also have more mycorrhizae. Also, because

mycorrhizae are specific for distinct species, we hypothesize that in more diverse areas, different types of AM and therefore more types of fungi species will be found.

SITE DESCRIPTION

Our survey was conducted at University of Michigan Biological Station burn plots in Pellston, Michigan (S72, T37, R4W). The plots are located on glacial outwash from the Valdres glacial period about 12,000 years ago. Upon retreat of the glaciers, the ice began to melt forming Lake Algonquin. Part of the outwash became an island in Lake Algonquin. The University of Michigan burn plots are located on this former island. Therefore the soil of the plot is a sandy series of glacial outwash (Reza 1987). It was once part of a thriving northern hardwood-pine community (Dorr and Eschmann, 1977). Intensive logging began in 1840, causing the area to evolve into an oak-maple community with big-tooth aspen and white pine interspersed. Repeated fires also had a serious impact on the area's vegetation. The first fire occurred in 1880 and was followed by others in 1890, 1896, 1899, 1907 and 1911.

The burn plots were initiated by Dr. Frank Gates in order to study forest succession (Meyers, 1982). In 1936, Gates clear cut a 100 x 100 m plot and then burned it to continue the study of succession (Scheiner and Teeri 1981). Gates then burned two more plots in 1948 and 1954. The study continued, being taken up by Scheiner and Terri in 1980, since then a plot was again burned in 1998 (figure 7). Because of the precise knowledge of when and how the plots were disturbed, they offer a unique opportunity to study secondary succession.

METHODS

The plots we chose to survey were burned in 1936, 1954, 1980 and 1998. To measure each plot we used a 100-meter long tape measurer. To find our 'start point' we began at the northeast corner of each plot. Next we measured twenty meters south and twenty meters west from the northeast end. We then walked directly west until we were twenty meters from the northwest corner of the plot. We walked using the 100-meter long tape measure to have a reference point for our transecting.

This method helped to avoid the effects of the edge of the forest. Edge effect was taken into account because of the abiotic and biotic differences between the edge and the inner portions of the plot. For example, temperature on the forest edge is greater than temperature within the stand as a result of higher light intensities. Similarly, moisture and relative humidity are less on the perimeter than within the stand (Harris, 1984).

We used meter sticks to walk along the rolled out tape, surveying the area two meters out on each side of the tape. While surveying, we identified and tallied up the amount of different fungal specimens found. To identify we used the book "Mushrooms Demystified" by David Arora and the help from researcher Luke Nave at UMBS.

We performed five transects per each plot in total (two on July 28, 2010; one on August 1, 2010; two on August 4, 2010). Each subsequent transect began five meters south of the previous transect. To test diversity we surveyed the amount and type of fruiting bodies found in a given area. We statistically analyzed our results using a species richness test (the number of different species in a given area) for each burn plot. We also tested the average specimens per transect for each burn plot. We tested for biodiversity using the Shannon Diversity Index and the Simpsons Diversity Index. We also compared the Shannon Diversity Index using a tukey test.

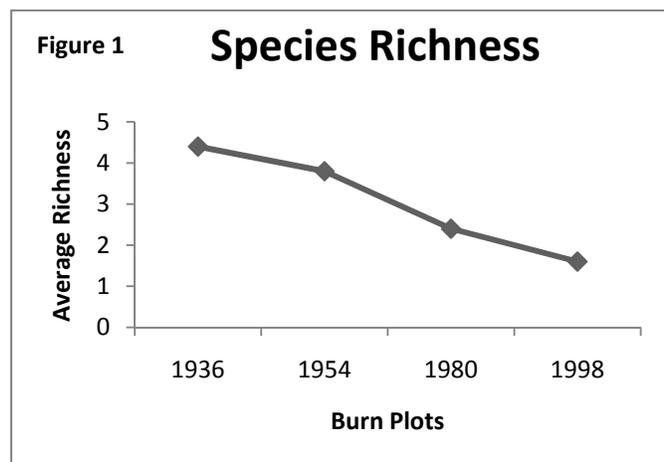
Figure 1	Species Richness	Figure 4	Biodiversity
Figure 2	Average specimen # / plot	Figure 5	Regression and Nova test
Figure 3	Above graphs w/out <i>C minor</i>	Figure 6	Map of Burn Plots

*Key to Data Analysis

RESULTS

In the 1936 plot, 1,875m² of area was covered and 375 meters were transected, 141 fungal specimens were found. In the 1948 plot, 1,650m² of area was covered and 330 meters were transected. 547 fungal specimens were found. In the 1980 plot, 2,000m² of area was covered and 400 meters were transected. 36 fungal specimens were found. In the 1998 plot, 1,855m² of area was covered and 371meters were transected. 11 fungal specimens were found.

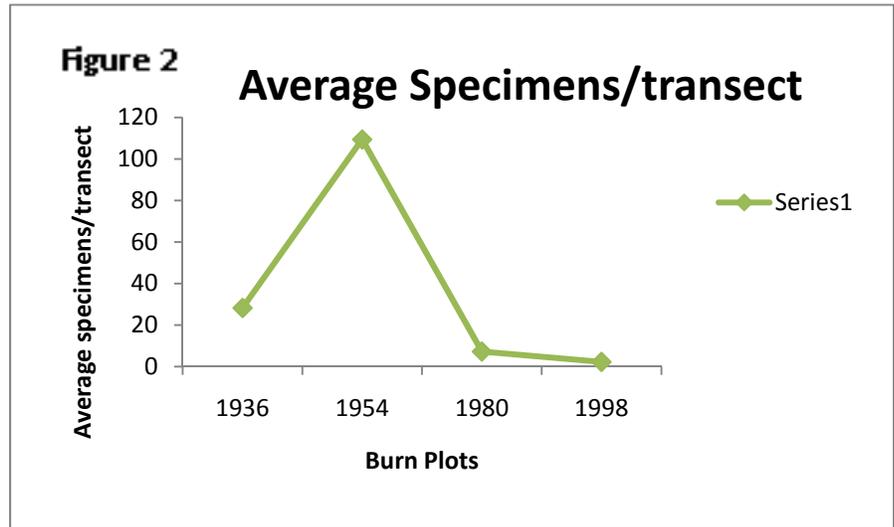
The species richness graph (Figure 1) shows a negative correlation with species richness and years of fire. Species richness was the highest for the oldest burn plot surveyed in 1936 with an average of 4.4 species per transect. It declined to 3.8 species per transect in 1954 followed by an



average of 2.4 species per transect in 1980 and lastly and declined to 1.6 species per transect in the 1998 burn plot. The average biomass per transect was 28.2 for the 1936 plot, 109.4 for the 1948 plot, 7.2 for the 1980 plot, and 2.2 for the 1998 plot. (figure 2)

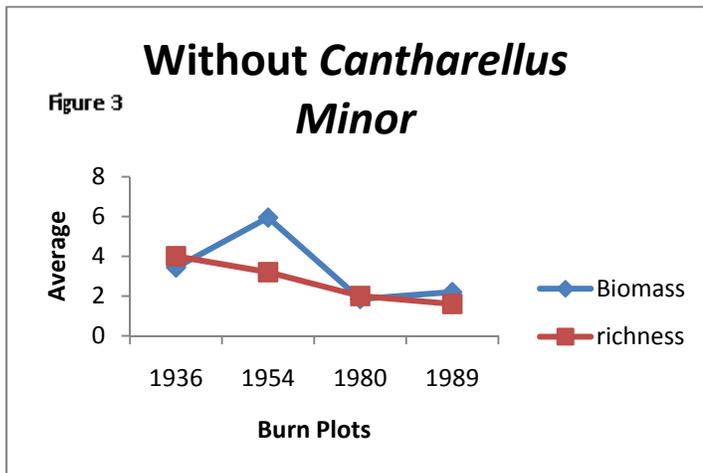
Without *Cantharellus minor* the average specimens per transect lessens drastically (figure 3).

The average specimens per transect was 3.4333 for the 1936 plot, 5.94667 for the 1948 plot, 1.85 for the 1980 plot and 2.2 for the 1998 plot. The species richness without *Cantharellus minor* (figure 3) shows a less



negative slope than the species richness with *Cantharellus minor*. The line decreased by .4 to 4.0 species per transect in 1936, it declined to by .6 to 3.2 species per transect in 1954 followed

by a drop of .4 to 2.4 species per transect in 1980, lastly it stayed the same at 1.6 species per transect in the 1998 burn plot, for there were no *C. minor* specimens there. The drop was the largest in 1954 because the largest amount of *C. minor* was in that plot.



Biodiversity Data Analysis

Transect	average Shannon	average Simpsons
1936	0.86984636	0.44620864
1954	0.523152	0.36247
1980	0.63657	0.61869
1998	0.46648	0.72222

The average Shannon Index for Biodiversity (Figure 4) was highest in 1936,

Figure 4

followed by 1980, 1954 and 1998. The Average Simpson's Index shows the most diversity in the 1998 plot followed by the 1980 plot, 1936 plot, and lastly the 1954 plot. We also ran a 'tukey test' to see if there were any significant differences in Shannon's Diversity between the four plots 1,2,3 and 4. The results are insignificant when looking at the significance column.

Regression Statistics

Multiple R	0.793193
R Square	0.62915513
Adjusted R Square	0.4437327
Standard Error	0.13312745
Observations	4

ANOVA

	<i>df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>
Regression	1	0.06013547	0.06013547	3.39309068	0.206807
Residual	2	0.03544584	0.01772292		
Total	3	0.09558131			

	<i>Coefficients</i>	<i>Standard Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>	<i>Upper 95%</i>	<i>Lower 95.0%</i>	<i>Upper 95.0%</i>
Intercept	0.89818236	0.16304716	5.50872727	0.03140891	0.19664703	1.59971769	0.19664703	1.59971769
transect total	-0.10966811	0.05953641	-1.84203439	0.206807	-0.36583259	0.14649637	-0.36583259	0.14649637

Figure 5

According to the p-value, regression was insignificant for the transect totals. (Figure 5)

DISCUSSION

A complete data set regarding how long it takes for mycelium to colonize and how succession and diversity influence mycorrhizae would be very useful in understanding forest and fungal ecology. Though our data is promising, more could be said if a yearly survey was conducted. That said, much was learned from the three-day survey and our hypothesis still holds.

For Species Richness, we fail to reject our hypothesis. Indeed, the data shows the older burn plots being more diverse than the younger ones. The 1936 burn showed the greatest species richness followed by the 1954 plot, 1980 plot, and lastly the 1998 plot. In the 1998 plot only bracken fern and aspen were found in the area. The 1980 plot was almost 20 years older in growth, but did not have too many more species. The 1980 plot did have some conifers, but if one recalls from the introduction, conifers do not form AM with fungi. Instead they have ectomycorrhizae, which can occur with or without the plant species with which they form a symbiosis. The 1954 plot was in a stage of intermediate succession and was quite diverse with many conifers, huckleberries, blueberries, big toothed aspen, red oak, red maple, and bracken fern. The 1936 plot being 22 years older than the 1954 plot was at a later stage of intermediate succession.

Studies completed by Scheiner, Sharik, Roberts, and Vande Kopple found that the successional trend in the burn plots went from bigtooth aspen to white pine, red pine, red oak, and red maple (Scheiner et. al. 1988). They also observed species richness to increase with time, which was the trend we also found in our data.

According to Connells Intermediate Distribution hypothesis; species richness is maintained by an intermediate level of disturbance. If disturbed too much or too little, diversity will be low. The 1936 and 1954 plot may be at mid level disturbance, thus accounting for the

greater number of species observed. Since the 1998 and 1980 plots were recently burned the frequency of disturbance may be too high for the species to be rich. They also may have had low species richness because mycorrhizae take time to colonize.

Shannon's Biodiversity Index was useful in that it takes into account the number of species accounted for and the evenness of the species. The index is increased either by having additional unique species, or by having a greater Species evenness. Since our 1954 sample was filled with fruiting *C. minor*, the index was higher for the 1980 plot due to unevenness of the 1954 plot. Although there were more species present in the 1954 plot, the evenness factor through off the data.

The Simpsons Diversity differs from Shannon's Index in that it takes into account the number of species present, as well as the relative abundance of each species. The Simpson index represents the probability that two randomly selected individuals in the habitat belong to the same species so a lower percentage represents a more diverse area (Krebs 1994). That means that in the 1998 plot there is a 28 % chance that two randomly selected specimens will be the same. Though only eleven specimens were found in total for the 1998 plot, they were evenly distributed amongst six different species. In the 1954 plot, 473 specimens out of the 548 collected were *Cantherelles minor*. This makes for a very uneven sample with a 64 % chance one will pick a specimen of the same species in the 1954 plot.

Because *Chanterelles minor* is so frequent in number, we ran the data analysis for species richness and average specimens per transect with and without it and the slope of the graphs lessened. Many fungi fruit seasonally (for example on can usually only find morels around mother's day) and we happened to survey when the *C. minor* fruiting bodies were active. Though morel mycorrhizae may be in the area, they are not observed because it is not their

fruiting season (Mueller 2004). Since our survey was only conducted over a period of one week, the biodiversity is skewed and a survey needs to be conducted from spring to fall to accurately assess the true biodiversity and species richness. Based on the insignificant results of the regression data and the tukey test, a more thorough survey is needed to find more significance.

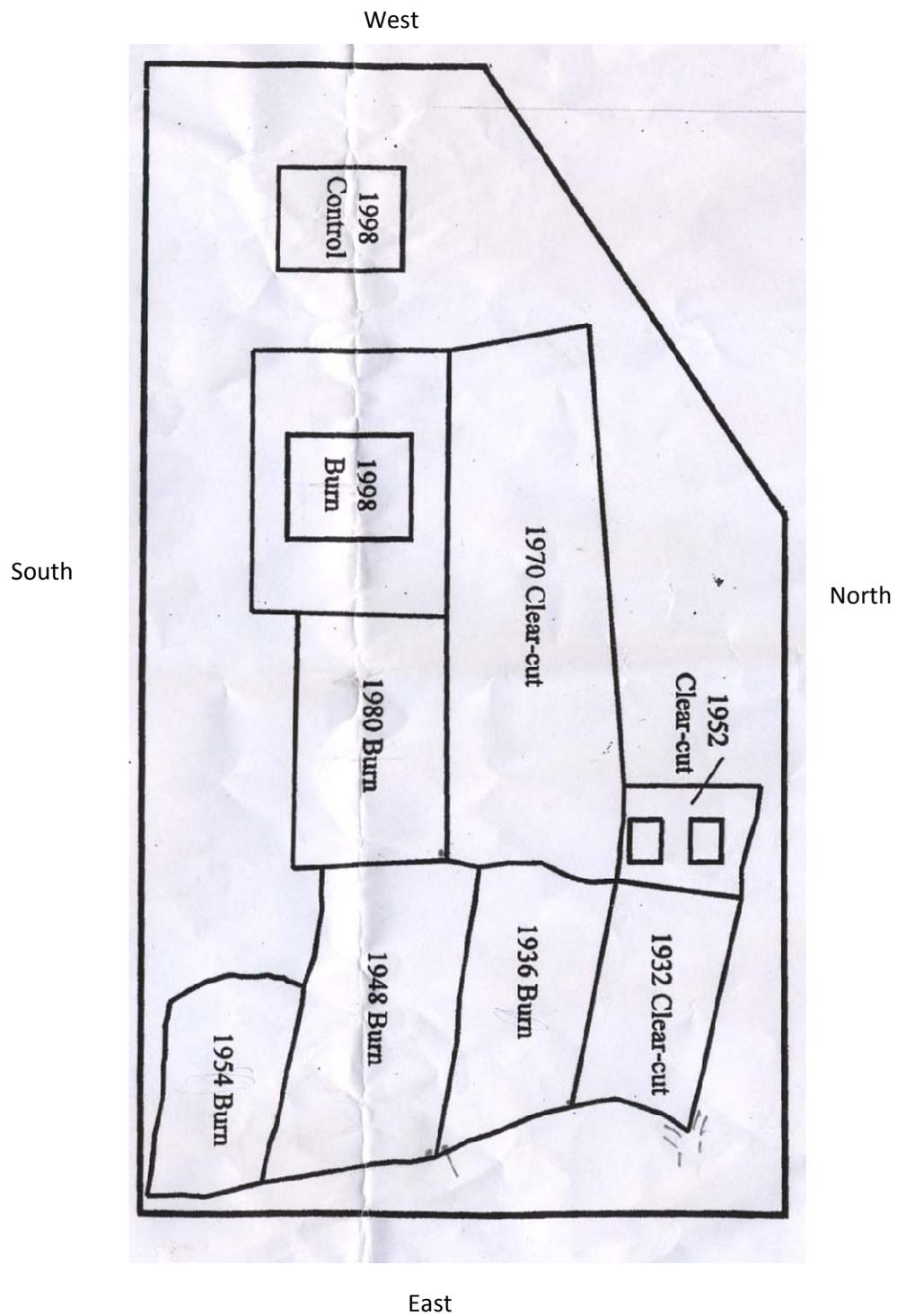


Figure 6 This is a map of the burn plots we are analyzing. Taking edge effect into account, we start from the northeast corner in each plot, going parallel to the adjacent plots, we work our way to the northwest corner. We are surveying the 1936, 1954, 1980, and 1998 burns.

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