

Soil Ecology Project

The Effect of Buildings on Mycorrhizae

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Background

Mycorrhizae are an important part of soil ecology that play a key role in the survival of plants. In a symbiotic relationship between plants and fungi (New York Botanical Garden, 2003), the mycelium of the fungi attach to the roots of the plant and the soil surrounding it (Swift, 2004). Then the fungi colonize the root using what is known as a Hartig net, a complex network of fungal hyphae that exchange nutrients between the fungus and the host plant. The mycelium can cover the cells of the root on the outside or penetrate into the root cells (New York Botanical Garden, 2003). But either way, the fungi thereby form a connection between the soil and the roots of the plant (Swift, 2004) with the hyphae transporting water and mineral nutrients such as phosphorus, nitrogen, zinc, copper, nickel, chloride, and sulphate (Swift, 2004) from the soil to the roots (Trappe, 2012). This enables the plant to get more nutrients and more water and therefore, without mycorrhizae, many of the plants on land would not be able to survive as easily. In return for helping the plant to absorb nutrients, the fungi receive carbohydrates made by the plant during photosynthesis. The fungi use the carbohydrates for their own energy needs (New York Botanical Garden, 2003).

Mycorrhizae also provide other benefits that increase the plant's longevity. The network of hyphae, for example, helps to hold the soil together, making irrigation easier and providing weed resistance. Experiments have shown that plants are more diverse and grow taller when mycorrhizae are added. Furthermore, the mycorrhizae help protect the plant's roots from diseases, and build up stress resistance in it (St. John, n.d.).

Two particularly important nutrients that mycorrhizae help a plant to obtain are nitrogen and phosphorus. Mycorrhizae assist the plant in absorbing nitrogen by sending out enzymes that can mobilize organic nitrogen, enabling the plant to get it more easily (Swift, 2004). Nitrogen

also improves plant growth and helps the plant quicken the circulation of nitrogen in its environment (New York Botanical Garden, 2003). Another nutrient, phosphorous, improves a plant's growth and resistance to the effects of drought (St. John, n.d.) because phosphorus assists the plant in photosynthesis and is an essential part of ATP, DNA, and RNA. Both phosphorus and nitrogen are essential to plant life because the enzymes formed by RNA control chemical reactions that run the cell. Without the phosphorus and nitrogen, cells could not live. Nitrogen also improves plant growth and helps the plant quicken the circulation of nitrogen in its environment (New York Botanical Garden, 2003). However these nutrients cannot help the plant during construction, when the mycorrhizae are disturbed (Todd, 2004).

When the school building was built, the soil was ripped from the ground, disturbing the mycorrhizae living in it. The soil surrounding the school was also disturbed with the creation of gardens, and other landscaping, and when an area like the RPCS campus is affected by construction, it lacks important components of the soil that help plants to survive. When the soil is frequently taken from the ground during construction, the mycorrhizae and relationships it has built can be disturbed (Todd, 2004).

Our problem is questioning whether or not a tree's distance from the school has an effect on the population of mycorrhizae in the soil surrounding it. We are testing the soil surrounding two of the same species of trees and the negative control, all of which are different distances from the school, and finding the population of the mycorrhizae in the soil. The soil can be disturbed by construction, and the areas in which construction is most prominent are in closer proximity to the school. The problem for our test is important because we want to find out whether or not humans are having a significant effect on the health of the trees surrounding buildings. Mycorrhizae are beneficial to trees, and if their population is suffering due to the

effects of humans, then that may be negatively affecting the trees surrounding us. This is an issue because if the population of mycorrhizae in the soil surrounding the tree is low then that could be negatively the health of the tree. Using this information we developed the hypothesis that the soil surrounding the trees closer to the building will have a smaller population of mycorrhizae living in it, compared to soil surrounding trees farther away from the building.

Experiment

- I. Problem: Does the proximity of trees to a building increase or decrease the density of fungi around those trees' roots?
- II. Hypothesis: As the distance from the building increases, the density of fungi will increase around the trees' roots
- III. Procedure:
 - A. Independent Variable: the distance from the tree to the building
 - B. Dependent: the density of Fungi found in the soil surrounding the tree's roots
 - C. Negative Control: sample of soil from front lawn
 - D. List of Controls:
 1. Sugar vs. red maple tree to control for plant diversity
 2. Amount of water used in the serial dilutions
 3. Amount of soil used in the serial dilutions
 4. Type of petri plate with nutrient agar
 5. Type of water
 6. Amount of water used to dilute the soil
 7. Time allowed for fungus to grow

8. Time at which the samples were taken
 9. Type of device used to extract the soil
 10. Amount of soil extracted
 11. Age of the soil
 12. The degree of dilutions
 13. the amount of the dilution plated
 14. The dilutions that are plated
- E. Step- by- Step Instructions:
1. **All soil samples in steps 1-2 must be taken at the same time on the same day.** Find two types of trees that are different species (sugar maple and red maple) and different distances from the building, so that:
 - i. One sugar maple tree (1) is 25 m. away from the building at coordinates N: 39.35763 W: 076.63570.
 - ii. The second sugar maple tree (2) is 82 m. away from the building at coordinates N: 39.35845 W: 076.63538.
 - iii. the first red maple tree (3) is 144 m. away from the building at coordinates N: 39.35907 W: 076.63546
 - iv. The second red sugar maple tree (4) is 81 m. away from the building at coordinates N: 39.35938 W: 076.63549
 - v. The front lawn (5) is 42 m. away from the building at coordinates North: 39.35809 West: 076.63549.
 2. **Take one soil sample from each location at the same time on the same day** that is 30.5 cm away from each tree by turning the soil core (2cm diameter)

clockwise until it reaches 15 cm and put the sample in a plastic bag that is labeled with the corresponding number.

3. **Serial dilutions in steps 3-14 in test must be done at the same time. Use the same sample bag for all trials, but each trial must be done at a different time.** Use a clean, new transfer pipette to add 10 ml of sterile water to a 15 ml culture tube. Label the tube "tree 1, 10^0 ."
4. Label a second 15 ml culture tube "tree 1, 10^{-1} ." With the same pipette that was used to put sterile water in the "tree 1, 10^0 " tube, add 9 ml of sterile water to the "tree 1, 10^{-1} " tube.
5. Label a third 15 ml culture tube "tree 1, 10^{-2} ." With the same pipette that was used to put sterile water in the "tree 1, 10^0 " tube and the "tree 1, 10^{-1} " tube, add 9 ml of sterile water to the "tree 1, 10^{-2} " tube.
6. Using a 1 cc scoop, put 1 cc of soil from "tree 1" into the "tree 1, 10^0 " tube.
7. Cap the "tree 1, 10^0 " tube and shake it vigorously.
8. Using a new, clean pipette, take 1 ml of the water and soil mixture from the "tree 1, 10^0 " tube and add it to the "tree 1, 10^{-1} " tube.
9. Cap the "tree 1, 10^{-1} " tube and shake it vigorously.
10. Using the same pipette that was used to transfer 1 ml of the mixture from the "tree 1, 10^0 " tube to the "tree 1, 10^{-1} " tube, take 1 ml of the water and soil mixture from the "tree 1, 10^{-1} " tube and add it to the "tree 1, 10^{-2} " tube.
11. Cap the "tree 1, 10^{-2} " tube and shake it vigorously.
12. You should now have a total of three culture tubes with water and soil mixtures.

13. Label three 3M petrifilm™ Yeast and Mold Count Plate petri plates containing nutrient agar with "tree 1 trial 1 10^0 ," "tree 1 trial 1 10^{-1} ," and "tree 1 trial 1 10^{-2} ." Place 100 μ l samples from the 1st, the 2nd and 3rd tubes (dilutions 10^0 , 10^{-1} , & 10^{-2}) onto their corresponding petri plates.
14. Repeat steps 3-15 with the soil samples from trees 2, 3, 4 and 5.
15. Allow to grow for 48 to 72 hours.
16. Examine each of the plates for individual fungus colonies and start with the plate with the lowest dilution that has fungus on it. From the lowest dilution to make your estimates of the number of mold which are fuzzy dots and yeast which are the more defined dots in the original 1 cc soil sample using the formula below:

Note: the molds and yeast may come from different dilutions.

Microbes in 1 cc of soil = # Colonies on sheet $\times 10^2 \times 10^{\text{dilution \# at which these colonies were found}}$.

17. Do trial one on day one in the morning, then go back up to step three and do trial two in the afternoon on day 1. Finally go back up to step three once again and do trial three on day 1 after trial 2.

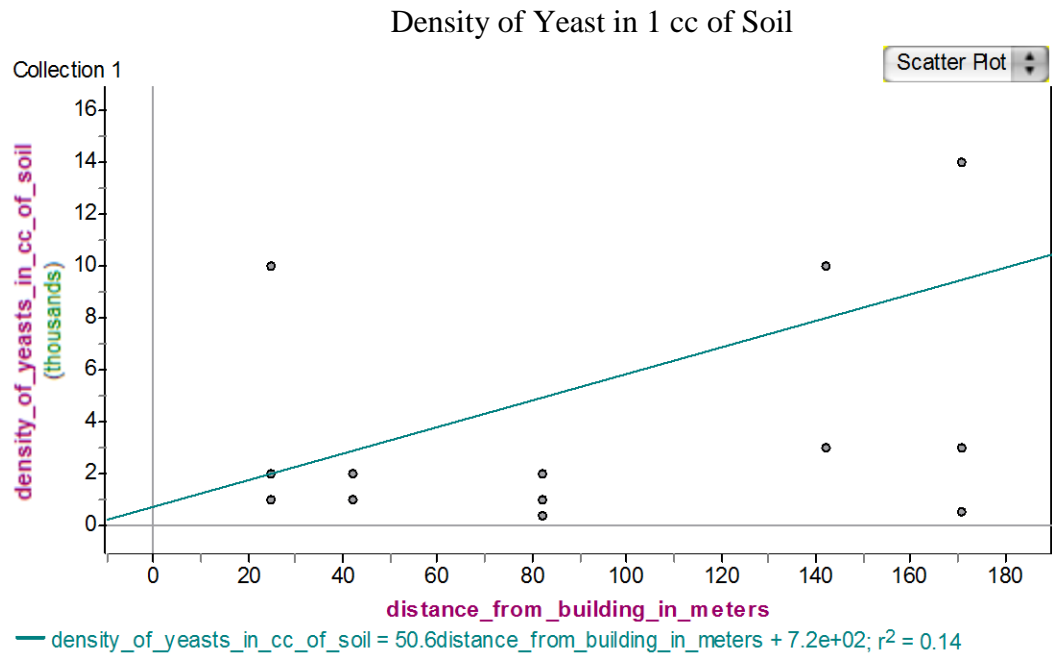
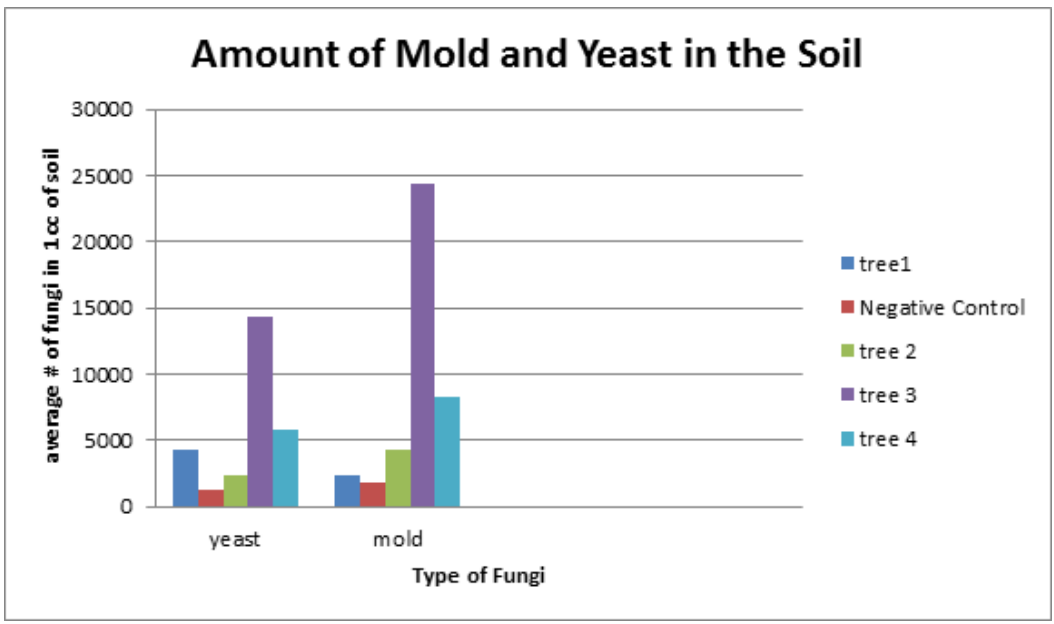
IV. Data and Analysis

a. Data Table

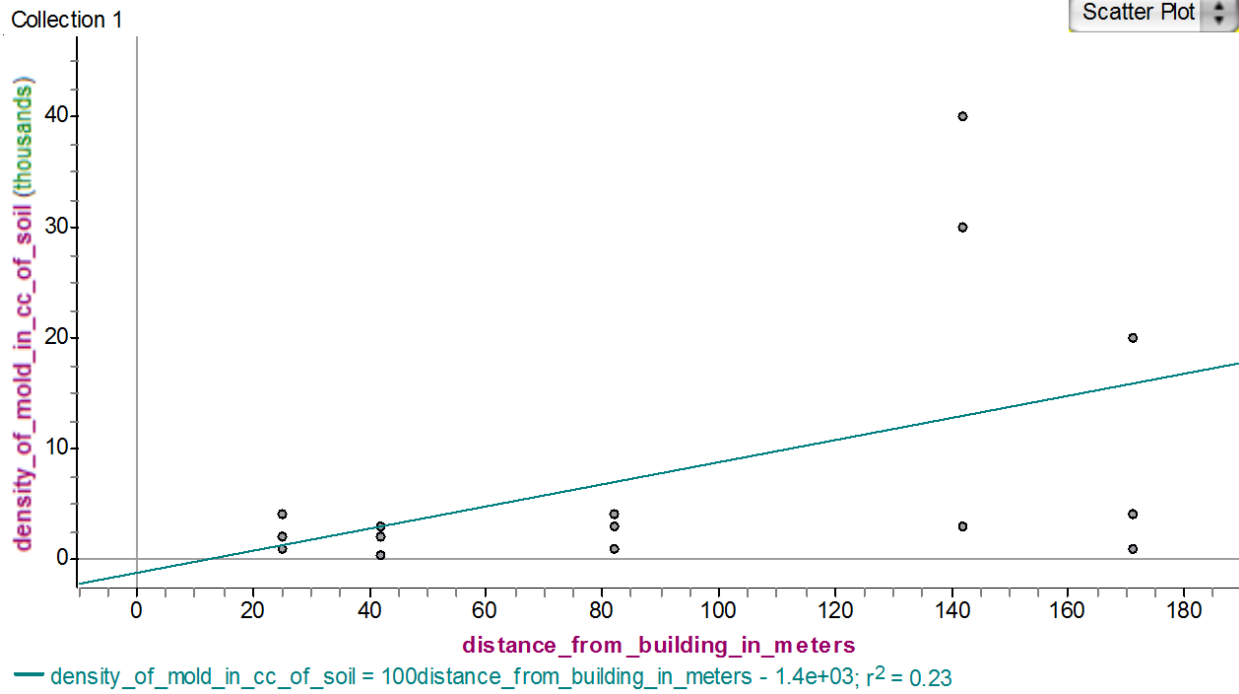
Amount of Fungus

Sample	Trial	Mold density in 1cc of soil	Average Mold density	Yeast density in 1 cc of soil	Average Yeast density	Total fungi density	Average
Greg (42.0624 m.)	1	3000	18000	2000	1333	5000	3133
II	2	400		1000		1400	
II	3	2000		1000		3000	
Sasha (24.6888 m.)	1	4000	2333	1000	4333	5000	6667
II	2	1000		10000		11000	
II	3	2000		2000		4000	
Cody (171 m.)	1	20000	8333	14000	5867	34000	14200
II	2	4000		3000		7000	
II	3	1000		600		1600	
Bertha (82.2655 m.)	1	4000	2666	400	1133	4400	3800
II	2	3000		1000		4000	
II	3	1000		2000		3000	
Zack (142 m.)	1	3000	24333	10000	14333	13000	38666
II	2	30000		3000		33000	
II	3	40000		30000		70000	

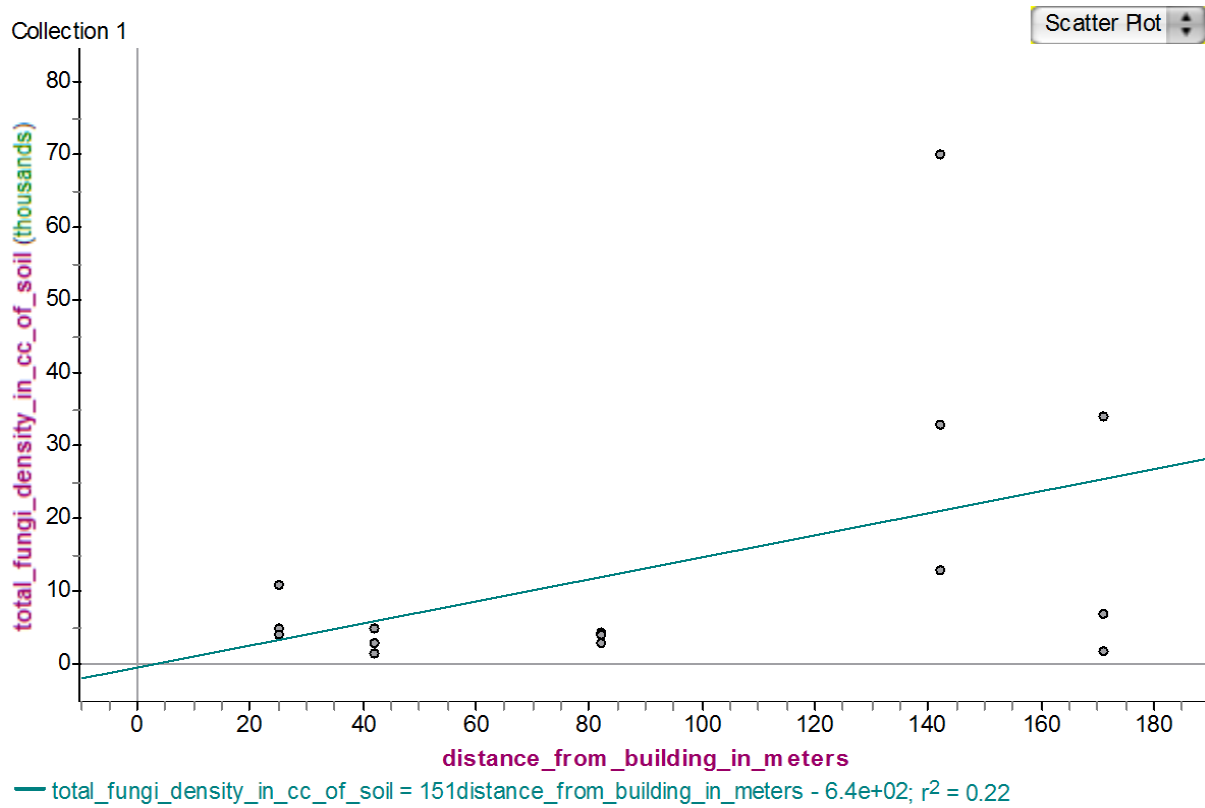
b. Graphs:



Density of Mold in 1 cc of Soil



Total Fungi Density in 1 cc of Soil



Conclusion

Our hypothesis stated that as the tree's distance from the building increase, the density of fungi increases around the tree's roots. After testing soil samples from the area around 4 trees and one sample from the front lawn, we learned that our hypothesis was correct. From each soil sample, we counted the number of yeast, mold and total fungi. Our results showed that the overall correlation for the total density of fungi was 0.22, proving that the fungi density increases as the distance from the building increases.

If soil has more yeast, it means that its environment is less suitable for fungi, and if the soil has more molds, it means that its environment is more suitable for fungi. In 1cc of tree 1's soil sample, there was a higher yeast count than mold count, showing that the soil around tree 1 was not very suitable for fungi. However, in the soil samples from the front lawn and trees 2, 3 and 4, we found a higher mold count than yeast count in 1 cc of soil. These results showed that the soil around trees 2, 3, and 4 is more suitable for fungi than the soil around tree 1.

The correlation for yeast density was 0.14, while the correlation for mold density was 0.23. Overall, this shows that the soil around the trees is suitable for fungi because there is more mold than yeast in the soil. Because of the current drought, the yeast count was generally higher than normal. Tree 3 had a higher density of both yeast and mold, showing that tree 3 has a much more suitable environment for fungi.

In our experiment, we should have taken new soil samples from each of the locations for each trial, but we did not. Instead, we used the same sample from each location for all three trials. However, the trials were done at different times, which allowed us to see the results from the soil over different amounts of time.

In the future, we could test to see what about tree 3's soil environment made it more suitable for fungi. We would look for factors that would make the environment more suitable for the fungi, if we tested the soil of tree 3.

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