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Fungi Soil Report

Background

Imagine being a helpless cell, dependent upon the sugars from a plant bigger than the cell, and fighting to survive in order to receive those necessary sugars because the sun did not shine on the leaves of the plant. Sunlight a plant recieves allows photosynthesis to occur and energy to be made by it. Then the plant gives sugar to the fungi in the ground in exchange for carbon dioxide, water, and many other essential minerals that only the fungi can supply (Nardi, 2003). The fungi then use the sugar, which is one of the five biological chemicals, and through chemical bonding, the sugar is used as energy to produce the four tasks that cells use to live. Those four tasks are reproduction, regulating temperature, synthesizing new materials, and transforming energy. This then allows the fungi to perform and live normally and send the essential minerals back to the plant. This is partly how the ecosystem works: the exhange of carbon dioxide and water for sugars between animals, plants, and minerals.

This exchange process in the ecosystem is being destroyed each day by humans who take advantage of natural resources. Soil plays such a major role in the world because it fuels the plants. The plants then provide shelter, food, and oxygen for both humans and animals. They also take the carbon dioxide out of the air so they can live. Once the soil dies and the fungi and minerals are killed away, the whole cycle becomes disrupted. This also applies to trees, if the number of trees steadily decrease, humans, animals, and the soil all suffer. The most basic level of the ecosystem consists of minerals in the soil. Without trees providing energy and sugars for the fungi and different minerals, the fungi can no longer preform the four tasks; causing the basis of our ecosystem to suffer. The ways to destory our ecosystem include uprooting trees, pollution, and allowing toxic chemicals, such as herbicides, to be put into our soil.

Fungi are very complex organisms, and can be argued as the most essential organism in the expansion of the plant world. One of the most recognizable fungi are called mycorrhizae. These mycorrhizae were first discovered by a professor; A.B. Frank in 1881 through a research project (Freedman, 2004) . He noticed that there were certain networks being created around the roots of plants. These networks were later described as mycorrhizae, which were very complex in creating a stronger bond between the plants, roots, and fungi. They are called mycorrhizae because of the latin words, myco, meaning fungus and rihae, meaning roots (Freedman, 2004).

There are also two different subdivisions of mycorrhizae; Ectomycohrrhizae and Endomycorrhizae. Ectomycorrhizae have short roots and are more visible to the naked eye. This type of myorrhizae is mostly found on forest trees and forest mushrooms. Ectomycorrhizae also increases the surface area of the root system, allowing more minerals and nutrients to be consumed. This explains why it is found on such large trees as the forest species. With the constant depletion of mycorrhizae the bond between the roots and the essential minerals and fungi will be strongly impacted (Tree Phisology, 2007). The second type of mycorrhizae is Endomycorrhizae. This is less complex and acts more like the actual mycorrhizae, but it does not create the shield around the root. Since it would cause this type to be less complex and important, the roots of

Endomycorrhizae are very specific fine roots (Tree Philology, 2007). The roots of plants are a very complex and important characteristics and help control the safty and well-being of the plant.

Fungi are also classified by the organisms they eat. The first fungi at any plant root, are the sugar fungi. These fungi eat up all of the sugars given to them by the plant. Fungi live in harsh temperatures because of the burning of the sugar. Once these sugar fungi are finished with their jobs the sac fungi are brought in. These sac fungi are much slower and less populated and will eat any dead plant remains. An important characteristic of fungi are that water and damp weather encourage reproduction. A second characteristic of fungi are that they eat dangerous organsims such as nematodes and insects before they have a chance to harm the plant or its roots (Nardi, 2003). They can be thought to act as a type of shield for the plants. Besides being on the plants roots, they are also found on wood, or animal waste. Fungi can normally be found on dead organisms and come in many shapes and forms, ususally adapting to the weather and plant conditions.

The plant roots are a very complex characteristic and are important in order to help the plant to survive. The roots can break down rocks and allow them to release minerals and create space for more roots to grow. The roots grow to such depth and width that if people were to study them in great detail, they would be amazed at their expansion. However, one study has shown that the harder the weather conditions, the deeper the roots move into the soil, which is done in order to receive the proper amount of minerals and nutrients. There are many substances in the soil that could provide the roots with nutrients. For example; algar, linchens, mosses, and bacteria could all provide the plants with the essential minerals and nutrients. However, it is shown that the relationship between fungi and plants are better than that between plants and other organisms. (Nardi, 2003). When fungi are trasmitting organisms to the plant roots it was proven that the amoutn fungi produce are two times the normal amount of minerals and nutrients. Fungi are also known for allowing humans to notice their presence. Mushrooms; normally colored, are made from the “fruiting bodies” of the fungi (Nardi, 2003). Misconceptions about fungi are mostly known because of what they are commonly seen to be inhabiting.

Everyday a new chemcial enters the earth’s ecosystem, furthur destroying our world. A way to destroy the fungi in the soil would be to apply a toxic chemical onto the soil. When people try to kill off weeds or unwanted plants they will spray an herbicide onto the plant, which then goes into the soil. That herbicide is toxic and could potentially kill off the needed fungi. However, some people are lazy decide to just kill unwanted shrubbery in order to make their lawns look better. Instead of pulling out the unwanted plant or weed, they choose to spray a toxic chemical onto the soil; not understanding the circumstances. This is one of the first steps to the destruction of the ecosystem.

Round Up™ is the most popular and strongest herbicide. However, not the fastest relief to the unwanted shrubbery. The active ingredient in Round Up™ is isopropylamine salt of glyphosate. This chemical sits within the plants roots not acting as a disturbance. However, it slowly sits and weakens the plant until about a week later it attacks and kills the plant. In the process of killing the plant it will kill of fungi (Nature’s Country Store, 2007). Garlon™ is almost the opposite of Round Up™ in the way it kills the plant. Garlon™ is a plant controlling substance. The way Garlon™ works is that once it is sprayed onto the target it immediately destroys the plant. Then, about a week later, it will allow the plant to grow back enough so that the fungi re-appear. The active ingredient in Garlon™ is triclopyr (American Bird Conservatory, and Arborchem Online, 2007). Preference of the user is normally the main distinguishing mark between Round Up™ and Garlon™.

Because of the many different types of herbicides. Each different herbicide has a specific chemical or ingredient that targets a part of the unwanted shrubbery in order to help kill the plant or weed. This allows there to be sects of herbicides without the classification of names. Chlorophenoxy acid herbicides are mixed in with fertilizer and are mostly used to control the growth of broadleaf weeds. They are toxic towards plants, helping to kill them, but before they kill the plant they "blend in" with the plants hormones. This allows the herbicide to not be noticed as easily. Triazine Herbicides are mostly used on corn plants and are not good for the soil. They cause contamination of the groundwater, hurting both the soil and even potentially humans. Although, they can sometimes be used as a soil sterilant. Organic Phospurous Herbicides are most important in killing forestry and will kill basically everything in reach or near the real threat. However, a postitive characteristic of organic phospurous herbicides is that they specifically attack the chemical glysophate, which will help in the killing of the plant and make it a smoother death. This type of herbicide is also not harmful to animals, allowing the dead plant to not harm the animal if eaten (Science Encyclopedia, Volume 3, 2007). With all the different herbicides it allows people to find the least toxic and most effective for their purpose of killing the unwanted shrubbery.

Therefore, in our group experiment we studied the affect herbicides have on the soil and the killing of the fungi. We learned through out our ninth grade biology year how cells and ecosystems work. With our curiosity we decided to look more in depth how herbicides kill important fungi in our soil everyday. By determining what everyday activities can harm the soil, we decided to research herbicides. We learned that people spray toxic chemcials onto plants, which harm the soil; destroying the ecosystem. The big picture of killing off the fungi are that they give essential minerals and plants, which provide humans with oxygen. The plant then provides the fungi with sugar in order to live using the four tasks. Since we were so curious, we decided to have nine plots of soil and test each plot three times. We separated the plots into threes; the first three were Round Up™, the second three plots were Garlon™, and the last three were water. In each of the three plots we took a before, after 1, and after 2 sample. This gave us a large range of data and a chance to really examine the probability of our problem.

Being scientists, we then wrote out a lab in order to follow proper guidelines. After we collected the soil we diluted each plot three times. Placing each soil solution onto a yeast, and mold petrifilm™ plate. This allowed us to count the number of fungi per cm^3 in the soil. We waited two days before counting the petrifilm™ plates. After doing our before samples, we waitied four more days before completing the dilution process a second time for the after 1 samples. In addition, we performed the process a third time for second dilution samples. After each dilution test, we recorded the data and then used the mathematical problem to get the final data numbers.

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Experiment Lab: Herbicide and Soil Problem

I. Problem: Which herbicide - Round Up™ or Garlon™ - kills less fungi in the soil?

II. Hypothesis: If both Round Up™ and Garlon™ are added to the soil, then Garlon™ will kill more fungi.

III. Procedure:

A. Independent Variable: Adding Round Up™ and Garlon™ to the soil

B. Dependent Variable: Number of fungi in the soil

C. Negative Control: Site were only water is sprayed onto the soil

D. List of Controlled Variables:

- day of collection of soil
- place of soil collection
- collection on same weather conditions
- other vegetation outside
- amount of herbicide
- amount of soil
- condition of soil; moist, or hard
- number of sprays of herbicide and water
- distance nozel is from ground
- vegetation within soil

- time of day of collection and use of materials on that day
- time soil sits in test tube
- number of test tubes used for dilution
- amount of soil put into each test tube for dilution
- type of nutrient (petrifilm™ yeast and mold plates) used to grow fungi
- amount of sunlight tube recieves
- amount of water each dilution tube recieves
- amount of solution put onto plates
- number of times diluting soil

E. Step-by-Step Procedure:

1) Gather all materials; one bottle of round up, one bottle of surge, water, 20 flag markers, 1 GPS, 1 soil tube, 9 plastic bags.

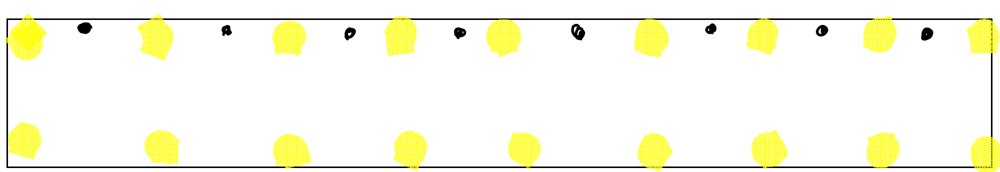
2) Go outside onto RPCS front law. (N 39.35801 / W 76.63620)

3) Place four yellow flags in a square (39 cm by 39 cm)

4) Then place yellow flags to form eight more squares of the same size yellow flags in a row next to the first (39 cm apart from the next) and see the diagram below.

5) Then on the edge of each square, place one white flag.

6) Mark the white flag with the plot number and solution (see diagram below).



7) Then take the soil core tester and take a cylinder (10 cm by 2 cm) of soil from each of the nine plots.

8) As the soil from each plot is being extracted, take a plastic bag and mark the plot number, solution and “before” and place the soil into a bag.

9) Each plot should have a different plastic baggy; labeled and sealed.

10) Then after the before sample of soil is collected wait, let the soil sit in the classroom for two days.

11) Perform a serial dilution for each of the nine plots. (make sure that all dilutions are done on the same day, for controlling purposes)

12) Get a test tube stand and place three test tubes for the first plot into the stand.

13) Write 10^0 on the first test tube, 10^{-1} on the second test tube, 10^{-2} on the third test tube, and the plot number on each of the caps of the three test tubes.

14) Then using a serilogicial pipette, place 10ml of sterile water into the first (10^0) test tube.

15) Also using the serilogicial pipette place 9ml of sterile water into the second (10^{-1}), and the third (10^{-2}) test tubes.

16) Then place 1 cc scoop of the soil of the plot being tested and place into the first test tube (10^0).

17) Shake the first test tube with soil vigiously.

18) Then using a different seriliogical pipette, place 1 ml of the (10^0) test tube into the (10^{-1}) test tube.

19) Shake the (10^{-1}) test tube with the recently added soil solution vigiously.

20) Then using the same serilogicial pipette, place 1 ml of the (10^{-1}), test tubes soil solution into the (10^{-2}) test tube.

21) Then shake the (10^{-2}) test tube and then return all three test tubes to the test tube stand.

22) Then place three yeast and mold petrifilm™ plates on the table, each marked with the plot number, “before”, dilution number, and the solution being tested.

23) Then shake the (10^{0}) test tube vigiously and using the micro pipette place 100 µl of the (10^{0}) test tube solution onto the center of the first 10^{0} petrifilm™ plate.

24) Then close the petrifilm™ plate and push down on the petrifilm™ plate with the plastic spreader.

25) Then repeat steps 22-24 with the second (10^{-1}), and third (10^{-2}) test tube soil solutions, placeing the samples on different pertriflim™ plates.

26) Once all of the petrifilm™ plates are completed, place the plates in a cabinet for two days.

28) Then after letting the plates sit for two days, take the plates out.

29) Use a magnifiying glass to then count the number of yeast and mold on each plate.

30) Look on the most diluated pertrifilm™ plate and count at least five yeast and/or mold colonies. You are looking for a blue blotch or a blue dot.

31) Then record all data and make sure to add up the number of yeast and mold for a total number of colonies.

32) Also, record the dilution number that the colonies were counted on.

33) Then use the mathematical equation of $\# \text{ of colonies} \cdot 10^{|\text{dilution}|} \cdot 10^2 = \# \text{ of organisms/cm}^3$.

34) Then record the number that is found with the mathematical equation and make it into a data table, corresponding with the plot it refers, too.

35) Then spray two squirts of Round Up™, Garlon™, and Water onto the plots as follows:

36) Plots 1-3 will have two sprays of Round Up™

37) Plots 4-6 will have two sprays of Garlon™

38) Plots 7-9 will have two sprays of Water

39) Then let the soil with the chemicals sit for four days.

40) Then recollect soil, repeating steps 7-34, but this time mark everything as AFTER; the after samples.

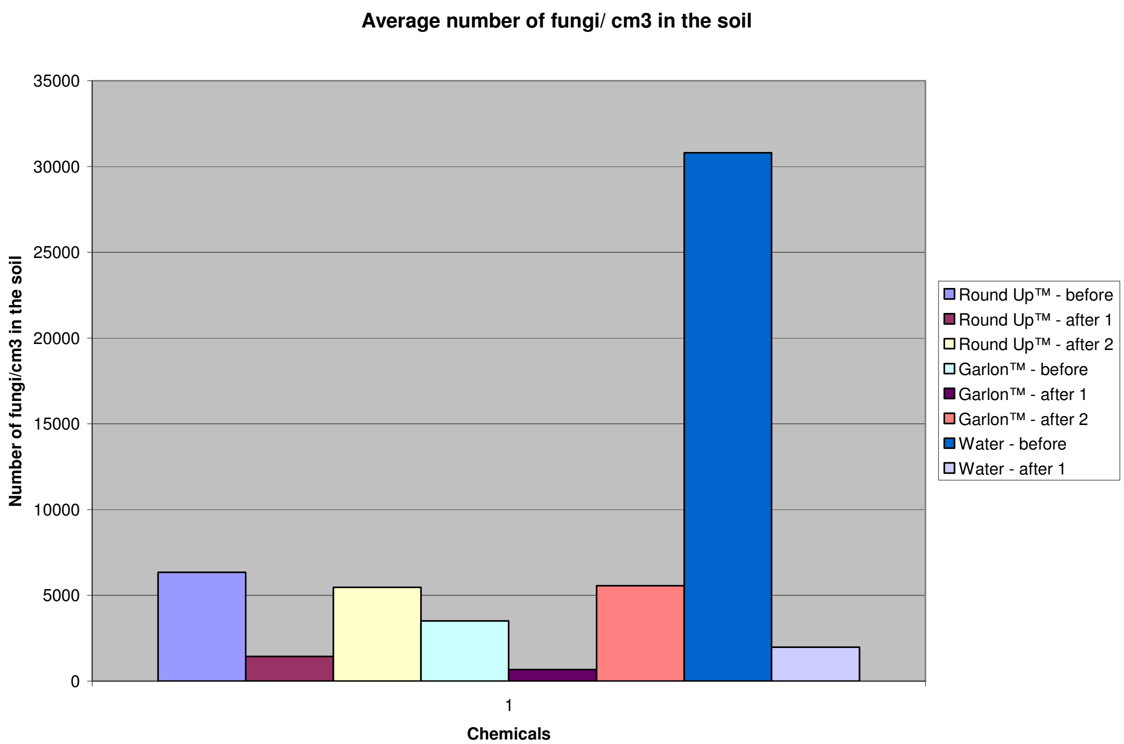
41) Wait 4 days in between collecting the first and second after samples

42) Then for our second "after" sample, repeat steps 7-34, but this time mark everyththing with "after 2" on the samples.

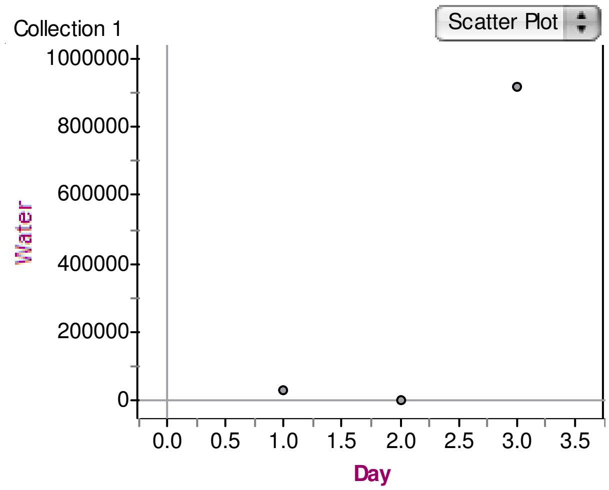
IV. Data and Analysis

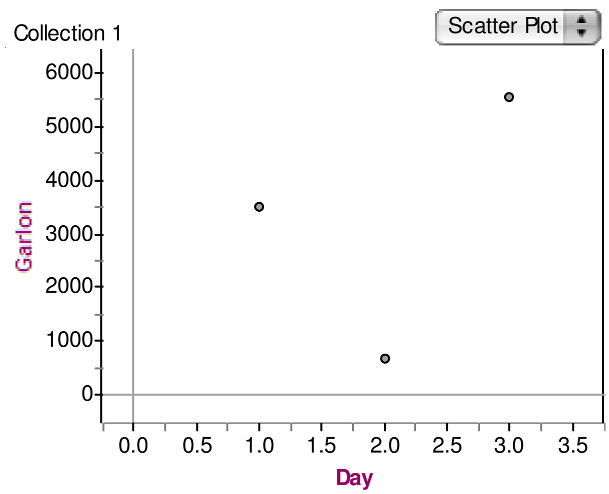
Average Number of fungi/cm^3 in the soil

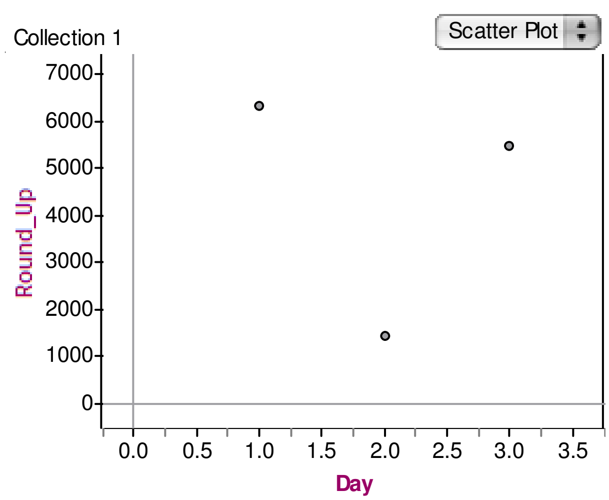
	Round Up™			Garlon™			Water		
	Before	After 1	After 2	Before	After 1	After 2	Before	After 1	After 2
Trial 1	7000	17000	1100	8000	800	9000	900	2000	9000
Trial 2	6000	1500	2300	1400	500	1700	90000	2200	18000
Trial 3	6000	1100	13000	1100	700	6000	1500	1700	500
Average	6333.33	1433.33	5466.67	3500	666.67	5566.67	30800	1966.67	916667



Not listed in the graph is the Water – after 2 plot. The number received from this plot was 916667 fungi/cm^3 in the soil. This number was approximatley 8000 times larger than the rest and threw off the proportion of the graph.


Collection 1
Scatter Plot
1000000
800000
600000
400000
200000
0
Water
0.0
0.5
1.0
1.5
2.0
2.5
3.0
3.5
Day


Collection 1
Scatter Plot
6000
5000
4000
3000
2000
1000
0
Garlon
0.0
0.5
1.0
1.5
2.0
2.5
3.0
3.5
Day


Collection 1
Scatter Plot
7000
6000
5000
4000
3000
2000
1000
0
Round_Up
0.0
0.5
1.0
1.5
2.0
2.5
3.0
3.5
Day

Analysis

The data table above clearly shows the three trials tested and the numbers received. There are nine colums because each chemical has a before, after 1, and after 2 step. The final column is an average column to average together the numbers found in each trial. In this data table, one can observe that Round Up™ takes longer to kill the fungi showing that once added, it increases the number of fungi (7000 – 17000 – 1100). Garlon™, however, seems to decrease the number of fungi immediatley and then allow them to grow back (8000 – 800 – 9000). For the negative control, water was used. Water just helped the fungi grow and reproduce. Water started with (900 – 2000 – 9000). Showing that different chemcials affect the fungi in different ways each different time period. By performing the t-test, we found the probability of the likliness of our test. This would show us the probability our data would have been the same, or similar for every time we tested the soil. For our first t-test, we got a p value of p=.55. This was while testing the before and after 1 numbers of our data table. We also tested our before and after 2 and our t-test gave us a p=.61. Both of these values show that if we had more time to continue testing the soil, our data would have been similar. This test is very important to the science world because it lets the scientists know if the amount of data is really reliable. This also helps people undersand how important testing your experiment more than three times is beneficial. Each extra time the experiment is tested, it increases the reliability of the outcomes of the experiment.

Conclusion

Our hypothesis was incorrect. We said that if both Round Up™ and Garlon™ are added to the soil, then Garlon™ will kill more fungi. This was proven wrong through our three trials of tests. We also studied the effectiveness of Round Up™ and Garlon™ in killing the fungi in the soil. We used water as our negative control because we decided that it would work perfectly since the fungi use water. The fungi also eat dead plants, give water, carbon dioxide, and other minerals to plants. We thought that the fungi would use the water as one of the five biological chemcials in production of the four tasks. In more detail; reproducing more fungi in the ground. My group found that for each of the three before plots the number of fungi were relaviley similar. Round Up™ had 7000, Garlon™ had 8000, and water had 200. Although water was significally smaller we can understand that because the soil was closly enough related that each plot had roughly the same amnount of fungi when we took our first before samples. We then sprayed our Round Up™, Garlon™, and water. However, after reading that Round Up™ takes about a week before it starts to really work, we decided to collect two after samples rather than just one. The first sample showed Round Up™ as having 1700 fungi, Garlon™ having 800 and water having 2000. This showed us that Garlon killed more fungi for the first trial. Then about four days later we tested for our second after sample. We saw that Round Up™ had 1100 fungi, Garlon™ had 9000 and water had 9000, too. This made us wonder if Garlon really did kill the most fungi. We then went back and added together the two after averages; Round Up™ - 6900 and Garlon™ - 6233.34. This shows us that Round Up™ kills more fungi in the ground overall, However, this is not a positive characteristic of Round Up™. The reason we thought Garlon™ would kill more fungi was because it worked faster so thought it might kill more fungi before Round Up™ even started working, which is true, but then Round Up™ really did lots of damage later and Garlon™ let the fungi grow back. Then after researching Garlon™ and Round Up™ in more detail we found that Garlon™ is a plant controller, which exhemplifies that Garlon™ would not kill off more fungi than Round Up™.

Besdies performing our (t-tests) my group could have structured this experiement differently. If we were asked to re-perform this experiment certain details would have been rearranged or changed. Instead of having the nine plots and grouping the three chemcials together, we should have had one Round Up™, one Garlon™, and one water in every three plots. This way we could have tested one of the "sections" with each chemical at a time, instead of cramming with all nine in one day. Making it easier for oursevles in controlling the weather conditions and the data. Also, for future groups and reference, my group should have tested close to a tree and its roots because we learned that was where the mycorrhizae and fungi are most plentiful. This would have made the fungi numbers larger. We also learned that fungi reproduce in large quantities after rain or a damp night. This would have been helpful for our soil collection in trying to collect more damp soil; hoping for more fungi to be present. All of these factors were seen in the data tables and charts above because the bars on the graph are not tall. However, in the last plot the fungi numbers were larger. Those last numbers throw off the graphs by not showing the comparision of the other plots; smaller numbers. The first one with the multiple bars on the single graph are showing overall the comparison between the plots and trials. The second three graphs are scatter plots to compare the differences between the specific trials for that chemical; before, after 1, and after 2. In the first graph there are eight bars that show the eight plots. The reason that the ninth plot is not showed is because it throws off the rest of the data because the number was about 8000 higher than the others.

Overall, our hypothesis was incorrect because Round Up™ kills more fungi because of the chemicals present and their intenstiy. Garlon™, kills more immediate fungi, but then over a period of about a week allows the fungi to reproduce and grow back. Unlike Round Up™, which lets the fungi live for a little and then in large quantities rapidly kills of more fungi.

We have acted honorably:

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