Soil Ecology Project

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May 26, 2010

We have acted honorably

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April 30, 2010
Mr. Brock
Biology 9

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Background

Bacteria found in the soil are very small and are predominantly single prokaryotic cells. They are extremely important and are necessary in order to have healthy soil because they are what complete the tasks of transforming chemicals, debasing organic materials, restraining diseases in the soil, and changing nutrients in the roots of plants (UWA, 2004). Hence the level of macronutrients and the amount of bacteria in the soil are dependent on each other, and anything that alters either one of these alters the overall health of the soil.

Two such things that can alter these are compost and artificial fertilizer. The first of these, compost, is made up of natural ingredients such as air, water, microbes, carbon and nitrogen and when broken down by soil bacteria, fungi and protozoa, slowly introduces into the soil the critical nutrients which living things need. As it decomposes, compost produces carbon dioxide, water, heat and humus (the dark organic material in soil) (Dictionary.com, 2010), and it is this humus compost is relying that adds critical chemicals such as potassium, phosphorus and nitrogen (Expert Village, 2008).

These three macronutrients are found in both compost and artificial fertilizer (as well as natural soil) and the difference is the level of these macronutrients is what strengthens or harms the natural soil. (Expert Village, 2008)

Indeed one of the many benefits of compost is that by giving essential nutrients such as nitrate, phosphorus and potassium to the soil in smaller doses at a time, the plants growing there receive the nutrients they need slowly and without overwhelming them. Plants only use a small amount of each nutrient at a time and so receiving them this way allows plants to grow more gradually, making them more stable and better able to survive. The reason the smaller doses of macronutrients are more stable for the plants growth is because the plants. The other nutrients are not wasted in this way, but instead are used by the plant individually after it is done with one (Moore Solutions Agency, 2008).

While compost and artificial fertilizer are similar in that they supply the same essential nutrients to plants, artificial fertilizer is chemically altered to give plants all the nutrients it needs at one time. This is not always a good thing as the plants can only use the nutrients they need at the moment and the rest are washed away by precipitation. The water that contains the unused nutrients can then be washed into waterways and lower ground areas and alter the natural environment negatively by adding chemicals where they are not supposed to be and contaminating things such as food and water sources. Artificial fertilizer can also cause plants to grow at an abnormal rate and make the top of the plants heavier than the roots which can lead to damage such as the plants falling over or possibly breaking.

In addition, artificial fertilizer can cause the plants to become dependent on it because the excess nutrients can harm the soil bacteria that normally produce them. One nutrient in particular that can interfere with the normal relationship between plants and bacteria is nitrogen compounds.

The way the nitrogen cycle works is that the nitrogen in the air is sucked by the bacteria into the soil. Then the bacteria in the soil turn it into lots of different forms of nitrogen such as ammonium, nitrite and nitrate. Those forms then go into the plants that are growing from the ground. The plants with the nitrogen in them are then eaten by herbivores, which are then eaten by carnivores or omnivores and continue on through the food web process. Once the organisms die they go in the ground where the bacteria decompose them and turn them back into the nitrogen forms where the cycle starts over again. The reason nitrogen is necessary is because its amino acids contain nitrogen that make up proteins. Also the nucleotides such as DNA and RNA contain nitrogen that make up nucleic acid. Without the nitrogen proteins and nucleotides are not produced, and they are necessary for cells to be made. So, without the nitrogen cells would not be made. This is why the nitrogen cycle is so important because it allows all cells to be made.

When the artificial fertilizer adds that in to the soil the bacteria ends up sitting in its own waste. As a result of this the bacteria either die or do not function to their full extent. When the bacteria aren't doing their job of creating nitrogen for the plants, the plants cannot get the nitrogen they need, and therefore rely on the fertilizer to provide the nitrogen for them. The nitrogen cycle is what takes nitrogen and converts it into usable forms. One of the main ingredients in both fertilizer and compost is nitrogen. Nitrogen is also the main component of the nitrogen cycle and the chemical that is most involved with bacteria that live in the soil. The fertilizer adds processed nitrogen in the form of nitrates to the soil whereas the compost adds in a lesser concentration of nitrogen. The processed nitrogen is more concentrated and therefore will most likely have the greatest result. (Engelstad, Orvis P., and F. C. Boswell. , 1985)

The phosphorus in the fertilizer comes from the molecules that make up the cell's membrane known as phospholipids, the ATP of the cell which is the source of energy for cells, as well as DNA and RNA. Potassium is in the ion of the cells and therefore is anywhere from 1%-2% of the plant's weight. Potassium is necessary for the plant's metabolism. Obviously, all three of the macronutrients must be present in the soil additive in order to grow the most productive organisms in the soil (Moore Solutions Agency, 2008). Nitrogen, one of the three macronutrients is vital for soil life and is classified using the nitrogen cycle.

We are trying to test how fertilizer and compost will affect the bacteria in the nitrogen cycle of the soil because we want to see if fertilizer or compost will be better for the soil. The fertilizer and compost will be added to samples of soil from the front lawn. Compost and fertilizer are both additives for soil; however they are composed differently and contribute different benefits for the soil. By testing the soil with fertilizer and compost in it we are going to observe the changes made in the nitrogen cycle. Therefore, our experiment is important because without the nitrogen cycle, no cells or living things would be made.

Our Experiment

I. Problem- How do the following applications, fertilizer and compost, alter the nitrogen cycle?

II. Hypothesis-The fertilizer will cause the most significant changes in the density of bacteria and the nitrate levels in the soil.

III. Independent Variable- what you put in the soil, fertilizer or compost

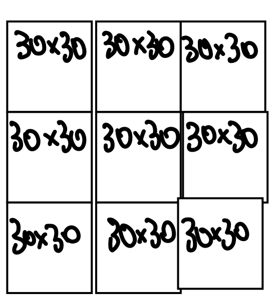
IV. Depended Variable- the density of bacteria and the amount of nitrate in the soil samples

V. Negative Control- water applied to the 3 negative control soil plots

VI. Controlled Variables- amount of each application added to the soil, time that the application sits in the soil, the amount of soil in each sample, same size jugs, the use of Sta- Green Lawn Fertilizer and the use of Plant-Tone for Organic Gardening organic fertilizer for each trial, number of samples from each plot, same kind of chemical tests, concentration of solutions for their given plots, amount of sterile water used in the bacteria test, same type of petri plates with nutrient agar, same size of soil plots- 30 cm x 30 cm, same size soil samples- 15 cm deep x 2 cm in diameter, and same units of measurement

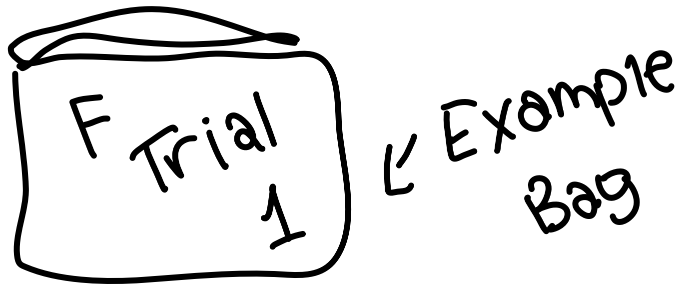
VII. Step-by-Step:

1. Go to N 39.35810^{0} and W 076.63590^{0}
2. Label 3 plots with 30 cm x 30 cm measurements for fertilizer, 3 plots with 30 x 30 measurements for compost, and 3 plots with 30 x 30 measurements for the negative control. You should have a total of 9 plots. Labels- F= Fertilizer, C= Compost, NC= Negative Control.
   Your plots should look like this:

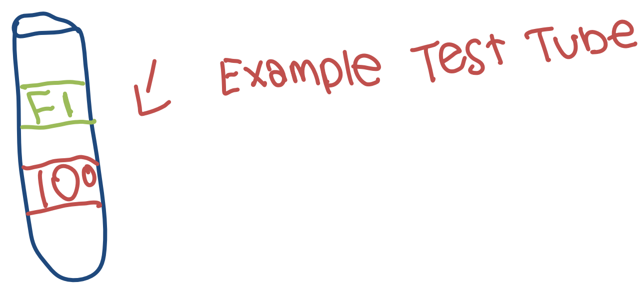


3. Use the soil auger and rubber hammer to collect **ONE** soil sample that is 15 cm deep and 2 cm in diameter from each 30x30 plot for the **"before soil samples". Make sure that you take all before samples at the exact same time.**
4. Place the sample from each plot into individual plastic baggies- be sure to label each bag with the number of the trial (1,2, or 3), and an F, C, or NC, for fertilizer, compost, or negative control, and which plot it came off of. Label the "before soil samples" with blue sharpie so you can tell them apart from the "after soil samples"

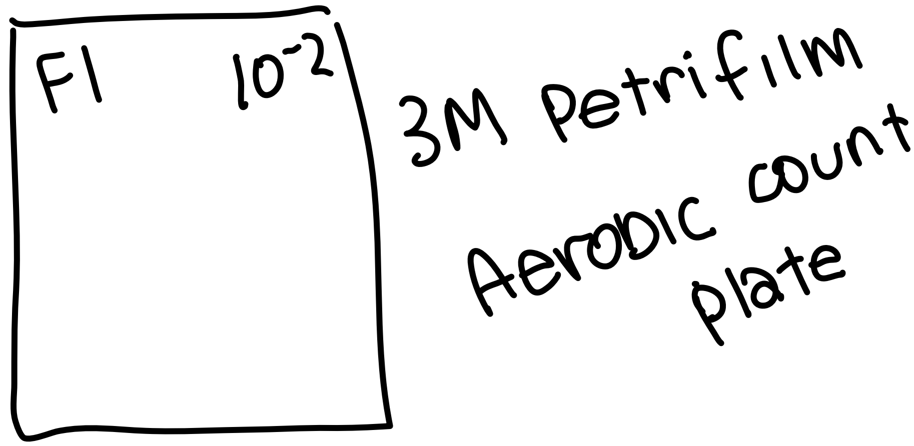
5. Make sure to clean off the soil auger when each plot is finished- you should have 9 soil samples total, 1 from each plot
6. Carry the bags back into the lab
7. Place 4.8 grams of Sta-Green Lawn Fertilizer in a clean mortar and pestle, and grind up
8. Pour into a jug containing ½ a liter of water and shake each jug to make the fertilizer application
9. Repeat steps 7-8 two more times, into separate jugs so that you have a total of 3 fertilizer solutions
10. Place 8.8 grams of Plant- Tone for Organic Gardening organic fertilizer (compost) in a clean mortar and pestle, and grind up
11. Pour into a jug containing ½ a liter of water and shake each jug to make the organic application
12. Repeat steps 10-11 two more times, into separate jugs so that you have a total of 3 compost solutions
13. Fill 3 more jugs with 1/2 liter of water each for the negative control application
14. Pour each jug of fertilizer, compost, and negative control into their corresponding correctly labeled plots. Make sure to spread the liquid throughout the plot, and not in just pour in one place
15. Let the fertilizer, compost, and water sit for 72 hours
16. Once the 72 hour period has ended use the soil auger and rubber hammer to produce **ONE** soil sample that is 15 cm deep and 2 cm in diameter from each 30x30 plot for the **"after soil samples"**. Every sample should be 15x2 cm.
17. Place the sample from each plot into individual plastic baggies- be sure to label each bag with the number of the trial (1,2, or 3), and an F, C, or NC, for fertilizer, compost, or negative control. Label the "before bags" with blue sharpie so you can tell them apart from the "after bags"



18. Make sure to clean off the soil auger when each plot is finished- you should have 9 soil samples total, 1 from each plot
19. Carry the bags back into the lab
20. **Make sure you test for the bacteria density and the nitrate levels for all 9 "Before" soil plots at the exact same time.**
21. Use the STH-14 Combination Lamotte kit in order to perform the Nitrate procedure for the "Before Soil Samples".
22. The following steps are for bacteria:
23. Label 9 sets of 4 15 ml. culture tubes for all 9 plots in the following way: label which application, fertilizer, compost, or negative control, the trial, 1, 2, or 3, and then label one with 10^0, one with 10^{-1}, one with 10^{-2}, and one with 10^{-3}



24. Use a clean, new transfer pipette to add 10 ml of sterile water to each 10^0 culture tube.
25. Use the same pipette to add 9 ml of sterile water to each 15 ml. culture 10^{-1} tube.
26. Use the same pipette to add 9 ml of sterile water to each appropriately labeled 15 ml. culture 10^{-2} and 10^{-3} tube.
27. Place 1 cc of soil sample F1 into the 10^0 culture tube, labeled "F_1 $10^{0.}$
28. Cap the tube shake vigorously
29. Using a new clean pipette, remove 1 ml. of the soil/water mixture from this appropriately labeled 10^0 tube and place into the correspondingly labeled 10^{-1} tube
30. Cap and shake vigorously
31. Using the same pipette in step 29, remove 1 ml. of the soil/water mixture from the 10^{-1} and place into the correspondingly labeled 10^{-2} tube
32. Cap and shake vigorously
33. Using the same pipette in step 31, remove 1 ml. of the soil/water mixture from the 10^{-2} tube and place into the correspondingly labeled 10^{-3} tube
34. Cap and shake vigorously
35. You should now have a total of 4 culture tubes
36. Place 100 ul samples from the 3rd and 4th tubes (10^{-2} and 10^{-3}) onto their own separate and appropriately labeled petri plates containing nutrient agar. Be sure to replace the tip each time. Use the 3M Petrifilm Aerobic Count Plates. Label the petri plates like the example:



37. Allow the bacteria to grow for 72 hours
38. Examine each of the plates for individual bacteria colonies and choose the plate with the lowest dilution value (but at least 5), to make your estimates of the number of bacteria in the original 1 cc soil sample using the following formula:

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

39. Record your results
40. **Make sure you test for the bacteria density and the nitrate levels for all 9 "After" soil plots at the exact same time.**
41. Repeat steps 22-36 for all "After" soil samples (F1, F2, F3, C1, C2, C3, NC1, NC2, and NC3)
42. Repeat step 21 (nitrate procedure) for the "After Soil Samples"

Data Tables

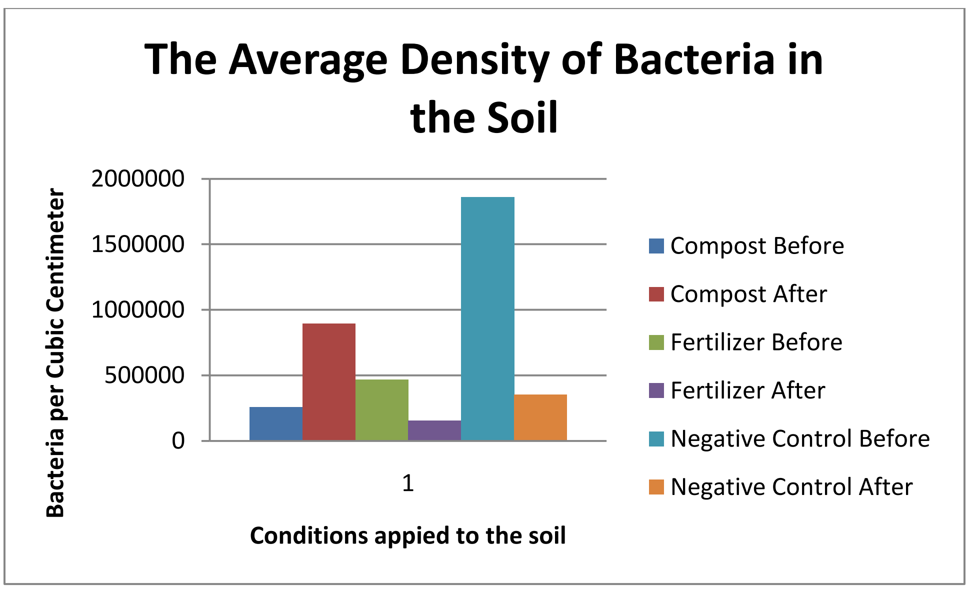
Before Treatment Applications

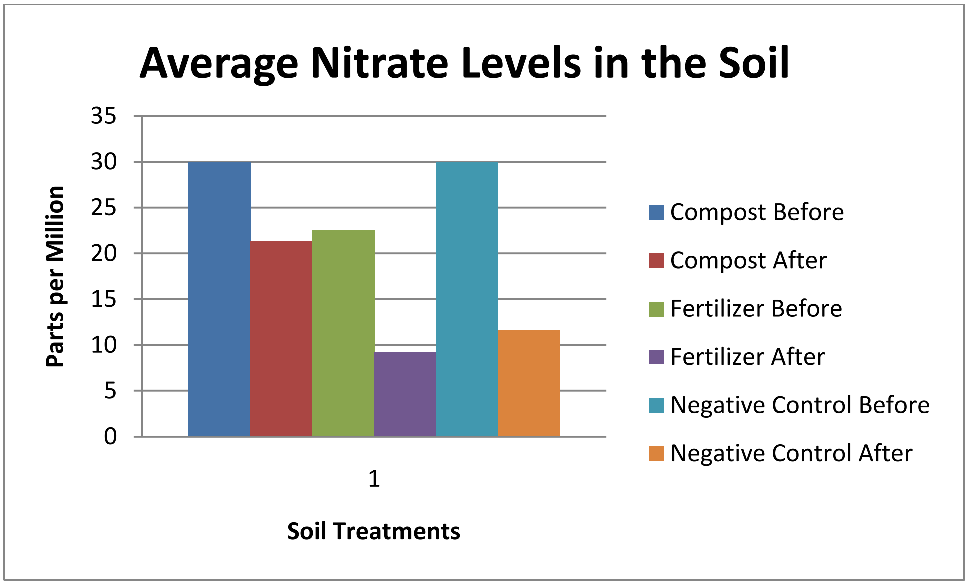
Type of Treatment	Nitrate Level (ppm)	Bacteria (#/cc soil)
Compost 1	10 ppm	270,000 cc
Compost 2	40 ppm	4,100,000 cc
Compost 3	40 ppm	1,200,000 cc
Avg.	30 ppm	256666.667 cc
Fertilizer 1	30 ppm	150,000 cc
Fertilizer 2	12.5 ppm	80,000 cc
Fertilizer 3	25 ppm	540,000 cc
Avg.	22.5 ppm	466666.69 cc
Negative Control 1	10 ppm	700,000 cc
Negative Control 2	75 ppm	410,000 cc
Negative Control 3	5 ppm	270,000 cc
Avg.	30 ppm	1856666.669 cc

After Treatment Applications

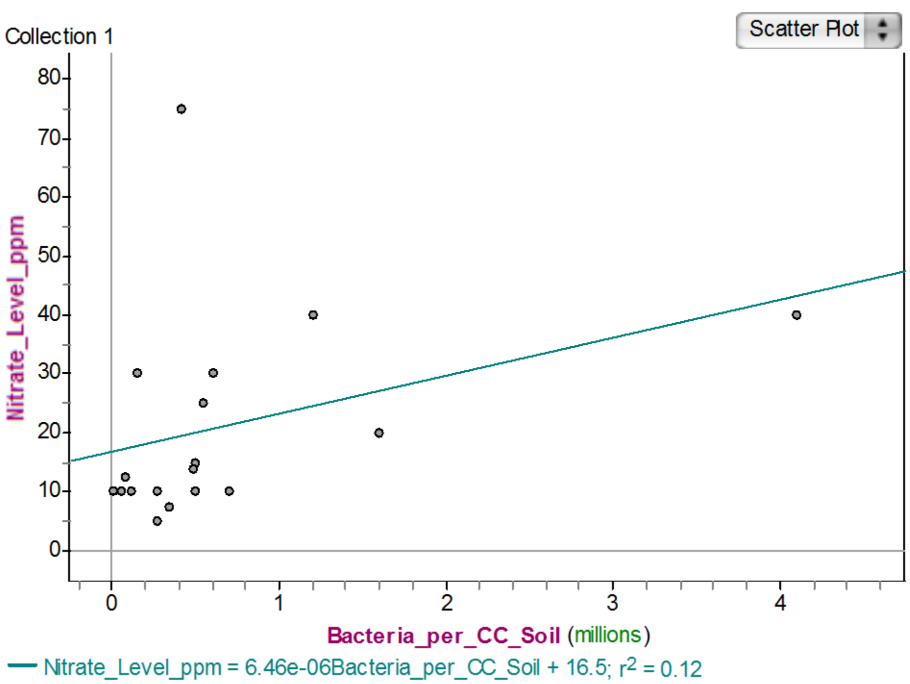
Type of Treatment	Nitrate level (ppm)	Bacteria (#/cc soil)
Compost 1	30 ppm	600,000 cc
Compost 2	20 pp,	1,600,000 cc
Compost 3	14 ppm	490,000 cc
Avg.	21.33 ppm	896666.667 cc
Fertilizer 1	10 ppm	5,000 cc
Fertilizer 2	10 ppm	120,000 cc
Fertilizer 3	7.5 ppm	340,000 cc
Avg.	9.167 ppm	155,000 cc
Negative Control 1	15 ppm	500,000 cc
Negative Control 2	10 ppm	500,000 cc
Negative Control 3	10 ppm	60,000 cc
Avg.	11.667 ppm	353333.33 cc

Data Analysis





Bacterial Density vs. Nitrate Levels in the Soil



Conclusion

Our experiment proved our hypothesis to be false. We predicted that artificial fertilizer would cause the most significant changes in the density of the bacteria and the nitrate levels in the soil. However, through our tests we found that when added to the soil, compost caused the most significant change in the density of bacteria and the nitrate levels in the soil. We found this result because in order for the density of bacteria in the soil to increase the most significantly, the amount of nitrogen in the soil must not overwhelm the bacteria in the soil. In the before treatment applications the average levels of nitrate for compost averaged 30 ppm, and the levels of bacteria averaged 2556666.667 cc. In the before treatment applications the average levels of nitrate for fertilizer averaged to 22.5 ppm, and the levels of bacteria averaged to 466666.69 cc. In the before treatment applications the average levels of nitrate in the negative control averaged 30 ppm, and the levels of bacteria averaged 1856666.669 cc. In the after treatment applications the average levels of nitrate for compost averaged 21.33 ppm, and the levels of bacteria averaged 896666.667 cc. In the after treatment applications the average levels of nitrate for fertilizer averaged 9.167 ppm, and the levels of bacteria averaged 155,000 cc. In the after treatments applications the average levels of nitrate for the negative control averaged 11.667 ppm, and the levels of bacteria averaged 353333.33.

Only one of the three nitrogen tests for compost resulted in an increase in the parts per million of the nitrogen levels, and two of those three tests resulted in a decrease of the parts per million of the nitrogen levels. All three of the nitrogen tests for artificial fertilizer resulted in a decrease in the amount of parts per million of the nitrogen levels. Two of the three nitrogen tests on our negative control plots resulted in an increase of parts per million in the nitrogen levels and one of those three resulted in a decrease of parts per million in the nitrogen levels. Two of the three tests of the amount of cubic centimeters of bacteria in the compost plots resulted in an increase in bacteria and one of the tests resulted in a decrease of the amount of cubic centimeters of bacteria in the soil. Only one of the amount of cubic centimeters of bacteria in the soil in our artificial fertilizer plots resulted in an increase of bacteria in the soil and two of the tests for the amount of bacteria in the soil with added artificial fertilizer led to a decrease of the amount of bacteria per cubic centimeter. Two of the three tests for the amount of bacteria per cubic centimeter for the negative control plots had a decrease in the amount of bacteria and just one had an increase in the amount of bacteria in the soil.

Citations:

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