Soil Ecology Project

We have acted honorably.

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Soil Ecology Background

There are several different types of soil bacteria, and each play their role in how healthy the soil is. One kind, the decomposers or saprotrophs, are bacteria that “convert energy in soil organic matter into forms useful to the rest of the organisms in the soil food web” (Brandt, Frey, Hermanson, Kealy, Ketter, Liddicoat, 2010; and Ingham, 2000). They are crucial to keeping the soil healthy because they help break down dead organisms into a form of energy plants can use for food, thereby making it available to all the other organisms in the ecosystem. In a long, multi-step process, detritivores such as burying beetles and earthworms first eat the structures and tissues of dead organisms that fall to the ground, and then their feces and other wastes leave the surface to go underground into the “hands” of the bacterial decomposers (Staff of Glow Web Services, 2010). These decomposers come in and release powerful enzymes which break down the leftover organic molecules into nutrients, such as nitrates, phosphates, and carbon dioxide that contain the key chemical elements, nitrogen and carbon that are the building blocks of all the biological molecules (Gemmell, 2008; and User of Teachers’ Domain, 2002). Once in these forms, the nutrients can be absorbed and used by plants, which then use these nutrients to make food for themselves. The decomposers, in turn, take a share of some of the nutrient compounds to provide energy for themselves, and so this entire process continues over and over again (User of Teachers’ Domain 2002; and Gemmell, 2008). Hence, decomposition is crucial because without it, everything that died would pile up over time and never disintegrate. It would just sit there forever and the biological molecules all life needs to survive would remain trapped and unavailable (Brandt, Frey, Hermanson, Kealy, Ketter, Liddicoat, 2010).

A second major type of bacteria found in the soil are the mutualists, or nitrogen-fixing bacteria. These bacteria work with plants in a symbiotic relationship, where the plant needs the bacteria to survive and vice versus (Kimball, 2010). Plants reuse these bacteria because the bacteria provide the plants with one of the necessary nutrients they need to survive, nitrogen, in the form of nitrates (NO_3^-), with which the plants make critical molecules such as amino acids (Staff of Lenntech, 1998). In turn, the plants provide the bacteria with some of the nutrients needed to survive, which typically include sugars for cellular respiration (Ingham, 2000).

The entire process is known as the Nitrogen Cycle, and it is the foundation for most of the life on this planet. In this cycle, the soil bacteria first absorb nitrogen gas from the atmosphere (N_2) and convert it to ammonium (NH_4^+) (Ritter, 2006), which can be released through decomposition (Harrison, 2003), and through nitrification, nitrifying bacteria then convert the ammonium into nitrites (NO_2^-) by combining the NH_4^+ with oxygen (Ritter, 2006). Oxidization of the nitrites into nitrates (NO_3^-) follows, and this is the form plants can most readily assimilate for their own biological needs. (Staff of Freshwater Madness, 2010 and Ritter, 2006). Then, after the plants and the organisms that eat the plants die, the decomposing bacteria break them back down into ammonium which various anaerobic bacteria convert back to nitrogen gas and the process starts all over again (Staff of Lenntech, 1998).

Nitrogen is such an important chemical element for multiple reasons. For starters, the plants and the organisms that eat them, (such as sheep, and the other carnivorous animals such as wolves who eat the sheep) use nitrogen in their cells to create nucleotides and amino acids. Cells then combine the nucleotides and amino acids to create DNA and proteins that allow cells to perform their four tasks; homeostasis, reproduction, respiration, and the synthesis of new material. By doing these four tasks every day, all the time, the organism is able to function, and so without the element nitrogen and its cycling through the ecosystem nothing would be able to function, reproduce, live, and therefore the ecosystem would be unable to grow and develop.

But, there are consequences if the nitrogen movement is disturbed. For example, the bacteria in the soil play a major role in the converting of and healthiness of nitrogen in the soil. If the bacteria in the soil are affected in a negative way then the nitrogen is affected in a negative way. This causes the whole cycle to be blown off course because then the plants are absorbing faulty nitrogen which causes them to not function properly, which causes the sheep who eat the plants and the wolves who eat the sheep to not function properly as well. It is a vicious cycle that can lead to an unhealthy environment. (Harrison, 2003; and Staff of Lenntech, 1998).

One potential group of organisms that might have a negative impact on the nitrogen movement, as described above, and therefore all organisms that live in an ecosystem, are invasive species. Because of the intimate relationship between plants and this cycle, invasive plants may have a particularly negative impact on this process. Also, invasive plants and non-invasive plants may affect the bacteria in the soil by interacting with the bacteria in the soil differently. Invasive species are potentially a major problem also to the soil communities because they will suck everything from the soil. The invasives have an extremely high supply-and-demand rate and need these nutrients desperately because they grow so quickly. So, invasive plants have the potential to form a strong relationship with the bacteria in the soil that make the nutrients the plants need. This can also be positive for the bacteria because the invasives are also supplying them with nutrients they need. However, this can be detrimental to the other surrounding plant species because the invasive plants are hijacking all the bacteria which are making all the nutrients. Looking at the negative aspects, this is a problem because the invasives are sucking up all the nutrients faster than they can be created by the microorganisms in the soil, meaning there are less nutrients for the other plants.

Not only do invasive plants steal nutrients from the soil, but they also affect certain microbes, such as bacteria within in the soil. Colorado State University, for example, has shown that soil samples collected from areas occupied by the invasive plant species, Spotted Knapweed, contain significantly different amounts of fungi in the areas with large amounts of Spotted Knapweed as opposed to in areas with no Spotted Knapweed, and these changes occurred even when a small amount of Spotted Knapweed is present (Spero News, 2007). Hence, invasives alter the ecology of the soil dramatically even when there is only a small amount present. Ultimately, this can be a terrible consequence because it can cause the soil to become unhealthy.

If the soil is healthy then the plants that use the soil will be healthy too because plants use the soil for all of things they do to keep themselves alive, such as absorbing a variety of nutrients. But, this process can be reversed, because if harmful plants are growing they can have a negative impact on the soil by absorbing too little or too many nutrients at a certain time. This can have a drastic effect on the soil because if the invasive plants are absorbing too many nutrients the microbes in the soil cannot produce that particular nutrient fast enough, so there is too little of that nutrient in the soil. But, if the invasive plants absorb too little nutrients and the microbes continue to make that nutrient, there may be too much of that nutrient into the soil which can also be harmful to the soil. But, this problem cannot only be applied to invasive plants. It is also possible that non-invasives plants could absorb too little or too many nutrients causing the microbes in the soil below to make too much or too little of a nutrient. The key is that the plants and microbes in the soil need to be in balance in order for the soil to be healthy and productive.

What kind of plant covering would cause there to be more bacteria present in the soil? A ground-covering of invasive or non-invasive plants? To test this idea an experiment was designed to take soil samples from three different areas; one, with a ground covering of invasive plants, another with a ground covering of non-invasive plants, and the third area(negative control) with a monoculture covering. Then, the samples collected would each be tested to see how many bacteria were present in one cubic centimeter of soil. It was thought that the soil with a ground covering of non-invasive plants would have the most bacteria, and therefore be the healthiest.

Experiment:

Invasive vs. Noninvasive

I. Problem: Are there more bacteria in the soil covered by invasive or by non-invasive plants?

II. Hypothesis: Soil occupied by non-invasive plants will have a denser population of bacteria living in it.

III. Procedure:

   A. Independent Variable: soil covered with invasive vs. non-invasive species
   B. Dependent Variable: the density of bacteria in each of the soil samples
   C. Negative Control: soil sample from an area which has a mono culture of native species
   D. List of Controlled Variables:
      - ✓ 3 separate samples for each area
      - ✓ Depth of soil corer (15 ½ cm.)
      - ✓ Width of soil corer (2 cm)
      - ✓ The time we gather the soil
      - ✓ The time we turn the corer clockwise (after the first minute)
      - ✓ The time we have constant (2 minutes) to dif the corer into the soil
      - ✓ 10 mL for the 10^0
      - ✓ 9 mL for the 10^{-1}, 10^{-2}, and 10^{-3} for trials 1,2, and 3 in each area (pond, east, west)
      - ✓ Constant calculations
      - ✓ Use a new pipette for each area
      - ✓ Use the same location to write down the numbers in the GPS

- ✓ same day, hour, minute, second, to collect the soil (so the weather is constant)
- ✓ 48-72 hours for bacterial growth process
- ✓ Do each dilution the same way at the same time
- ✓ Sterile water
- ✓ Always dilute the same degrees for each test tube
- ✓ Always use 1 cc of soil into the 10^0 tube
- ✓ Always use 10 mL for the 10^0
- ✓ Always use 9 mL for the 10^{-1}, 10^{-2}, and 10^{-3}

E. Step By Step Instructions:

1) Label 3 plastic bags for non-invasive area, one for each trial: NI (1), NI (2), and NI (3). Label 3 plastic bags for invasive area, one for each trial: I (1), I (2), and I (3). Label 3 plastic bags for controlled area: C (1), C (2), and C (3).
2) Collect three samples from the invasive site located at (west) N. 39. 35807^0 W. 076.63864, and three for the non-invasive area (east) N 39. 35787^0 W 076.63857, and three from the controlled side (pond) N 39.35749 W 076.63888 using the soil corer at a depth of 15 ½ cm and 2 cm in width. Place the soil samples in the corresponding plastic bags that you have labeled before.
3) Return to the lab and prepare to test the bacteria in the non-invasive, invasive, and controlled areas. Note: the first soil sample (first trial) from the non-invasive area, the first soil sample (first trial) from the invasive area, and the first soil sample (first trial) from the invasive area always need to be tested at the same time. Same for each area for each of the second and third trials.
4) Starting with the non-invasive area, put the five tubes in order according the trial number and dilution. Example: East 1 10^0, East 1 10^{-1}, East 1 10^{-2}, and East 1 10^{-3}
5) Take the lids off while on the rack. Make sure you don't switch the lids.
6) Take 10 mL of the sterile water and add to the non-invasive trial 1 10^0. For 10^{-1}, 10^{-2}, and 10^{-3}, add 9 mL of sterile water.
7) Scoop 1-cc of the non-invasive soil and put it into the (trial 1) 10^0 non-invasive test tube. If you need to, get a glass rod to push the soil through the 1-cc scooper to push the soil out.
8) Put the cap back on and hold down to shake the 10^0 test tube very thoroughly.
9) Using a new pipette, take 1 mL from tube non-invasive (1) 10^0 and put it into the non-invasive (1) 10^{-1} tube; cap and shake vigorously.

10) Using a new pipette, take 1 mL from tube non-invasive (1)10^{-1} and put it into the non-invasive (1) 10^{-2}; cap and shake vigorously.
11) Using a new pipette, take 1 mL from the non-invasive (1) 10^{-2} and put it into the non-invasive (1) 10^{-3}.
12) You should now have a total of four culture tubes.
13) Plate 100 micro liters samples from the 2nd and 3rd (dilutions 10^{-2} and 10^{-3}) onto their own separate, labeled 3M Petrifilm Aerobic Count Plate nutrient agar sheets. Make sure you use a new pipette.
14) Repeat steps 4-13 for the first trials in the controlled area and invasive area as well.
15) Wait 48-72 hours for the bacteria to appear.
16) When ready, lay the nutrient agar sheets in order to count the individual bacterial colonies with a magnifying glass.
17) Start with 10^{-3} to count. If there are less than five bacteria on the nutrient agar paper 10^{-3}, count the bacteria on the 10^{-2}.
18) If the bacteria are too small to see, then simply sharpie the dots.
19) Now use this calculation to find how many bacteria are in colonies. Microbes in 1-cc of soil = # of colonies on a sheet x 10^2 x | dilution at which the colonies are found |
20) Record the number of bacteria per cubic centimeter in each sample.
21) Repeat steps 4-20 for trials 2 and 3 of the invasive area, non-invasive area, and controlled area.

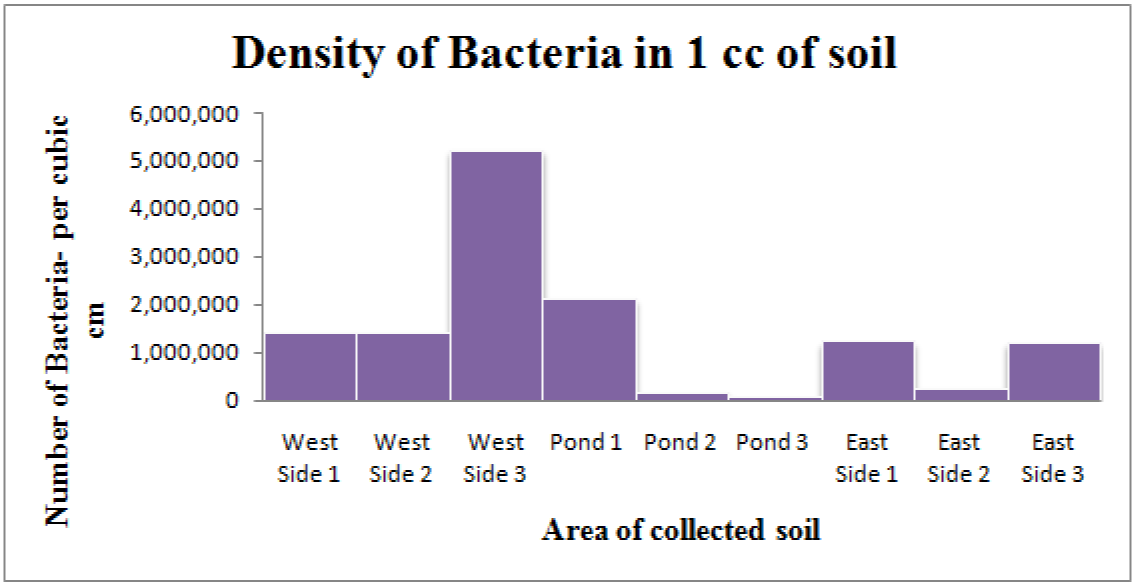
<u>Data and Analysis:</u>

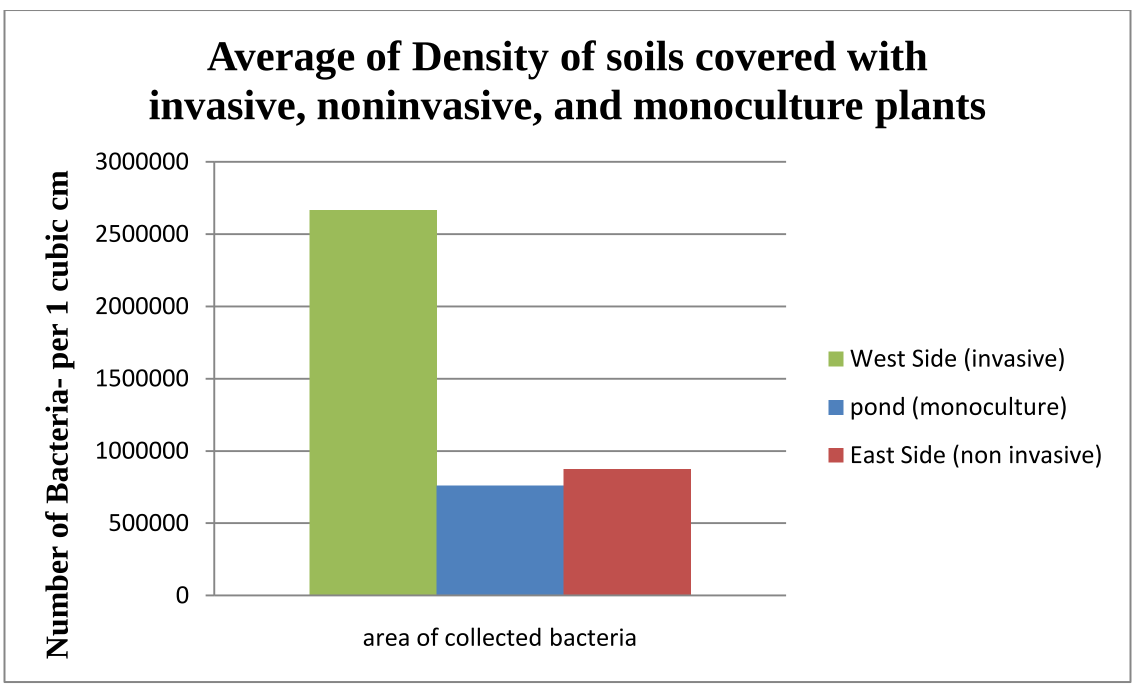
Data Tables:
Density of Bacteria in Soil Samples in the Backwoods at Roland Park Country School

Area of collected soil	**Density of Bacteria per cc of soil**
West side 1 invasive	1,400,000
West side 2 invasive	1,400,000
West side 3 invasive	5,200,000
West side average	2,666,666.667

Area of collected soil	**Density of Bacteria per cc of soil**
Pond 1 monoculture	2,100,000
Pond 2 monoculture	150,000
Pond 3 monoculture	30,000
Pond average	760,000

Area of collected soil	Density of Bacteria per cc of soil
East side 1 non invasive	1,200,000
East side 2 non invasive	230,000
East side 3 non invasive	1,190,000
East side average	873,333.3333





Conclusion:

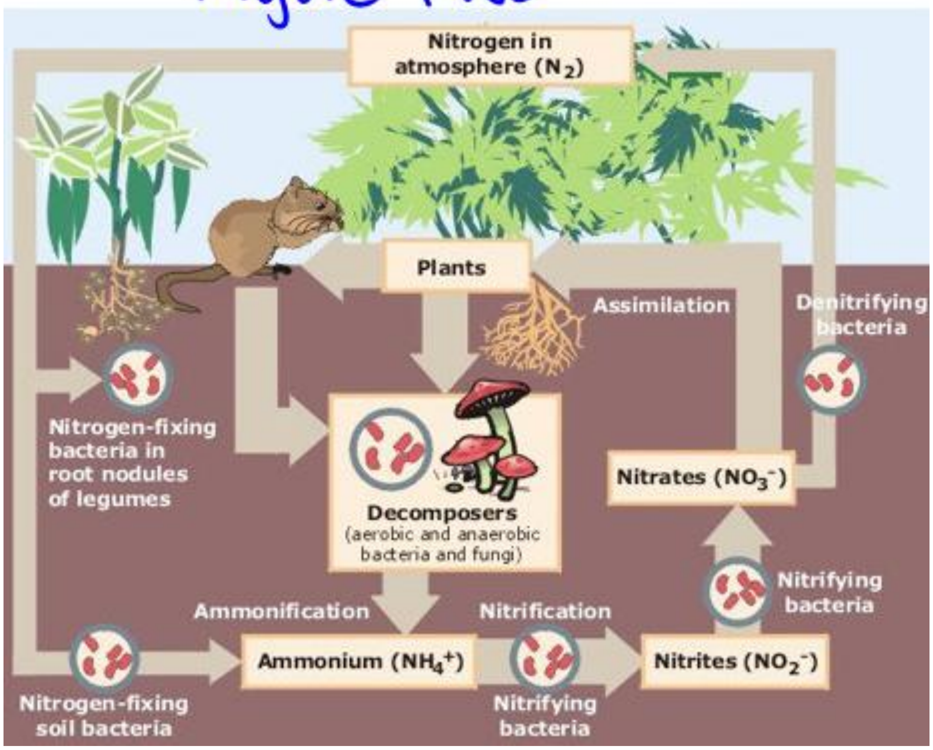
Soil Ecology Conclusion: Invasive and Noninvasive plants

My team and I understand that our hypothesis which states soil is healthier with a covering of non invasive plants is incorrect, we know this as there is much more bacteria found in the soil on the West slope side of the Roland Park Country School backwoods. The west slope is where the majority of the invasive species are found on Roland Park's campus. We believed that the soil would be healthier with a covering of noninvasive plants, because invasive plants grow quickly and aggressively (disturbing the ecosystem and the amount of bacteria in the soil and nutrients taken in by the plants), while non-invasive plants are a species that are not aggressive growers and capable of a full blown takeover. My team and I also assumed that invasive plants were bad for an ecosystem, because of their invasive nature, that they would cause havoc with the non invasive species of plants as they are disturbing the bacteria in the soil and the relationship of the native plants with the soil (ecosystem). We knew for a fact that our hypothesis was wrong when the East side (noninvasive) plant coverings had a difference of 1,793,333 more bacteria with the West side of invasive plant coverings. In comparing the East side, West side and the Pond area's amount of bacteria in soil, we found that the large difference in the numbers is due to what we believe is the diversity of the soils. To find our data, we used the Pond area as our negative control because it is a monoculture environment. Looking at our graph (Average of Density of soils Covered with invasive, noninvasive and monoculture plants), there is little difference in the amount of bacteria between the East side and Pond side slopes with only 113,333 more bacteria on average per cubic centimeter in the East Slope of the backwoods. As mentioned above, when observing the East Side and West Side densities of bacteria there is a large difference, on average 1,793,333 per cubic centimeter. This has led my group to believe that the small difference between the East side and Pond bacteria levels is due more to the fact that the East Side appears to be close to a monoculture itself with Rhododendron in its environment(Average of Density of soils Covered with invasive, noninvasive and monoculture plants). As seen in graph one, the East and West sides are significantly different in plant diversity, and they are hard to compare in "soil health" depending on the number of bacteria, because the diversity in their plant species. This data shows that having biodiversity in the Roland Park backwoods is beneficial to the ecosystem.

Diversity in an ecosystem, or biodiversity, like the West Side slope of the Roland Park out backwoods is a healthy environment for the plants and all the microorganisms in the soil! Biodiversity is important, because different plant species means a greater variety of crops (Shah, 2009.) More plants (and a greater diversity of those plants) mean much more nutrients are used in the Nitrogen Cycle. In the Nitrogen cycle, bacteria produce waste products that are stored by plants. If more plants are taking nutrients in, then there are fewer waste products that the bacteria are living in. With more variety comes more responsibility as plants depend on each other like the Nitrogen cycle for all in an Ecosystem to survive (Figure two: Nitrogen in Atmosphere) (Shah, 2009).

For future research options on our project, we would want to focus on whether the diversity of plant species had an effect on the number of bacteria found in the grounds of the East Slope, West Slope and Pond area in the Roland Park backwoods. Next time, my group and I would find more areas in the backwoods, with a more diverse ecosystem in those areas where we would test the density of bacteria. My group could also choose to test for the nutrients in the soil as invasive plants (when invading a prior ecosystem) can change nutrients in that soil. For example, there are some of the Nitrogen compounds in the soil. For further research of this project, we would test for nutrients and the density of bacteria in the Roland Park Country School Backwoods.

Figure Two



<u>Background and Conclusion Citations in APA style Refrences:</u>

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URL of an image used in the conclusion: See Shah, A reference for full citation:
http://www.globalissues.org/article/170/why-is-biodiversity-important-who-cares#WhyisBiodiversityImportant