Compaction and its Effects on Percent Moisture Content and Bacteria Density

By Morgan Alexander, Shannon Quinn, and Maddy Wilson

I have completed this assignment honorably.
Bio Final Lab

Background

In addition to the organisms commonly associated with soil such as earthworms and other organisms that can be seen with the naked eye, there are many different microscopic organisms that inhabit the ground as well. These microbes help supply key soil nutrients which plants use to grow by aiding in the decompositional and biogeochemical cycles, and some of the most significant of these microbes are the soil bacteria (Zucker 2011). Simple prokaryotes, these organisms do a number of important tasks in the soil ecosystem, including causing organisms to decay and release their nutrients back into the soil, forming soil particles that bind to each other more strongly to improve soil texture, and increasing access to water and oxygen for all the organisms living there (Duiker 2017).

Soil bacteria especially aid in the biogeochemical cycles, including the nitrogen and carbon cycles. In the first of these, nitrogen-fixing bacteria in the soil form nodules on plant roots and convert nitrogen gas from the air into a form of nitrogen, ammonium, that plants can use for their metabolic purposes. Then, other nitrifying bacteria change any excess ammonium to nitrate, a form more easily used by specific types of plants such as grasses and row crops. Finally, denitrifying bacteria convert any excess nitrate back into nitrogen gas or nitrous oxide, which enters the cycle again (Ingham n.d.).

In addition to cycling nitrogen in the ecosystem, bacteria also cycle carbon. Bacteria called actinomycetes are responsible for decomposing organic compounds that are hard to break down, such as cellulose and chitin, so that these major sources of carbon can be released as carbon dioxide for the plants to use as a nutrient as well (Ingham n.d.).

Overall, bacteria play a role in 18-90% of all processes in soil that make nutrients more easily available to other living things (Zucker 2011), and without these bacteria, plants would be unable to access the raw materials they need for photosynthesis. Plants use photosynthesis to
produce the glucose they need to feed themselves, and without bacteria’s role in the carbon and nitrogen cycles, nitrogen in particular would not be available for plants to make necessary chemicals, such as amino and nucleic acids, which plant cells need to engage in this photosynthesis (as well as complete all the necessary tasks of life: reproduction, regulation, synthesis, and respiration). Furthermore, because plants are producers, the excess energy they manufacture through photosynthesis travels through every trophic level, from themselves to primary and secondary consumers, etc. Hence, if the plants are unable to access the necessary carbon and nitrogen provided by bacteria to produce energy, then this energy will be lost to the entire food chain. This heavy impact that bacteria have on the whole of the natural world is the reason they need to be protected from certain outside forces, especially human-generated ones such as compaction.

Compaction is the process in which the weight of human-created structures, such as a building or a sidewalk, or other natural forces push down on soil, decreasing the presence of pores and other open spaces in it (Dictionary.com 2017). Compaction can greatly affect the amount of moisture in the soil (Daum 2017) and can also damage soil structure by making the size of the pores in the soil too small to allow the exchange of gases involved in the nitrogen, oxygen, and carbon cycles. Since soil structure determines the ability of soil to hold water, nutrients, and the air that is necessary for plant growth, damaging it through compaction forces plant roots to exert more force to extend into the compacted layers and can cause a decrease in nitrification. Compaction can also cause potassium and nitrogen deficiencies, an increase in the risk of crop diseases, and can increase bulk density in the soil, consequently causing root growth restriction (DeJong-Hughes, Moncrief, Swan 2001). This increase in bulk density can also cause decreased habitat for larger microorganisms, such as nematodes (Duiker 2017).

Hence, in general, compaction slows the biological activity of the microbes living in the soil. It leads to decreased habitat for the nematodes that eat the bacteria, allowing the bacterial population to grow too large, thereby decreasing the microbial diversity that normally helps keep
dangerous organisms under control by making the balance between predators and prey more even. Slower water percolation due to compaction also leads to longer periods of higher saturation, causing anaerobic soil bacteria to use nitrate instead of oxygen for their metabolism, and denitrification occurs more often, removing from the soil more of the nitrogen plants need. Other anaerobic bacteria also release hydrogen sulfide due to this compaction, which is poisonous to plants (Duiker 2017). Therefore, because of the effects of compaction, it is likely that the bacterial population will decrease.

In our experiment, we would like to examine the effects of compaction by a concrete sidewalk on the level of moisture in the soil next to it. Based on our research, we have found that soil that is closer to a site of compaction, such as by a sidewalk, should have a lower moisture level at the time of sampling than that of the soil that is further from the compaction site. This is due to the compression of the pores in the soil by the source of compaction, causing them to be unable to hold as much water as unaffected soil. We will also be extracting the active bacteria from the soil samples, and observing how this variation in moisture level due to compaction affects the density of microorganisms, specifically bacteria, in the soils from each location. Our experiment is looking to prove and display how humans have made a negative impact on the health of soil and its inhabitants by excessively using heavy equipment and developing over this soil, causing compaction (DeJong-Hughes, Moncrief, Swan 2001). It is important to recognize the significance of bacteria and other organisms that live in the soil, because of how necessary they are in the biogeochemical cycles and the food chain, and to become aware of how our actions are affecting them, and in turn, affecting us.
References

https://www.youtube.com/watch?v=gSY-4AtqQG0


http://www.extension.umn.edu/agriculture/soils/tillage/soil-compaction/

http://extension.psu.edu/plants/crops/soil-management/soil-compaction/effects-of-soil-compaction


Lab Report

I. Problem: Does compaction increase or decrease soil moisture level and density of bacteria life?

II. Hypothesis: Heavy compaction decreases the moisture level in the soil, as well as the density of bacterial life in it.
III. Procedure

Independent Variable: The degree the sampled soil has been compacted

Dependent Variable: The density of bacteria in the soil (number per cc), amount of moisture in soil (% water loss)

Negative Control: Soil sampled from an area without any compaction

Controlled Variables: Distance between compaction sites, unit of measurement, day and time soil sample is collected, coordinates of locations, temperature of oven (in Celsius), size of pipette, type of GPS device, amount of soil collected, type of soil core extractor, size of soil core extractor, plant life on soil, size of culture tubes, type of weighing device (in grams), size of container used to dry soil, pipette used for sterile water, pipette used for each soil sample for each serial dilution, amount of nutrient agar, type of nutrient agar plate, time allotted for bacteria to grow, time allotted for soil to dry, temperature of growth environment of bacteria, amount of soil used in serial dilution, amount of sterile water used, dilutions observed for bacteria growth, amount of soil solution plated, degree soil is diluted to

Step by Step

1. Go to coordinates N 39.35899, W 076.63636 and place a small marker flag in the ground 10 cm straight into the grass from the sidewalk and label it A1.

2. Place one small marker flag on both sides of this marker flag so that they are 85 cm away from it and 10 cm away from the sidewalk and label them A2 and A3, respectively. (See diagram)
3. Place three more small marker flags northeast 4m away from these three marker flags and label them B1, B2, and B3. (See diagram)

4. Place another small marker flag at the coordinates N 39.35804, W 076.63599 and label it C.

5. Wait 48 hours.

6. Using a 2 cm in diameter soil core extractor, take one soil core sample at each of the flags by inserting the soil core extractor into the soil and rotating until the top of the soil is at the 10cm mark on the extractor. All samples should be taken at 9:43am on the same day and placed each in individual airtight correspondingly labeled plastic bags.

7. Bring them inside to a lab bench.

8. Wait 48 hours.

9. Do steps 10-22 on the same day at the same time.

10. Use a clean, new transfer pipette to add 10 mL of sterile water to a 15mL culture tube. Label the tube “10 A1”.

11. Use the same pipette to add 9 mL to a second 15 mL culture tube. Label the tube “10 A1”.

12. Repeat step 11 two more times to 2 additional 15 mL culture tubes, and label the culture tubes “10 A1” and “10 A1”, respectively.

13. Place 1 cc of soil sample A1 in the “10 A1” culture tube.
14. Cap the tube and shake vigorously.

15. Using a new, clean pipette, remove 1 mL of the soil/water mixture from the “10-A1” tube and place it in the “10-'A1” tube.

16. Cap the tube and shake vigorously.

17. Using the same pipette in step 15, remove 1 mL of the soil/water mixture from the “10-'A1” tube and place it in the “10-'A1” tube.

18. Cap and shake vigorously.

19. Using the same pipette in step 15, remove 1 mL of the soil/water mixture from the “10-'A1” tube and place it in the “10-'A1” tube.

20. Cap and shake vigorously.

21. Place 100 microliter samples from the “10-'A1” and “10-'A1” tubes onto their own separate, correspondingly labeled 3M Petrifilm™ Aerobic Count Plates.

22. Repeat steps 10-21 with each of the soil samples, labeling the tubes “A2, A3, B1, B2, B3, and C”, along with the dilution number, respectively and using a new pipette for each soil sample.

23. Allow to grow for at least 48 hours.

24. Starting with the lowest dilution, examine this plate for individual bacterial colonies and if it has at least 5 colonies, use it to make estimates of the number of bacteria in the original 1 cc soil sample. Move up to the next lowest dilution to count if there are not at least 5 present. Record the number of colonies and the dilutions they were found growing on. Use the following formula: # Microbes in 1 cc of soil = Colonies on sheet x 10^x 10\[^{|dilution \# at which colonies were found|}\].

25. Do steps 26-27 on the same day.

26. Take each remaining soil sample and determine the mass of the test samples and record this mass as the “wet mass”.

27. Dry to constant mass at 110 ± 5°C for at least 10 hours. To reduce drying time break lumps into small fragments and spread in a thin layer over bottom of container.
28. Remove the sample from the drying device and cool to room temperature.
29. Determine the mass of the test sample and record this weight as the “dry mass”.
30. Determine the percent change in moisture using these equation: Mass of water in sample = wet mass minus dry mass; Percent moisture = Mass of Water/Dry mass of sample x 100
31. Repeat steps 6-30 two more times at the same time of day each time approximately every other day.

IV. Data and Analysis

a. Data Table: Impact of Compaction on Water Content & Bacteria Density in the Soil

<table>
<thead>
<tr>
<th></th>
<th>Location A: Site of Compaction</th>
</tr>
</thead>
<tbody>
<tr>
<td>% Water in Soil</td>
<td></td>
</tr>
<tr>
<td></td>
<td>% Water in Soil</td>
</tr>
<tr>
<td></td>
<td>A1</td>
</tr>
<tr>
<td>Trial</td>
<td>A1</td>
</tr>
<tr>
<td>1</td>
<td>9.27%</td>
</tr>
<tr>
<td>2</td>
<td>8.3%</td>
</tr>
<tr>
<td>3</td>
<td>8.61%</td>
</tr>
<tr>
<td>Average</td>
<td>8.73%</td>
</tr>
</tbody>
</table>
### Location B: 4m from Site of Compaction

<table>
<thead>
<tr>
<th>% Water in Soil</th>
<th>Bacteria Density (# per cc of soil)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Trial</td>
<td>Sample Number</td>
</tr>
<tr>
<td>1</td>
<td>24.44%</td>
</tr>
<tr>
<td>2</td>
<td>19.95%</td>
</tr>
<tr>
<td>3</td>
<td>16.74%</td>
</tr>
<tr>
<td>Average</td>
<td>20.38%</td>
</tr>
</tbody>
</table>

### Location C (negative control)

<table>
<thead>
<tr>
<th>% Water in Soil</th>
<th>Bacteria Density</th>
</tr>
</thead>
<tbody>
<tr>
<td>Trial</td>
<td>Sample Number</td>
</tr>
<tr>
<td></td>
<td>C</td>
</tr>
<tr>
<td>1</td>
<td>19.81%</td>
</tr>
<tr>
<td>2</td>
<td>18.14%</td>
</tr>
<tr>
<td>3</td>
<td>17.93%</td>
</tr>
<tr>
<td>Average</td>
<td>18.63%</td>
</tr>
</tbody>
</table>
Conclusion

Based on the data found by measuring the percent water in samples from both compacted and uncompacted sites, we can see that the expected relationship between compaction and the percent water content in soil was present: the moisture content was lower at the site of compaction and higher as the distance from it increased. At locations A1, A2, and A3, which were directly adjacent to a site of compaction, a sidewalk, the moisture content of the samples were 8.73%, 11.42%, and 13.01%, respectively. These percentages are significantly smaller than those from the site that was 4 meters away from the sidewalk, locations B1, B2, and B3, which were 20.38%, 18.597%, and 17.68%, respectively. The decrease in percent moisture content in the compacted soil is likely due to the pores in the soil being minimized by the heavy impact of the method of compaction, in this case the sidewalk, therefore causing there to be less space for water in the soil, causing decreased moisture content. Therefore, because the level of moisture was lower in the soil samples taken from the locations that were compacted and higher in those that were farther from the site of compaction, it can be
concluded that compaction causes a decrease in soil moisture level, which proves that our hypothesis could be correct.

However, in our hypothesis, we stated that compaction and its effects on the moisture levels in the soil would decrease the number of bacteria per cc of soil, or bacteria density, but, based on our data, this is not the case. As shown on the trend line of the scatter plot that compares percent water content to bacteria density, we can see that, since the slope of the trendline is negative (-715510), as the percent water content increases, the bacteria density decreases. This is the exact opposite of what our prediction was, and it directly opposes the natural relationship between bacteria and moisture: when there is less moisture in the soil, there should be fewer soil bacteria, and vice versa, because bacteria, as do all organisms, need water to survive. Therefore, as seen on the scatter plot, since the bacteria density was higher at the sites of compaction (where there was less moisture), and lower at those that were not, we can conclude that compaction, in fact, had no impact on the bacteria density in the soil, and that there was instead an outside force that affected the growth of bacteria in these specific areas, making our hypothesis incorrect, in a way. However, we cannot make any claims about our hypothesis because of this known relationship between bacterial density and moisture content.

This unexpected data set could be due to the fact that, as seen in previous experiments, the relationship between bacterial density and water content, when depicted on a graph, is represented by a parabolic shape, meaning that it is shown to be increasing and decreasing at different percent water contents. It is possible that we chose points that were best represented by the negative sloped section of the bacterial density vs. water content parabola. It is also possible that, since the sections of the lawn where we chose to place our sites were essentially surrounded by sidewalk, water runoff from rainfall may have rolled from the sidewalk, an impervious surface, onto the lawn and changed the water saturation of the soil in the lawn. This would mean that compaction was not the only force that impacted the moisture content, and consequently the bacterial density, in the soil.
One way we could examine the causes for this inconsistency in data would be to do more tests in different places around the front lawn, where our original sites were. This would confirm whether we just happened to pick a spot on the lawn that was best represented by the negatively sloped section of the parabola, or if the location of the sample site in regard to the surrounding sidewalk and its runoff instead affected the bacteria density. If we took samples from different places around the lawn, and the data was inconsistent, with compaction and placement in regard to the sidewalk seemingly not having an effect on bacteria density, we could infer that we simply chose a location where the data was best represented on the parabola as a negative slope. However, if we took samples based on proximity to the sidewalk, and observed that the samples that were in a location where runoff from the sidewalk could easily reach and permeate the soil had more bacteria, then we could conclude that this, in fact, was the reason our data had an unexpected result.