



Problem Statement: Climate change has brought with it so many agricultural concerns that farmers stay constantly worried about their crops. Issues such as increased droughts and therein water scarcity, favorable pest growth conditions, and decreasing soil health have permeated the globe.

Abstract

Crop rotation is an agricultural practice where species of plants are rotated out seasonally to improve the growth of the plants. Plants adapt to different soil conditions through biotic interactions (4). An increased crop diversity leads to improved soil health (5). We are trying to see the specific strains of bacteria that are altered based on the rotation of crops. One key factor in these improvements is the role of soil bacteria. However, there is still much to learn about the specific bacterial strains affected and how their presence directly influences soil quality. The purpose of this study is to investigate the impact of crop rotation on soil bacterial populations and their correlation with soil health. Crop rotation has been studied previously, however there is more research done on the benefits for agricultural, environmental sciences, and profitability. We want to focus on the community interactions within the soil, particularly how bacterial populations respond to crop rotation patterns. Understanding these microbial interactions will provide insight into soil ecosystems and improve sustainable agricultural practices.

Background

Climate change's effects are multifaceted in the agricultural world. Regenerative agriculture combats this but the practice still hinges on weather. Crop rotation is a farming practice feeling climate change's effect.

Plants mold to different soil conditions through biotic interactions during crop rotation. Large scale quantifications of crop rotation's benefits have been conducted, but little research has been done on a microbial side. There is a dear symbiotic link between the benefits of crop rotation and the soil microbiome.

This study documents the different microbial interactions within the soil before, during, and after crop rotation, so that we can infer why these levels increase and by what specific bacterial genera. Cataloging this bacterium into a website or infographic, this study will help to fulfill the isolate discovery pillar of the engineering process for crop rotation.

The information discovered could later be used to fulfill the remaining pillars and manipulate bacterial levels within the soil. The end goal is for crop rotation to retain some of its benefits for both the plant, and the general health of the rotated soil as we continue to battle the changing environment.

Materials

Materials:
2 Large Circular
Containers
2 Small Circular
Containers
Four Cloth Wicks
Soldering Iron
NPK Water
(previously made)
Deionized Water
Soil
Turnip Seeds
Radish Seeds

Materials:
Zymo Bead Beating
Kit
Nanopore
Sequencing Kit
Qubit
CP: Control Plant
P1: First Turnip Plant
P2: First Radish Plant
P3: Second Turnip Plant
P4: Second Radish Plant

Materials:
PPE (Gloves, Coats)

Method and Process Steps

NPK Water Creation

We first made our NPK water by combining roughly 2999 parts deionized water with 1-part NPK through an initial 0.5 – 1 oz NPK to 1 gal dilution and then combining. The mixture is composed of 100% Nitrogen (1% Ammonium, 2% Urea), 100% Available Phosphate, 100% Soluble Potash, & 60% Chelated Iron.

Soil Prep and Planting

We used pre-irrigated and nutrient treated soil with 0.004% Ammonium Nitrogen, 0.004% Other water soluble nitrogen, 0.18% Water insoluble Nitrogen, 0.04% Available Phosphate, and 0.04% Soluble Potash. The Endomycorrhizal species (all Glomus 0.05 propagules) are as follows: in tridens, mossae, agg. egyptum, etunicatum, 0.1% Wetting agent is also added. We made self-watering planters w/ protocol and used lab provided growth lights.

Measuring

We took note of leaf growth, root growth (limited), and the pre-flowering times for the plants. We were limited to business day rather than by-the-hour.

DNA Extraction

We used the Zymo Reagent Bead Beating Kit TM to conduct bead beating of 0.25g of our soil and genomic lysis buffer. We used Zymo Biomics Pillbug instructions for both bead and DNA extraction. Our samples were extracted on day 1, day 2, and day 3, and we repeated this process three separate times for the three stages.

Sequencing

We use the Nanopore Minion and Flongle kits along with our classmates to sequence our soil. Our reads revealed many different bacterial genera, but some showing significantly larger abundances. We investigated those with the highest and the lowest reads for all three stages of crop rotation.

bioinformatics

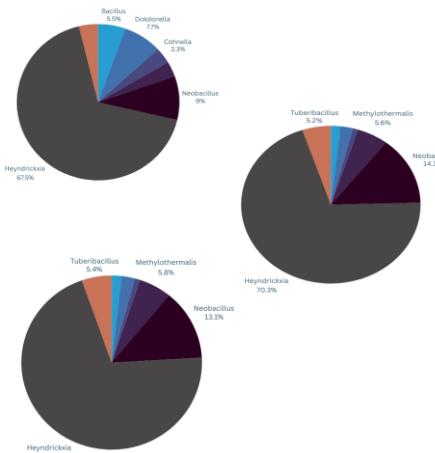
We took highlighted DNA samples and researched their effects on either crop or soil, with the goal of comparing the percentages and ratios of each stage to draw a conclusion.

Research Question: Will the bacteria and fungi that create a microbiome when a plant is grown interact and affect the growth of an alternate plant placed within the same soil?

Hypothesis or Criteria for Success

The information discovered could later be used to fulfill the remaining pillars and manipulate bacterial levels within the soil. The end goal is for crop rotation to retain some of its benefits for both the plant, and the general health of the rotated soil as we continue to battle the changing environment.

Results



We found that the soil profiles of the rotated bacteria were different from the pre-rotated profiles, but only slightly. The most significant changes were to the Neobacillus and Heyndrickxia profiles, with the rest of the genera having relatively the same percentages. Our first radish plant did not give us a read on anything except for the Pseudomonas.

*Note that we had to restart the first phase of our experiment, or the first growth stage, due to a fungal gnats problem. The high fungal gnats issue caused a high percentage of bacteria that could have mixed with our abundance reads, so we grew a retrieval phase within two weeks.

Conclusion

Most bacteria in the Neobacillus bacterial species help with ecological balance and regenerative compound generation.

Heyndrickxia bacteria help enhance nutrient availability, stimulate root growth, and soil particle aggregation.

These genus's showed the most change/ growth after crop rotation occurred. We noticed faster root growth and vegetation signs within our rotated plants, which accommodates these observations. Though we put the same standard NPK solutions during all phases of the trial, these bacteria clearly helped the rotated crops with their intake of the nutrients.

With this information, the inoculation of bacteria within the Neobacillus and Heyndrickxia genus can occur during crop rotation. This will allow the benefits to be retained and even enhanced as time passes and climate change makes the weather even harder to predict. An even closer scale investigation on these genus's could narrow down our findings to specific species, but that is too broad of an experiment scope for current nanopore technology.

Next Steps

To help this experiment we would have to repeat this process multiple times. With the limited amount of time we had, we were not able to run the experiment multiple times. Due to this, the results of our experiment could have been a fluke or not completely accurate. Multiple rounds of the experiment would significantly help. If this experiment is conducted again, it would be known that the soil could potentially attract gnats. We would know not to overwater the plants and keep them more controlled overall. If we wanted to change this experiment on another level, we could also mess around with the timings and the types of plants used.