ABSTRACT 2024-2026

The Illinois Junior Academy of Science
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CATEGORY	Agriculture	STATE REGION #	3
SCHOOL	Decatur Classical	IJAS SCHOOL #	3029
CITY/ZIP	Chicago/60645	SPONSOR E-MAIL	gjkovach@cps.edu
SPONSOR	Gerard Kovach	SPONSOR CELL (optional)	

MARK ONE: ☑ ~~EXPERIMENTAL INVESTIGATION~~ DESIGN INVESTIGATION ☐

NAME OF SCIENTIST*	Maizie Koentopp	GRADE	8
NAME OF SCIENTIST		GRADE	
NAME OF SCIENTIST		GRADE	
NAME OF SCIENTIST		GRADE	

* If this project is awarded a monetary prize, the check will be written in this scientist's name, and it will be his/her responsibility to distribute the prize money equally among all participating scientists.

PROJECT TITLE Endophytic Impact on Plant Root Hairs and Brix

Purpose or Problem: The purpose of this project was to figure out if buckwheat grown in different microbial environments impacted the root hair growth and plant tissue sugar content.

Procedure: This project was performed by growing buckwheat in sterilized soil (control), and sterile soil inoculated with: endophytes, native soil microbes, and a combination of the two. The roots were then collected at different stages of growth to measure root hairs. A refractometer was used to collect Brix levels.

Conclusion: The data shows that buckwheat inoculated with endophytes had higher root hair levels, and the control had the highest brix levels due to the sample group being less developed. This means that inoculation is an effective method at improving plant growth.

1) Limit Abstract to 3 paragraphs (about 250 words or less). a) Purpose - what you set out to investigate; b) Procedure - how you did it; c) Conclusion - based on your results. Label each paragraph.
2) Must be typed, single-spaced on the front of this form. Do not write on the back of this form.
3) Three copies of your complete paper are required at the State Science Project Exposition.
Four copies of your complete paper are required for the State Paper Session Competition.

This form must be used. This form **must** be displayed on the front of the exhibitor's display board. It may be reduced to half a sheet of paper; 8.5 inches (vertical) X 5.5 inches (horizontal).

SAFETY SHEET
2024-2026

The Illinois Junior Academy of Science

Experimentation or design may involve an element of risk or injury to the student, test subjects and to others. Recognition of such hazards and provision for adequate control measures are the joint responsibilities of the student and sponsor. Any projects that violate any of the safety rules without a special circumstance that has been presented to and accepted by the IJAS State board will be disqualified from the competition.

Follow each of the following steps:

1. The sponsor, student, and parent (when appropriate) need to read ALL of the safety guidelines in the current IJAS Policies and Procedures carefully **PRIOR** to starting the project.
2. Determine whether the project violates any of the IJAS safety regulations. If there are any questions related to any of the safety rules, the sponsor should contact the Regional Safety Chair. Sponsors, students and parents must not contact the State Safety Chair directly.
3. If the Regional Safety Chair has further questions, they should contact the State Safety Chair stating the region and specific questions related to the project. Sponsors, students and parents must not contact the State Safety Chair directly.
4. No project is without any hazards. A blank safety sheet or one without any listed hazards will not be accepted as complete.
5. If the project involves humans in any way, microorganisms, vertebrate animals, or cells or tissue cultures, the proper endorsement form must also be filled out and included with this safety sheet.
6. This Safety Sheet must be included in the paper, on the project board and turned in electronically when the project is entered into the State Competition. (Rules for regional competitions may vary).

Location(s) where procedure was conducted: School science lab and at home	______If project was done at an outside facility, check here and include a letter from the facility with this form assuring safety was adhered to and that the student role in the project.
Were microorganisms involved in any way? Yes	__X__ If yes, check here and fill out a Microorganisms Endorsement sheet
Were humans involved in as a test subjects? No	_____If yes, check here and fill out a Humans Endorsement Sheet
Were cells or tissue cultures involved in any way? No	_____If yes, check here and fill out a Tissue Culture Endorsement Sheet
Were cells or vertebrate animals involved in any way? No	_____If yes, check here and fill out a Vertebrate Animal Endorsement Sheet
Were there any violations of IJAS Safety Rules that were either approved by the IJAS SRC or by a qualified institution? No	_____If yes, check here and include the letter from the IJAS SRC or the qualified institution where the project was conducted.
List any chemicals used: Bleach	Precautions taken with use of chemicals: Gloves, goggles, and a face masks were worn.
List any other safety hazards that are not covered on the endorsement sheets: Handling of scissors, use of microwave, handling of hot soil.	For each listed safety hazard, describe what precautions were taken: (note this can extend to the next page if needed) Kept scissors away from face and used safety goggles when necessary. Let soil cool and used gloves when necessary.

I/We the student(s) who conducted this project have followed each of the steps listed above and have used all of the safety precautions stated.
Signature(s)____ **Date 1/13/2025**

I, the sponsor of this project take responsibility for the instruction of the student on safety precautions and/or verified the safety credentials of any institution with which they worked.
Signature____________________ ***E-mail*** **gjkovach@cps.edu Date 1/13/2025**

Microorganism Endorsement
2024-2026

The Illinois Junior Academy of Science

Students and sponsors doing a microorganism project must complete this form, prior to experimentation. The signature of the student or students and the sponsor indicates that the project was done within these rules and regulations. Failure to comply with these rules will mean the disqualification of the project at the state level. This form must follow the Safety Sheet in the project paper and on the project board.

1. It is the sole responsibility of all teacher(s)/sponsor(s) to teach students proper safety methods and sterile techniques.
2. The Illinois Junior Academy of Science prohibits the use of primary or secondary cultures taken from humans or other vertebrate animals in any project because of the danger from unknown viruses or other disease-causing agents that may be present. Pure cultures of microorganisms known to inhabit vertebrate animals may be obtained from reputable suppliers and used in proper settings.
3. Microorganism experiments must be conducted in a laboratory such as science classroom or research facility.
4. Projects involving viruses and recombinant DNA should be done with the help of a professional and should comply with the National Institutes of Health (NIH) Guidelines unless the project is limited to a kit obtained from a legitimate supply house.
5. All cultures should be destroyed by methods such as autoclaving or with a suitable NaOCl (bleach) solution before disposal.
6. All rules within the IJAS Policy and Procedure Manual are followed.

Complete all boxes of the following chart.

Genus and species of organism(s) being used.	Arthrobacter globiformis; Azospirillum brasilense; A. lipoferum; Azotobacter chroocochum; A. paspali; A. vinelandii; Bacillus amyloliquefaciens; B. atrophaeus; B. Licheniformis; B. megaterium; B. pumilus; B. subtilis; B. thuringiensis; Brevibacillus brevis; Lysinibacillus sphaericus; Micrococcus luteus; Pseudomonas putida; Rhodopseudomonas palustris; Rhodospirillum rubrom; Streptomyces grinseus
List the location where the lab work was conducted.	School science lab
Name of the reputable source that supplied the organism(s).	Advancing Eco Agriculture https://advancingecoag.com
Method of disposal of the organism(s) being used.	To dispose of the bacteria, the inoculated seeds were immediately planted in soil, preventing any contamination. All materials were thoroughly cleaned with bleach.
Were there any violations of IJAS Safety Rules that were either approved by the IJAS SRC or by a qualified institution?	_____If yes, check here and include the letter from the IJAS SRC or the qualified institution where the project was conducted.
Describe the use of microorganisms in this project.	The microorganisms were used to inoculate buckwheat seeds. They increased root hair amount and sped up development.
Other precautions taken to ensure microorganisms are used safely in this investigation.	To properly use the bacterial inoculant, the researcher followed all the proper safety instructions. When handling the powder inoculant, gloves, goggles, a lab coat, and a face mask were used. In preparation for the inoculation, the space and all materials were wiped with bleach. Afterward, the workplace was also wiped with bleach and the researcher washed their hands thoroughly.

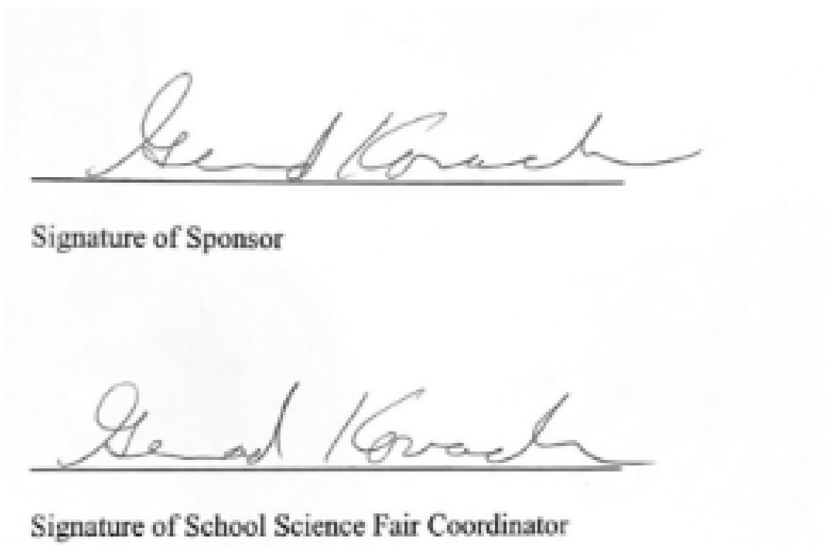
I/We the student(s) who conducted this project have followed each of the steps listed above and have used all of the safety precautions stated.
Signature(s) Maisie Koentopp **Date 3/18/25**

I, the sponsor of this project take responsibility for the instruction of the student on safety precautions and/or verified the safety credentials of any institution with which they worked.
Signature [signature] ***E-mail*** **gjkovach@cps.edu Date 3/18/25**

Endophytic Impact on Root Hairs and Brix

Agriculture

Experimental Investigation



Maizie Koentopp

Decatur Classical School

Chicago, IL 60645

Grade 8

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ACKNOWLEDGMENTS

I would like to thank my science teacher, Mr. Kovach. Throughout my project, he has supported and guided me through the challenges I faced. I am also very grateful to my parents, who have greatly helped me with my project. My dad has been an amazing mentor and guide during this endeavor. I also received mentorship from scientists, including Mike Ricketts of Argonne Laboratories, Dr. Shemuel Israel, a retired soil scientist and supporter of urban agriculture initiatives in Chicago, and Dr. Akilah Martin from the U.S. Army Corps of Engineers. They helped me hone my ideas and provided new insights on how to think about my experiment and collect and interpret my data. Finally, I would like to thank my peers, who have encouraged and supported me.

PURPOSE

The researcher chose to investigate endophytic bacteria and how they affect plant health because using beneficial bacteria can be an environmentally friendly alternative to harmful agrochemicals. Sustainable agricultural practices are the solution to problems originating from the use of synthetic chemical products to increase food production. Microbes offer versatile capabilities and are an attractive and feasible option for developing tools to replace harmful chemicals. Therefore, the first step people must take to replace these chemicals is to explore the microorganisms that reside in close proximity to the plant, also known as the rhizosphere. It is rich in nutrients compared to the soil outside of the rhizosphere and hence exhibits intense biological and chemical activities, causing it to be a lucrative option for developing soil fertility (Prashar et al., 2013). Harmful pesticides that farmers use are causing major environmental problems and disrupting soil ecosystems. Farmers still need an option that supports the number of healthy plants they want to grow, and one solution to this problem is using endophytic bacteria in the rhizosphere. This benefits plants in many ways and makes them healthier overall, without harming the soil and supporting different parts of the ecosystem. This experiment was designed to learn about and demonstrate how endophytes can be one of the best options to replace these chemicals.

The researcher chose buckwheat as the subject of this experiment because it is both a food crop and a beneficial cover crop. Buckwheat grows quickly and is used for weed suppression, a nectar source for native pollinators, and adding nutrients back into the already damaged soil (Boglaienko, 2014). It also adds value as a food source for both humans and livestock. It is a good option for farmers looking for an environmentally friendly cover crop. All

PURPOSE (Continued)

in all, if humans don’t change their farming practices, pesticides will continue to ravage our world. People must move away from these harmful practices, and begin doing things like inoculating seeds and soil, among many other regenerative agricultural practices, to support our environment before it is too late.

HYPOTHESIS

If the researcher compared buckwheat grown in different microbial conditions, then the plants inoculated with endophytes and naturally occurring diverse soil microbes will be the healthiest overall. Endophytic Bacillus is a genus that provides plants with a wide range of benefits, including protection against phytopathogenic microorganisms and insects. They elicit resistance and promote plant growth without causing damage to the environment. Bacillus such as Bacillus thuringiensis, B. subtilis, B. amyloliquefaciens, B. velezensis, B. cereus, B. pumilus, and B. licheniformis have been studied the most for their application in agriculture, although the genus as a whole shows great potential. Various bioactive compounds have been predicted to be present in the rhizosphere in higher numbers due to the increasing number of whole-genome sequenced endophytic Bacillus spp. strains. This data reveals endophytic Bacillus species as an underexploited source of novel molecules of biotechnological interest (Medison, 2022). One can ascertain from this that when implemented, the Bacillus species can positively affect plants. Therefore, when the researcher inoculates the buckwheat plants with Bacillus, as well as other endophytes, it will have a positive impact on the plant. The inoculated buckwheat will be aided by the endophytes and the microbes found in native soil. Bacillus subtilis exhibits many signs of using biocontrol mechanisms to suppress disease caused by pathogens. This includes the synthesis of many secondary metabolites, hormones, cell-wall-degrading enzymes, and antioxidants supporting the plant to defend itself. It also stimulates plant growth and the induction of acquired systemic resistance. Bacillus subtilis solubilizes soil phosphorus, enhances nitrogen fixation, and produces siderophores that promote plant growth and suppress the growth of pathogens. It enhances stress tolerance in plant hosts by inducing the expression of

HYPOTHESIS (Continued)

stress-response genes, phytohormones, and stress-related metabolites (Hashem, n.d., 1291-1297). This explains how Bacillus is beneficial in many direct and indirect ways and has a statistically relevant impact on overall plant health. If Bacillus is used as an inoculant, there will be a clear difference in the plant's health. It will allow the plant and the soil surrounding the plant to be exponentially healthier.

REVIEW OF LITERATURE

The researcher's project focused on whether bacterial endophytes can increase plant nutrient uptake and overall health. The researcher inoculated buckwheat seeds and grew them in different microbial environments to ascertain if the endophytes were beneficial to the plants. According to the USDA, around 1 billion pounds of pesticides are used each year in the United States to increase crop production, and these various chemicals cause many issues. This overuse of pesticides pollutes water, damages soil, and harms many organisms (Shoda et al., 2017). Farmers have come to rely heavily upon pesticides. There are many solutions to this problem, and one method the researcher chose to explore is using beneficial endophytes to support agriculture. The researcher wants to know if endophytic inoculants improve plant growth.

What Are Bacterial Endophytes?

Endophytes are beneficial symbiotic microbes that live in almost all plant species. Using endophytes offers an opportunity to maximize crop productivity. They promote plant growth in many ways, such as through nitrogen fixation (Kandel, 2017). Plants work with the soil biosphere, where they have a symbiotic relationship with endophytic bacteria, improving soil and plant health. Farmers could greatly benefit from adding endophytic bacteria to the soil because it will improve soil health and increase crop production.

Endophytic bacteria are microorganisms living in a plant's leaves, roots, stems, shoots, and flowers. They promote plant growth and benefit the soil by providing and enhancing nutrient availability and biological control mechanisms against pathogens and insect pests. These bacteria have multiple traits that promote plant growth, and they all work together to ensure the plant is

REVIEW OF LITERATURE (Continued)

healthy (Medison et al., 2022). When plants are inoculated with endophytes, the increased presence of specific bacteria ensures increased plant health. An easy environmentally friendly solution for farmers who want their plants to be healthier and absorb more nutrients is to inoculate their seeds with beneficial endophytes that aid their specific crops.

What Is the Rhizosphere?

The rhizosphere is the media in which the roots of a plant live, and it is populated by countless microorganisms. The rhizosphere was initially defined as the area surrounding a plant's root system that has a unique population of microorganisms. These are influenced by the chemicals released from plants' root hairs. These ideas have now been refined, and the rhizosphere includes three zones (Hartmann, 2007). This shows that the nuanced environments surrounding a plant's roots are extremely valuable because they open up doors for further exploration. The microorganisms work directly with the plant to improve both soil and plant well-being. When the rhizosphere lacks nutrients, the root hairs can't perform their job, and the plant suffers. This means people need to preserve the health of the soil so that the rhizosphere ecosystem can function properly in relationship with the plants.

Additionally, even though the rhizosphere is small, it contains vast amounts of microorganisms that all complete different processes that affect plant health. The rhizosphere is pictured as a small volume of soil around the roots of a plant, but in reality, it extends past that into complex systems of distinctly functioning zones. The systems include many things like nutrient fluctuation involving root hairs and nutrient absorption from various organisms (York et

REVIEW OF LITERATURE (Continued)

al., 2016). This demonstrates how the complicated systems that occur in the rhizosphere have a long-lasting impact on soil and plant health. Mycorrhizal fungi, fixing bacteria, plant growth-promoting rhizobacteria (PGPR), biocontrol microorganisms, mycoparasitic fungi, and protozoa are instrumental to the rhizosphere. If farmers want plants to absorb nutrients more effectively and promote crop health, a critical step is investing in soil health through the inoculation of endophytes.

What Are Brix and What Do They Mean in Plants?

Brix is a vital element in plant health that is an effective way to measure overall plant well-being. It is a way to determine the sugar levels of plants, and can be used to gauge the health of the plant. When you have crops with high Brix levels, it is an indicator that they are healthier because they are producing more energy (Brix Measurements and Its Role in Plant Nutrient Management, 2021). An easy way to quantify the nutrients in a plant is to measure the Brix levels. Farmers or gardeners who want to quickly and effectively determine the health of plants by using a Brix refractometer.

Furthermore, plants use sugar as energy, which they use for various biological functions, so sugars give us insight into the health of a plant. All organisms, including plants, use energy to grow. Plants do this by absorbing light and through photosynthesis they turn CO_2 and H_2O into $C_6H_{12}O_6$. The resulting glucose energy helps plants grow, is stored in reserves, and is released into the rhizosphere, providing energy for that ecosystem (Eveland, Jackson, 2011). When plants don’t have enough sugar, they cannot supply the energy needed for higher functioning. When

REVIEW OF LITERATURE (Continued)

Brix data is collected, it indicates the health of the plant. This is especially relevant to farmers because they can use Brix to determine crop health.

What is the Rhizophagy Cycle?

The rhizophagy cycle is a process that occurs within the rhizosphere benefiting plants and soil. In this cycle, microbes exit root hair tips depleted of nutrients, causing each hair to undergo a growth spurt. This happens about every fifteen minutes. Once the microbes enter the rhizosphere, they replenish their nutrients. Then, they enter the root cell periplasmic spaces with the nutrients they replenished. The plant then uses these nutrients to grow and complete different processes (White, 2018). It is evident that the rhizophagy cycle is vital to plant health. When increased amounts of endophytes are added into this cycle, it augments benefits to their plant hosts. This symbiotic relationship between the bacteria and plant is also beneficial to the overall soil health. Additionally, this explains why counting root hairs is a good strategy to determine plant health. If the cycle is functioning correctly, the plant should have many root hairs. If the researcher counts root hairs, it will be a good indicator of plant and soil health.

Why Are Endophytes a Better Alternative to Agrochemicals?

Implementing endophytes will protect the environment from the negative impacts of agrochemicals. Future agriculture will be difficult due to the lack of water and nutrients in many parts of the world because of climate change and decades of soil-degrading agricultural practices. It is therefore crucial to improve understanding of processes that can increase plants’ ability to

REVIEW OF LITERATURE (Continued)

use water and nutrients in the soil. Increases in root hair volume have been linked to increased plant productivity especially when soil resources are limited (Holz et al., 2017). Around the world, climate change is one of the biggest threats to the planet. If people want to continue to produce food for all the humans on Earth while at the same time protecting the environment, people must move away from harmful agrochemicals and instead turn to other methods. Inoculating seeds with endophytes to target the rhizosphere specifically is one of the best options for many reasons. It is easy for farmers to inoculate seeds in bulk quickly, instead of adding soil amendments to the entire field of crops. They benefit root hair growth, which in turn impacts overall plant processes. Inoculants can do the same job as harmful pesticides but benefit the planet.

For decades, farmers have utilized harmful agricultural practices when using industrial farming techniques that rely heavily on petrochemicals and over tilling the soil. The use of agrochemicals in agriculture has grown dramatically over the past 30 years. Currently, approximately 600 active pesticide ingredients are used, but people only have data available for approximately 100 of these. Humans are often exposed to agrochemicals, and this results in serious health effects, including acute and chronic neurotoxicity, lung damage, chemical burns, and infant methemoglobinemia. A variety of cancers also have been linked to exposure to various pesticides, particularly hematopoietic cancers (Weisenburger, n.d.). Pesticides also affect our environment. OC compounds, or volatile organic compounds, could harm the tissues of virtually every life form on the earth, the air people breathe, the lakes and the oceans, and all components of every ecosystem on earth (Actar et al., n.d.). People can no longer rely on

REVIEW OF LITERATURE (Continued)

harmful agrochemicals and must instead turn to more regenerative options. Using inoculants will protect human and ecosystem health while allowing farmers to produce sufficient crops.

Similar Research

Similar experimentation and research have been done at AL-Azhar University in Cairo, Egypt. They took medicinal plants, like Teucrium polium L., and isolated specific bacterial and fungal endophytes like Bacillus cereus and Bacillus subtilis. They then analyzed plant nutrients and root health when the isolated endophytes were applied. They found that the endophytes were beneficial to the plant, meaning that endophytes isolated from medicinal plants possess a vital role in promoting plant health (Hassan, 2017). The researcher has concluded that many other types of endophytes, as well as the ones isolated in that experiment, will be beneficial for plants. These endophytes will greatly impact crop production and sustainability, so more research must be done in this area.

In conclusion, using endophytic inoculants in crop production will greatly benefit plant health and reduce the harmful impacts of pesticides on our environment. The researcher will inoculate seeds with beneficial bacterial endophytes and examine the Brix levels and root hair numbers to ascertain the level of impact the endophytes will have. If applied commercially, the answer to this question will have a huge impact on agricultural systems and our environment.

MATERIALS

- 72 buckwheat seeds
- 5 plastic seedling trays, each with 9 8 cm cells
- Plastic trays to go under the seedlings to catch water runoff.
- Clear plastic covers for the trays
- 2.5ml Biocoat gold endophytic plant inoculant
- 1 optical Brix refractometer
- Broad spectrum LED plant grow lights
- 4400 ml of filtered water to water the plants
- 170 liters of filtered water to clean roots and sanitize pots
- 50ml measuring cup
- 440 grams of native organic soil
- 3080 grams of sterilized potting mix
- Sanitizing bleach spray
- 10 3-liter plastic bags
- Access to a microwave
- Access to a hose
- Three 118ml glass cups
- Safety goggles
- Face mask
- Nitrile gloves
- A pair of tweezers

MATERIALS (Continued)

- A digital microscope
- 1 microscope slide
- A ruler
- 26 milliliters of sulfur
- One roll of wax paper (23 meters)
- 1 Inline hose water filter

METHODS OF PROCEDURE

1. Bleach all the planters and trays by spraying them with bleach and letting them sit for at least five minutes. Then rinse thoroughly with water.
2. Fill each plastic bag with a potting mix. Microwave each soil-filled bag for 5 minutes at full power with the top unsealed to sterilize it. When done, seal the bags and let them sit until cool.
3. Measure 80 grams of sterilized soil and fill each of 18 cells in 2 trays with soil.
4. Measure 60 grams of sterilized soil and fill each of 18 cells in 2 trays with soil.
5. Repeat steps 3-4 adding 80 grams to 4 cells and 60 grams to 4 cells of the fifth tray.
6. Collect 880 grams of soil from a native, organic garden and sift through it thoroughly to ensure there is no non decomposed organic matter or macroinvertebrates and vertebrates.
7. Add 20 grams of garden soil to each cell in the two trays with 40 grams of sterilized potting mix. Also, add 20 grams of garden soil to the 4 cells in the fifth tray with 40 grams.
8. Add 36 buckwheat seeds and 20 ml of water to each of the two small glasses to dampen all the seeds.
9. While wearing PPE, add 2.5ml of inoculant to one glass containing buckwheat seeds, making sure not to contaminate the other. Stir until all seeds are fully coated.
10. Use sterilized tweezers to plant the seeds:
    a. Plant 2 inoculated buckwheat seeds in 9 cells of the tray with fully sterilized soil, and do the same for one of the trays with both sterilized soil and native soil.

METHODS OF PROCEDURE (Continued)

b. Plant 2 regular buckwheat seeds in 9 cells of the other tray with fully sterilized soil, and do the same for the remaining tray with both sterilized and native soil. In a fifth tray plant one inoculated seed in the sterilized soil, one inoculated seed in the native soil, one regular seed in the sterilized soil, and one regular seed in the native soil. You will have 1 empty cell in the fifth tray.

11. Add 100 ml of water to each plant and cover each tray with the clear plastic lids.
12. Move the trays under the grow lights, making sure the lights are at the appropriate height just above the lids.
13. Water with filtered water (use an Inline hose water filter) on days 3, 7, 13, 16, 19, 22, and 26. After 4 weeks, you will collect two plants from each tray. Continue watering the rest every three days.
    a. On day 7, test soil pH and if 7 or higher, add .5 milliliters of powdered sulfur into each container. Add 100 ml of water, ensuring the sulfur is absorbed into the soil.
14. After 4 weeks, collect the data from some of the roots.
    a. Take one plant from each group at a time, to make the data more accurate. Remove one of each plant. Carefully remove excess soil and lay the roots onto a window screen. Spray with water, and wash all the soil off without breaking the roots. Repeat this step with a second set of roots from each group.
    b. Lay the roots on a piece of wax paper, taping each stem down and labeling it with a Sharpie. Use tweezers to carefully separate all the roots from each other.

METHODS OF PROCEDURE (Continued)

  c. Let the roots dry for 2 hours. If they become too dry and brittle, mist them with a spray bottle to rehydrate.

15. Carefully remove the tape from each root. One at a time, put each root onto a microscope slide.
16. Use the microscope at 10x magnification. Take 15 pictures of different areas of the root, making sure to look for root hairs.
17. Repeat this process (steps 13a-15) with the 37-day-old plants taking 40 photos, but this time save the stems and leaves.
18. Take the leaves from each group and pulverize them, squeezing the liquid out of the leaves and onto the refractometer.
19. Look into the refractometer to collect and record brix-level data.
20. Let the plants in the 5th tray grow for 3 months, watering every 3 days. After 3 months, repeat steps 13a-15 a third time with one plant from each group, taking 40 photos.

RESULTS

Brix at 3 Months of Growth	Native Microbes and Inoculated	Inoculated	Native Microbes	Control
%	4	5	5	6

Day 28, Group 1, 15 Photos	Native Microbes and Inoculated	Inoculated	Native Microbes	Control
Root Hairs	33	129	30	25
Root Hair Sprouts	15	32	15	13
Areas With 15+ Root Hairs	2	6	0	0

Day 28, Group 2, 30 Photos	Native Microbes and Inoculated	Inoculated	Native Microbes	Control
Root Hairs	135	192	152	60
Root Hair Sprouts	29	46	43	25
Areas With 15+ Root Hairs	1	2	1	1

Day 37, Group 1, 40 Photos	Garden and Inoculated	Inoculated	Garden	Control
Root Hairs	214	692	284	186
Root Hair Sprouts	72	63	65	40
Areas With 15+ Root Hairs	2	22	2	2

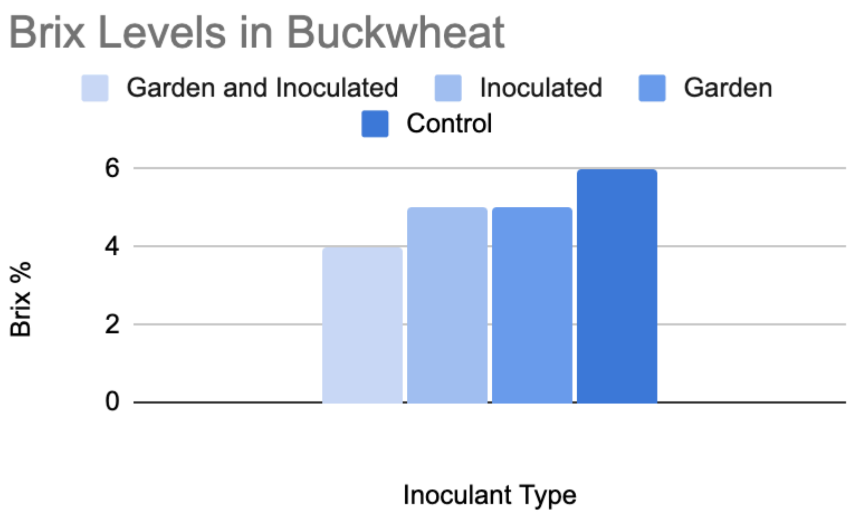
RESULTS (Continued)

Day 37, Group 2, 40 Photos	Garden and Inoculated	Inoculated	Garden	Control
Root Hairs	335	349	341	167
Root Hair Sprouts	83	68	56	59
Areas With 15+ Root Hairs	6	5	7	2

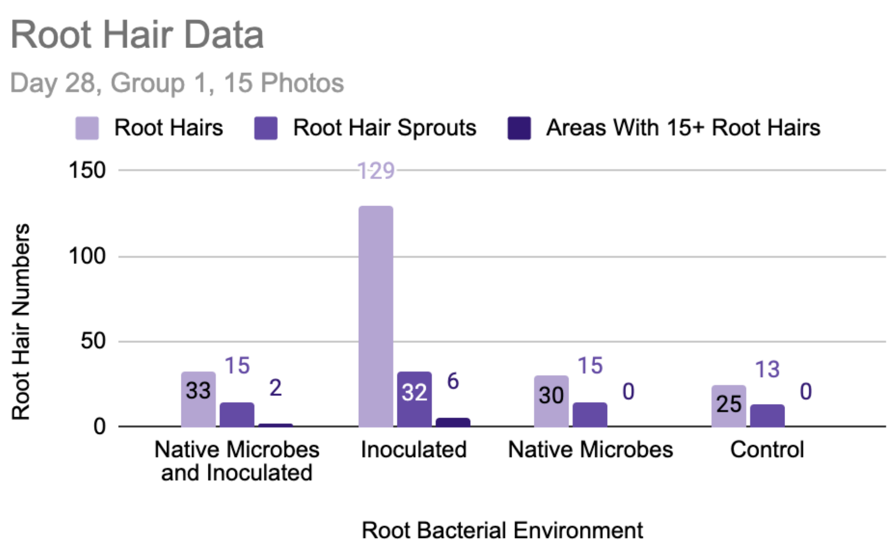
3 Months, 40 Photos	Garden and Inoculated	Inoculated	Garden	Control
Root Hairs	568	765	554	314
Root Hair Sprouts	41	61	69	51
Areas With 15+ Root Hairs	16	18	15	6

RESULTS (Continued)

Brix data from the buckwheat plants:

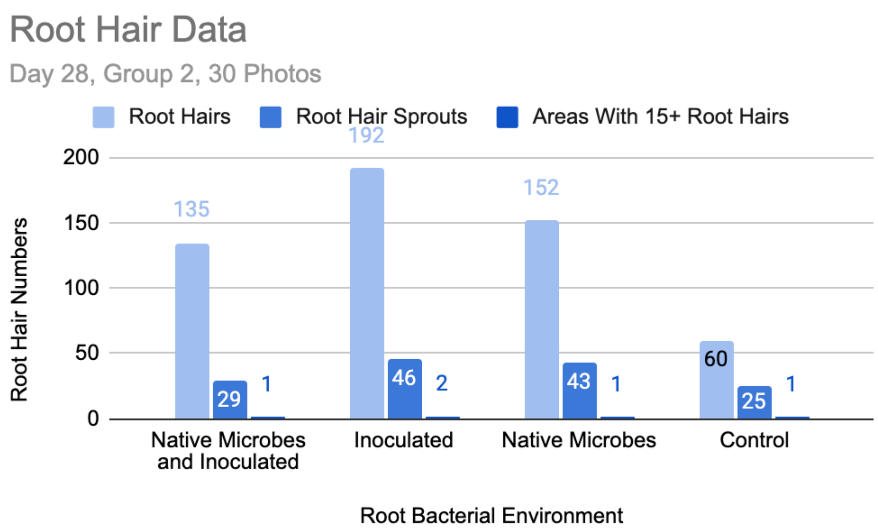


Root hair data collected after 28 days from 15 photos (group 1):

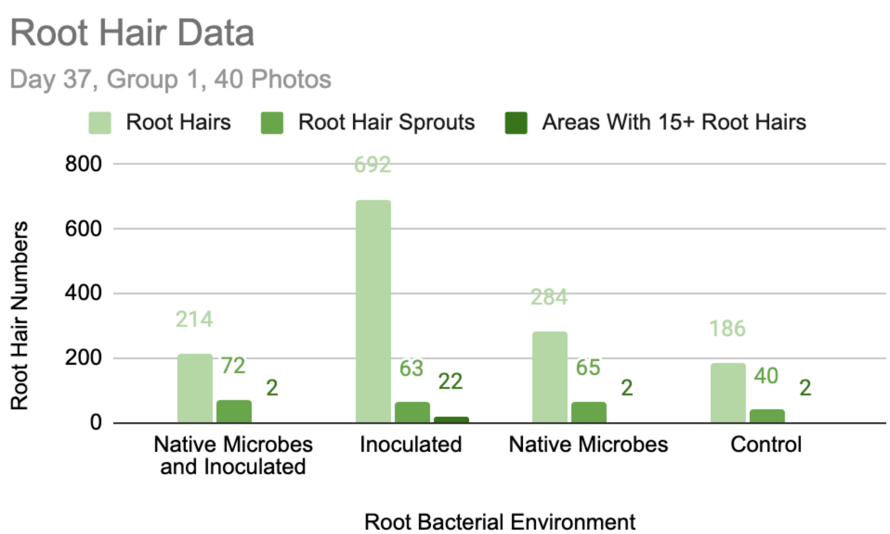


RESULTS (Continued)

Root hair data collected after 28 days from 30 photos (group 2):

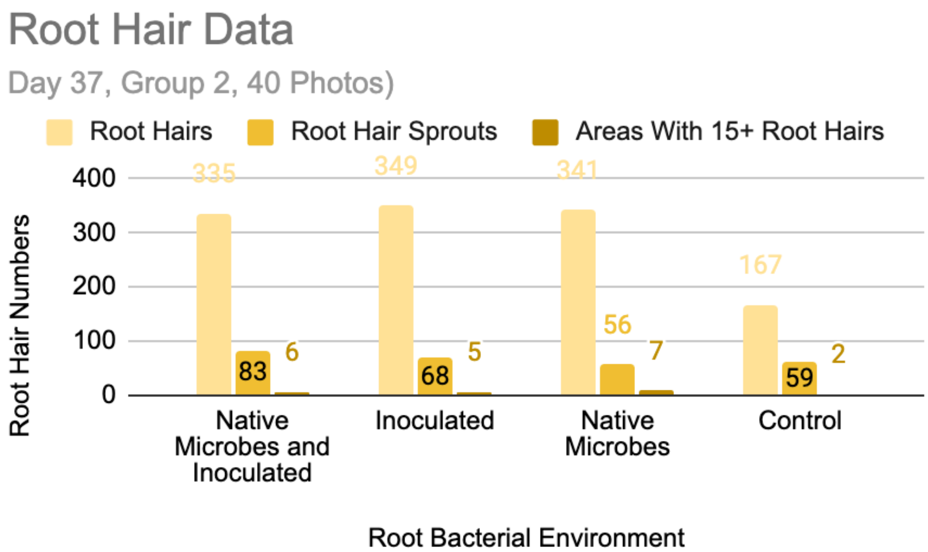


Root hair data collected after 37 days from 40 photos (group 1):

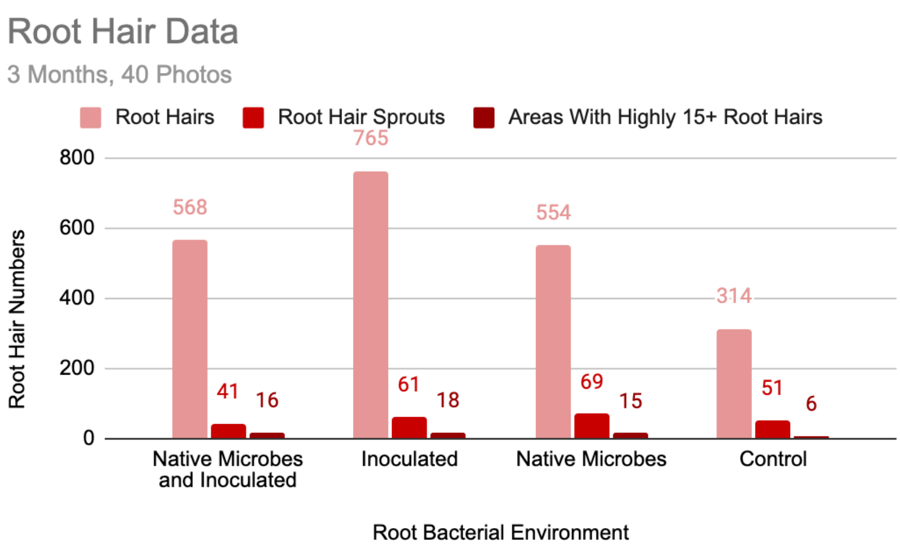


RESULTS (Continued)

Root hair data collected after 37 days from 40 photos (group 2):

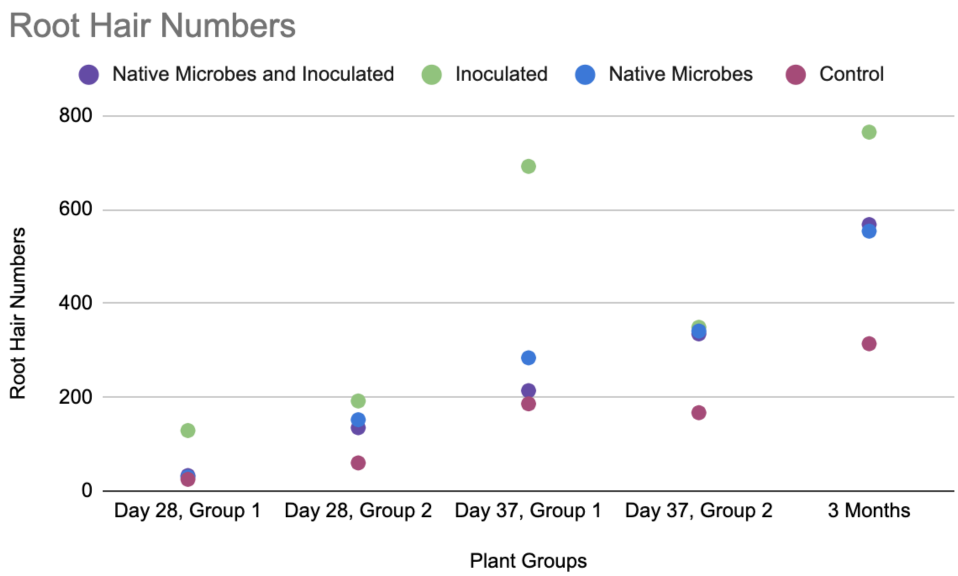


Root hair data collected after 3 months from 40 photos:



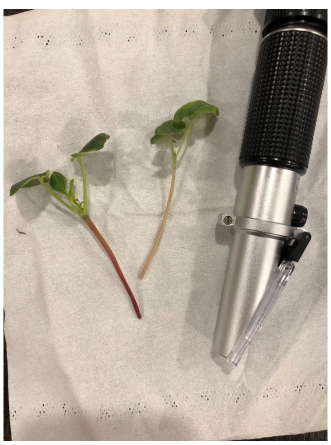
RESULTS (Continued)

Total comparison of root hair numbers:



RESULTS (Continued)

These photos compare plants grown with an inoculant (left) and plants grown in sterile soil (right) after 35 days of growth. Inoculation was observed to be an effective method to improve plant health and speed up development.



Here are the refractometer the researcher used and the plants from which the data was collected.

Here are the washed inoculated buckwheat roots, which were then put under a microscope to count the hairs on the roots.

RESULTS (Continued)

The data shows that the healthiest buckwheat overall was the inoculated buckwheat. The data from the plants 3 weeks old had 129 and 192 root hairs, 32 and 46 root hair sprouts, and 6 and 2 areas with 15+ root hairs. After 37 days, the data showed that they had 692 and 349 root hairs, 63 and 68 root hair sprouts, and 22 and 5 areas with 15+ root hairs. After 3 months, they had 765 root hairs, 61 root hair sprouts, and 18 areas with 15+ root hairs. The inoculated seeds in native microbes and native microbes groups averaged about the same. For 3 weeks, the inoculated buckwheat grown in natural microbes had 33 and 135 root hairs, 15 and 29 sprouts, and 2 and 1 areas with 15+ root hairs. After 37 days, they had 214 and 335 root hairs, 72 and 83 root hair sprouts, and 2 and 6 areas with 15+ root hairs. After 3 months, they had 568 root hairs, 41 root hair sprouts, and 16 areas with 15+ root hairs. The native microbes group had 30 and 152 root hairs, 15 and 43 sprouts, and 0 and 1 for areas with high levels of root hairs after 3 weeks. After 37 days, they had 284 and 341 root hairs, 65 and 56 root hair sprouts, and 2 and 7 areas with 15+ root hairs. After 3 months they had 554 root hairs, 69 root hair sprouts, and 15 areas with 15+ root hairs. The control, which was buckwheat grown in sterile soil, was the least healthy with the data from 3 weeks showing 25 and 60 root hairs, 13 and 25 root hair sprouts, and 0 and 1 for areas with 15+ root hairs. After 37 days they had 186 and 167 root hairs, 40 and 59 root hair sprouts, and both had 2 areas with 15+ root hairs. After 3 months, they had 314 root hairs, 51 root hair sprouts, and 6 areas with 15+ root hairs. For Brix levels, control had the highest percentage of 6, plants grown in native microbes and inoculated plants both had a percentage of 5, and inoculated plants grown in native soil had a percentage of 4. To conclude, the group with the most root hairs was the inoculated buckwheat.

CONCLUSION

The researcher hypothesized that buckwheat inoculated with native microbes and beneficial endophytes would have the highest amount of root hairs and Brix levels. After testing with current methods, the results partially support the hypothesis, however further experimentation with more advanced methods are needed to collect more accurate data. For the buckwheat grown in native soil and additionally inoculated, the data showed that it had the second lowest Brix levels and second highest root hair amount. The inoculated plants had the highest number of root hairs, but the control of buckwheat grown in sterile soil had the highest Brix levels. This occurred because the control plants were less developed due to the lack of bacterial life, and less developed plants have a higher concentration of sugar in the leaf tissue. When plants are more developed, they allocate sugars to flower and reproduce. Therefore, the inoculated buckwheat grown in native soil was the most developed overall. The researcher can learn from this that inoculation is an effective method for growing healthier plants faster. Although the root hair data collected from the plants grown in native microbes and plants inoculated with endophytes and native microbes had similar amounts of root hairs, the plants that were additionally inoculated were more developed. They had more healthy leaves and began flowering at a faster rate, showing how additional inoculation greatly benefited the plants.

Some errors that might have occurred in the researcher's experiment were that the plants weren't in a completely sterile environment, which could have led to contamination of bacteria influencing plant health. Additionally, root hairs could have been damaged during transportation from the growing site to the data collection location. If the researcher were to pursue this experiment further, they would have used the RhizoVision software. This allows the roots to be

CONCLUSION (Continued)

scanned and analyzed in more detail than with a microscope. This would make the results more accurate.

Additional factors influence soil fertility and plant growth, other than various bacteria. These include soil texture, micro-nutrients (such as calcium, sulfur, magnesium, etc.), amino acids, buffering of soil pH, and other physical and chemical factors that could positively affect plant growth. This experiment was only focused on the impact of inoculation on root hairs and Brix levels, so the overall health of the plant cannot be determined by just these factors. The results of this experiment were that buckwheat plants inoculated with beneficial endophytes benefit plant health.

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