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The Effects of Nitrogen and Potassium on the Growth of *Brassica rapa*

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Nominated by: Dr. Leonard Kenyon and Hannah Fazekas, PhD candidate

Kimberly is a senior pursuing a degree in Early Childhood Education. She works on campus and enjoys spending time with her family and friends. She also likes to watch movies with her cats.

Kimberly Notes:
When I began this project, I did not expect to become so invested in the research required to support my findings. I learned a lot about how nitrogen and potassium can affect plant growth when organic substances containing these elements are buried in the soil. I hope people will be able to utilize my findings for future research.

Dr. Kenyon Notes:
Students conducted a semester-long research project using Wisconsin Fast Plants. Students selected two different organic substances to test against control plants. They recorded data on germination, growth and development, flowering, pollination, fertilization and embryogenesis. This paper reflects Kimberly Truong's understanding of scientific writing, statistical analyses, data collection, literature review and clarity of thought.

Brassica rapa, which are more commonly known as Wisconsin Fast Plants® are very closely related to turnips, broccoli, cabbage, and other similar vegetables (Lauffer et al., 2016). Fast Plants® are most commonly known for how fast they can grow and how very little light is needed to make them grow since water, continuous light, and fertilizer is all that is required (Lauffer et al., 2016). Since we knew that very little was needed to make these plants grow, we designed an experiment to test how adding different organic substances can affect the growth of *B. rapa* compared to leaving the Fast Plants® alone.

Plants require chemical elements, which are also known as essential elements, from their surrounding environment to complete their life cycles, (Simon et al., 2016). Within the essential elements, there are macronutrients and micronutrients. A macronutrient is a chemical element that an organism must obtain in relatively large amounts, and examples of this would be carbon, hydrogen, and nitrogen, phosphorous and sulfur (Simon et al., 2016). Other macronutrients that come in smaller amounts are potassium, calcium, and magnesium. A micronutrient is a chemical element that an organism needs in very small amounts (Simon et al., 2016). Examples of micronutrients would be zinc, manganese, iron, and copper. A plant will recycle the atoms of micronutrients over and over, so that it is why only a small amount of them is needed (Simon et al., 2016). A deficiency of any of the micronutrients can kill a plant though (Simon et al., 2016), which is why it is important to make sure a plant can get all the nutrients that it needs.

The macronutrient nitrogen has many different uses within plants. An example of this would be that it is a component of all nucleic acids and proteins in a plant, and in annual plants, nitrogen is mainly used for reproduction (White et al., 2015). Proteins play a very important role in plant growth because stored protein provides building blocks for rapid growth upon seed and pollen germination (Herman et al., 1999). This means a plant could have rapid growth in the height after seed germination and in the growth of pods and seeds after pollination. Since proteins play such an important role in plant growth, then it is important for them to receive adequate amounts of nitrogen in order for proteins to be produced. In perennial plants, nitrogen can be used not only for reproduction, but also for growth and storage for future use (White et al., 2015). Plants use nitrogen by going through the process of mineralization in the soil, which is the conversion of organic nitrogen to ammonium (University of Hawaii at

Manoa, 2016). Once nitrogen has been converted into ammonium, it can be taken up by the plants to use in various ways.

A shortage of nitrogen is the most common nutritional problem for plants and when those same plants produce something like grains, the same nutrient deficiencies can be passed on to humans and livestock who consume them (Simon et al., 2016). In an experiment where different amounts of nitrogen were added to the soil, it was found that when 0% of Nitrogen was added to one of their experiments, plants were shorter in height than the control plants, while 20% of Nitrogen added was not any different from the control (Zhao et al., 2004). That experiment shows how not adding any additional nitrogen to the soil can hurt the growth of the plant. When a plant has a sufficient amount of nitrogen, a plant will usually display vigorous plant growth (University of Hawaii at Manoa, 2016). There are many organic compounds that contain nitrogen, but coffee grounds have about 2 percent nitrogen in volume, which provides bacteria with the energy it needs to make organic matter into a compost, and they can be added to the soil of a plant (Wise, 2008). Since coffee grounds are so rich in nitrogen, then they should be a good source of nitrogen if the grounds were to be incorporated into the soil of a plant.

Besides nitrogen, potassium--along with phosphorus--is another macronutrient that is usually deficient in soils (Simon et al., 2016). One reason potassium is deficient is because their ions diffuse very slowly when compared to the rates in which the roots can absorb those ions (Robinson, 1994). Only 0.1 to 2 percent of potassium is readily available for plants to absorb from the soil while 90 to 98 percent of potassium is relatively unavailable to use because it is resistant to chemical breakdowns. Moreover, the forms of slowly available potassium only make up 1-10 percent of the total potassium supply (The Mosaic Company, 2013). When plants are deficient in potassium, then they are more susceptible to infections when compared to plants that have adequate levels of potassium (Wang et al., 2013). When compared to the other nutrients, potassium plays a critical role in plant growth and metabolism, and contributes greatly with the survival of plants under abiotic or biotic stress (Wang et al., 2013). Since potassium plays such a heavy role with plant growth, but can often be deficient, it is important to make sure that plants have a way of receiving the potassium needed for proper utilization within the plants so they are less prone to infections and diseases.

Potassium is used in various ways within the plant and examples of this would be: the stimulation of early growth in a plant, increased efficiency of water use within the plant, and increased protein production (Rehm & Schmitt, 2002). Protein synthesis would not be possible without potassium because it is required for every major step of it; the "reading" of the genetic code in plant cells to produce proteins and enzymes that regulate all growth processes in the plant would not be possible without potassium (International Plant Nutrition Institute, 1998). There are many organic substances that contain potassium, such as bananas, but potato peels are also an excellent source of potassium (Chandler, 2015). Since potato peels have a lot of potassium in them, they should be very helpful with the growth of plants because potassium activates at least 60 enzymes that are involved with plant growth (The Mosaic Company, 2013).

If potassium is a nutrient that increases protein production, and we add potassium-rich potato peels to an experimental pot, then the Fast Plants® should have more overall growth in the plants than they would without any organic substance added. The growth would include the height, the number of leaves, the number of seeds, the number of flowers, the number of pods, the number of first true leaf hairs, and the length of the largest true leaf. We predict this will happen because an increase in protein production means that there should also be an increase in plant growth since potassium is also used for protein synthesis, which is what allows plants to regulate growth processes.

If nitrogen is a nutrient that is a component for nucleic acids and proteins, and we add nitrogen-rich coffee grounds to one of our experimental pots, then these plants should also have more growth than they would without any added organic compounds. The growth of the plants would include the height, the number of seeds, the number of pods, the number of leaves, the length of the largest true leaf, the number of first true leaf hairs, and the number of flowers. We assume this will happen since nitrogen is an essential nutrient needed in order to form nucleic acids and proteins and because stored proteins provide building blocks for rapid growth after seed and pollen germination.

Method

We started the experiment by using 2 Styrofoam® quads that had four openings in it, for a total of 8 openings, along with holes at the bottom of each opening. Then, we grabbed some felt wicks and cut a few to make sure they were the right size and stuck one wick halfway into each of the holes at the bottom of the openings, so some of the wick was hanging out of the bottom of the quads. Next, we added topsoil into each of the openings halfway up before inserting two pellets of Osmocote® fertilizer into them. More topsoil was added on top of the Osmocote® fertilizer and two Fast Plants® seeds were placed on top of all of the soil of each quadrant, for a total of 16 seeds planted. Finally, the Styrofoam® quads were watered until the felt wick at the bottom was wet and then the quads were placed under grow lights.

After about 5 days from the start of the experiment, we examined the growth of all of the seeds and picked the best nine plants of the sixteen to transport over to three separate pots. Before transporting the plants, we first adhered tape to each of the pots and labeled them Experiment 1 - coffee, Experiment 2 – potato, and Control. Each of the pots also had the numbers 1, 2, and 3 labeled on the tape to help indicate which plants we would be measuring in the future. We filled the pots with topsoil almost to the top of each pot. In experimental pot 1, one tablespoon of decaffeinated organic coffee grounds was added to the soil and mixed in throughout. We incorporated coffee grounds into the soil because the coffee grounds contain nitrogen. Nitrogen is a macronutrient that helps with the growth of plants. In experimental pot 2, about a tablespoon of fresh potato peels that had been just cut up were incorporated into the soil in the pot. Potato peels were used because they contain the macronutrient potassium in them. Potassium is used within the plant and helps with early growth in plants. The control pot was left alone with no organic substance added to the soil. We then carefully removed the best nine plants from the Styrofoam® quadrants before distributing them into the pots for a total of three plants in each pot. Small holes were dug into each of the pots close to the labeled number before the plants were transported. Each of the pots was then placed in its own glass dish and was watered until the water started to drip at the bottom of the pots. Finally, all of the pots were placed back under the grow lights.

One week after moving the plants, some of the plants experienced transplant shock, so we tied the stems of those plants to wooden sticks for support. We also started taking measurements of the plants twice a week for three weeks. We measured the plant height and the length of the true leaves in millimeters with a small ruler and then also counted the number of leaves, flowers, first true leaf hairs, pods, and seeds. At the beginning we were not able to count the number of flowers, pods, and seeds because they had not developed yet. All of the data was recorded in a table online using Microsoft OneNote®.

About two weeks after transplanting the plants, flowers started blooming, and eventually we took dead bees and pollinated each Fast Plants® by rubbing the body of the bees on all of the flowers to give each of the flowers pollen from another flower. This step was very important because without the bees, the flowers would not be able to cross-pollinate. After the flowers were pollinated, the plants started to grow pods. Two weeks after they started developing pods we took the biggest pods off the plants and split the pods open with our fingernails and a razor blade and then counted the seeds that were found in the pods and recorded the last information needed to add to the data table on OneNote®.

We analyzed the data by transferring all of the information recorded on OneNote® to a Microsoft Excel® worksheet. Once the data was transferred over to the Excel® worksheet, we used Pivot Table to organize all of the data and to help find the averages of all the plant traits that we measured. We also calculated the range for each measured variable over the course of the experiment. We then created a bar graph by using the data table with all of the averages of the plant traits measured. Using the averages of six measurement days of the length of the largest true leaf, we made a line graph (Figure 2). Once all of the data was made into graphs, it was easier to compare the results of the different traits that were measured for each of the treatments that were added to the plants.

Results

Table 1: Average of *B. rapa* Plant Traits

The average of the plant traits measured is represented by X (mean), R represents the range, and N represents the number of plants.

Plant Trait Measured	Control			Experiment 1 - Nitrogen			Experiment 2 - Potassium		
	N	X	R	N	X	R	N	X	R
Height (mm)	3	100.4	117.0	3	77.2	74	3	103.9	179
Number of leaves	3	3.3	6.0	3	2.5	4	3	3.1	8
Number of hairs	3	9.8	20.0	3	11.8	26	3	7.6	14
Number of flowers	3	5.4	8.0	3	1.8	4	3	4.2	7
Number of seeds	3	7.0	11.0	3	0	0	3	9.3	12
Number of pods	3	5.3	5.0	3	2	3	3	6.3	7
Length of longest true leaf (mm)	3	22	27	3	12.8	13	3	23.9	31

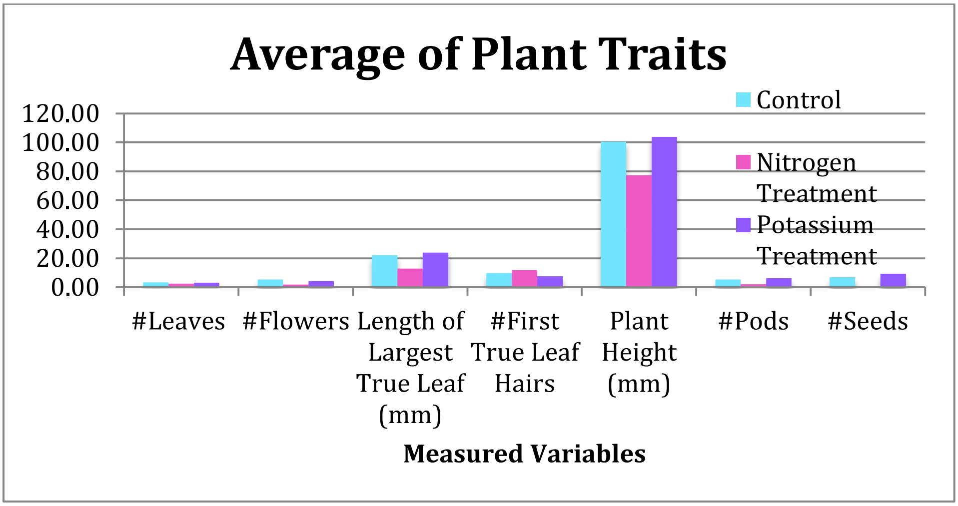

Figure 1: Average of *B. rapa* Plant Traits
The measured variables of all the plant traits with the nitrogen treatment and the potassium treatment that were added to *B. rapa* as well as the control were averaged over a period of six weeks.

Of the seven plant traits measured, four of them followed the same pattern, while the other three followed a different pattern. The potassium treatment had the greatest average height (103.88 mm), the average length of the largest truest leaf (23.88 mm), the average number of seeds (9.33 seeds), and the greatest number of pods (6 pods) (Figure 1). Within the same categories, the control had the next greatest averages, leaving the nitrogen treatment to have the lowest averages. The control had an average height of 100.44 mm, an average of 22 mm for the length of the largest true leaf, an average of 7 seeds, and an average of 5.3 pods (Table 1). The plant that received nitrogen had the smallest height (77.2 mm), the average length of the largest true leaf (12.81 mm), the average number of seeds (0 seeds), and the average of pods (2 pods) (Figure 1). The pattern changes with the control pot having the greatest average of the number of leaves (3.33 leaves), and the average number of flowers (5.42 flowers). The only time the nitrogen treatment had the highest average was with the number of first true leaf hairs (11.78 hairs), followed by the control with an average of 9.78 hairs, with the potassium treatment having the least amount of hairs (7.56 hairs) (Figure 1). The nitrogen treatment also did not produce any seeds (Table 1).

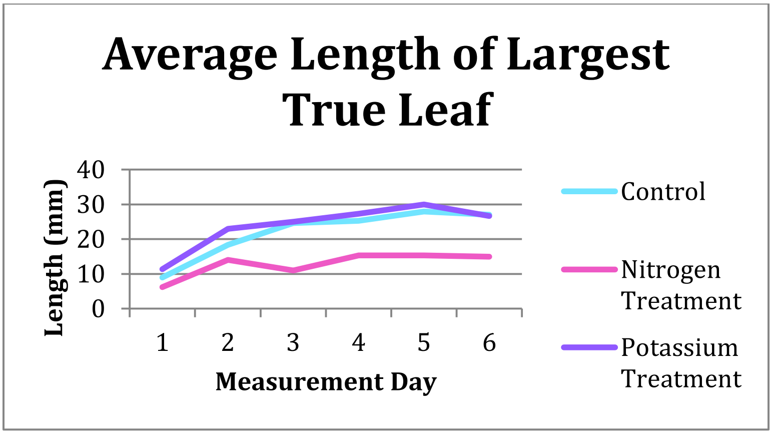

Figure 2: Average Length of Largest True Leaf of *B. rapa*
Over a period of six measurement days, the average length of the largest true leaves for each treatment is displayed on the graph.

During the first two measurement days, all three pots increased in the length of the largest true leaf, with the potassium treatment having the largest true leaf (23 mm), followed by control (18.33 mm), and then the nitrogen treatment (14 mm) (Figure 2). On the third measurement day, the largest true leaf for the nitrogen treatment was significantly smaller than it had been the other two days, while the other two pots continued to increase in length. The nitrogen treatment had a length of 11 mm, while both the potassium treatment and the control had a length of 25 mm (Figure 2). The nitrogen treatment had an increase in the length of the largest true leaf on day 4 before staying at the same length for the remainder of the measurement days. The potassium treatment and the control had an increase in the length of the largest true leaf on measurement days 4 and 5 until they both had a decline on measurement day six.

Discussion

The purpose of the experiment we conducted was to find out the effects of adding 2 different organic materials to the soil of the Fast Plants of growth in Fast Plants® compared to when nothing was added to the soil on various growths within the plants. We found out that one of our hypotheses was rejected while the other one was supported. We hypothesized that both the nitrogen treatment and the potassium treatment would have more overall plant growth than the control, such as the height, the number of leaves, the number of pods, the number of seeds, the number of flowers, the number of first true leaf hairs, and the largest length of the largest true leaf. We would have known this occurred by averaging out all the plant trait variables and comparing them to each other. We expected the nitrogen treatment to have more growth because of the fact that nitrogen is a component of all nucleic acids and proteins, which can cause a plant to have rapid growth after germination (Herman, et. al., 1999). We also hypothesized that potato peels would also have more growth than the control. We presumed this would occur due to the fact that potato peels contain potassium, which aids with early growth within plants because of the increased protein production. Our potassium hypothesis was supported since the potassium treatment had the greatest average of four out of seven measured variables (Figure 1). Similarly, we expected the coffee grounds to also have more height than the control because nitrogen is another important macronutrient, which aids in healthy plant growth. That hypothesis was rejected and can be seen in the graphs where the nitrogen treatment had the lowest averages of all of the plant traits that were measured except for the average number of first true leaf hairs (Figure 1).

The pot that received nitrogen grew the least compared to the other two pots that sprouted up. This could be due to the fact the coffee grounds added to the soil were still fresh. They had just been ground up the same morning that they were incorporated into the soil. If the coffee grounds had been used before mixing them into the soil, it could have made a difference in the growth of the plants. After coffee grounds are brewed, they are close to pH neutral (between 6.5-6.8), which means that the coffee grounds become less acidic once they are brewed (Wise, 2008). It also may have been a good decision to compost the coffee grounds before mixing them into the soil. When testing lettuce seed it was found out that coffee grounds that had not been composted showed poor germination and stunted growth (Wise, 2008). This means that we should have composted the coffee grounds first before mixing them into the soil. We might have also incorporated too many coffee grounds into the soil. Instead of adding a tablespoon of the coffee grounds, it might have been better to instead add just half of a tablespoon or maybe even just a teaspoon. Another reason why the nitrogen treatment did not give off the predicted results could have been because the nitrogen did not go through the process of mineralization. Mineralization is when organic nitrogen is converted to ammonium before it can be used by the plants (University of Hawai'i at Manoa, 2016). This process occurs with time. This means that the nitrogen that our Fast Plants® received may not have had enough time to convert the nitrogen to ammonium for the plants to properly use. It was also possible that the nitrogen was immobilized in the soil. Immobilization is a reverse process of mineralization: converting inorganic nitrogen to organic nitrogen and can occur when the decomposing organic matter contains low amounts of nitrogen and also when there is a high

Carbon to Nitrogen ratio (C:N) (University of Hawai'i at Manoa, 2016). As we observed our plants, the plants that received the nitrogen treatment also started to have a yellow coloration in the older leaves, which could be due to the fact that there was very little nitrogen in the soil for the plants to use properly since it could have been immobilized, causing a deficiency of nitrogen in the soil. Since the plants had a yellow coloration, there could have been a deficiency in the nitrogen, which causes stunted growth in plants.

The pot that received the potassium from the potato peels had the most height compared to the control, as well as the greatest average length of the largest true leaf, the number of pods, and the number of seeds (Figure 1), which supports our hypothesis. If the potassium treatment would not have helped plant growth, then the growth of the plants would have been stunted and have reduced yields (Rehm & Schmitt, 2002). Instead, the growth and development of *B. rapa* that received the potassium treatment flourished since they also had the greatest average in the number of pods and seeds and not just the height (Table 1). Since 60 enzymes associated with plant growth are activated from potassium (The Mosaic Company, 2013), this may have been the cause of greater height than the control. Adding in an organic material with an abundant amount of potassium in it caused a lot more growth to occur within those plants compared to those that received no organic material to the soil. The potassium from the potato peels could have also caused an increase in protein production. Proteins can be a building block for rapid growth to occur within plants (Herman, et al., 1999), which could have been another reason why potassium had the most growth.

Our results supported and rejected our hypotheses. The nitrogen treatment rejected one hypothesis, while the potassium treatment supported the other hypothesis. In a future experiment we could make some adjustments to try and support our original hypothesis for nitrogen, which was to have the nitrogen treatment have more height than the control. Instead of using decaffeinated coffee grounds, we could instead use caffeinated coffee ground to see if that would make a difference. We could also use regular coffee grounds instead of organic ones, and also add smaller amounts of the coffee grounds to the soil. We could have also had more coarse-grained coffee grounds to see if that would have had a significant change. Another method to try and support the hypothesis would be to let the coffee grounds sit in the soil for a while before transplanting *B. rapa* to the treated soil in order to let the bacteria in the soil break down the coffee grounds into a compost (Wise, 2008). We would measure the results of this new experiment the same way that we did for this experiment by recording the data of multiple plant traits, over a period of six weeks and see if any of the changes we made would have a different outcome. It also might make a difference if we make sure the same people measured the same plant traits in order to reduce human error in the experiment.

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