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The Application of Melatonin to Stimulate Growth in Mung Beans

18 December 2019

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Abstract

Fertilizers are a necessary factor in maintaining the high level of agricultural production all over the world. In heavy use, however, fertilizers can cause harm to the environment including groundwater pollution and high soil acidity. As the human population increases and farm land is used more, the continued use of fertilizers will result in more damage to the environment, so it is important for new ways of assisting plant growth to be researched. Melatonin is a naturally occurring hormone in many plants (and animals) that has demonstrated potential as a growth stimulator since its presence was discovered in the developing stages of some plant species. In this study, the possible growth stimulating properties of melatonin were investigated, with the objective of finding out whether mung bean plants grown in water with a melatonin supplement would significantly outgrow those growing in water alone. The mung beans were grown in the same location under the same conditions, and every day of the ten day growth period the water and melatonin were replenished to their respective group of beans. The results of the study showed no significant increase in growth by the mung beans given melatonin, warranting a rejection of the research hypothesis. The experiment in this study examines short term effects with a single melatonin dosage, however, and further research over longer periods of growth and with different melatonin dosages is suggested.

Introduction

Rationale

Agriculture is a primary source of food for humans and animals around the world. Normal agricultural techniques for growing plants involve the use of some type of fertilizer to help maintain soil quality and provide plants with the nutrients they need to grow. The problem is that fertilizers can often cause unintended harm to the environment around them, including but not limited to pollution of groundwater, pollution of water sources via runoff, and harmful levels of acidity in soil ("Fertilisers"). A possible solution to this problem could be the addition of new chemicals or hormones to help reduce the amount of fertilizer needed to grow the plants, which in turn will lessen the overall impact fertilizers have on the environment as less of them are used.

Melatonin, or N-acetyl-5-methoxytryptamine, is a hormone produced by many organisms spanning all three domains of living things (Hardeland). Its purpose differs from organism to organism, but it is most commonly cited as the "sleep hormone," which plays a role in regulating the body's internal clock in many animals, including humans (Bauer). Melatonin has been observed in varying degrees of concentration in a multitude of studies across many organisms in the plant kingdom (Reiter). Its function in plants has also been investigated, and multiple studies have linked melatonin to increased plant growth and have shown that it is present during the first stages of growth in many plants (Arnao). This information on melatonin in plants is compelling enough to warrant research into whether it is capable of increasing the amount of growth exhibited by mung beans (Janas). Mung beans were chosen for this experiment due to their short sprouting time, which will be 10 days for this experiment (N.), and their low requirements in terms of lighting and materials for sprouting (Moran).

Assumptions

- All mung beans will have the same initial mass
- Mineral content of tap water is consistent
- The time exposed to light during water exchange will not significantly affect the growth of the seeds
- Growing conditions for both groups will be identical (besides the addition of the supplement)
- The chosen growth period will be sufficient for the melatonin to have any of its possible effects

Objective

The objective of this research is to determine whether the addition of melatonin to sprouting mung beans seeds will significantly increase the amount of growth (by mass) of the mung bean sprouts.

Expectations

The student researcher expects that there will be an increase in growth in mung beans given melatonin compared to those given just water because of the evidence of melatonin's role in plant growth.

Hypotheses

Alternate Hypothesis (H_1)

Mung bean sprouts grown with a 3 mg melatonin supplement will have significantly higher growth (by mass) compared to mung beans grown without one.

Null Hypothesis (H_0)

Mung bean sprouts growth with a 3 mg melatonin supplement will not have significantly higher growth (by mass) compared to mung beans grown without one.

Materials

Item	Provider	Description of Use
Mung Bean Seeds (40)	Brad Miller	Subject of the research, will be grown
8 oz Mason Jars (2)	Brad Miller	Place of growth for mung beans
Mason Jar Sprouting Lids (2)	Brad Miller - Amazon	Used to close the top of the jars
Tap Water	Brad Miller	Given to both mung bean groups
3 mg Melatonin Tablets (10)	Brad Miller - Amazon	Added to the non-control group of mung beans during growth
Colored Tape	Brad Miller	Used to mark the jar of the non-control group
Dish Soap	Brad Miller	Used to clean out Mason jars
Stirrer	Brad Miller	Used to dissolve melatonin capsules
Cardboard Box (~ 12” x 6” x 8”)	Brad Miller	Used to block light from growth area

Equipment

Item	Provider	Description
Digital Scale (AMIR Digital Kitchen Scale)	Brad Miller	Used to weigh mung beans
Digital Camera (iPhone)	Brad Miller	Used to document research

Facilities

Needed Area	Facility
Growth area	Brad Miller’s house
Data analysis area	HTHS / Brad Miller’s house

Experimental Design Diagram

Title: The Application of Melatonin to Stimulate Growth in Mung Beans

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Null (H_0)
Mung bean sprouts growth with a 3 mg melatonin supplement will not have significantly higher growth (by mass) compared to mung beans grown without one.

Independent Variable: Melatonin content of the water in which the mung beans are grown

Levels:	No Supplement	3 mg Melatonin Supplement
# of Trials:	20	20
Control:	Control	

Dependent Variable: The growth of the mung bean sprouts after a 10 day growth period

Operational Definition of Dependent Variable: Mass of each mung bean sprout after growth period (grams)

Constants:

Growing environment (temperature and humidity)
Lighting (inside cardboard box; no light)
Amount of water given to each group per day (100 mL)
Mung bean seed source (Pride of India Whole Green Mung Beans)
Container for plant growth (8 oz mason jar)
Water source (tap)
Melatonin source (Pure Encapsulations)
Area of plant growth (Brad Miller's House)

Methodology: Experimental Setup, Graphics, Illustrations

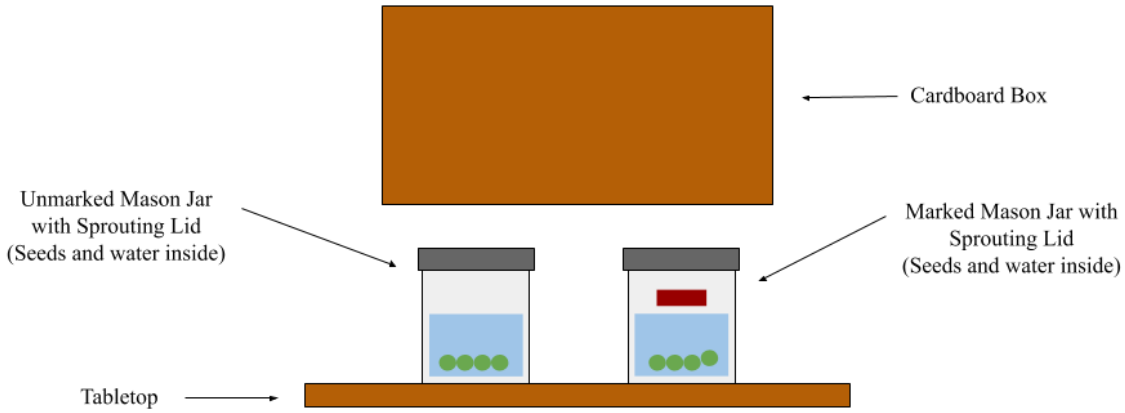


Figure 1: Growth Setup (The cardboard box above the jars will be placed over the top of them)

Figure 2: Materials

Figure 3: Completed Growth Setup

Figure 4: Final Day of Growth

Figure 5: Separation of Control Group for Weighing

Figure 6: Separation of Non-Control Group for Weighing

Methodology: Procedure

1) Randomly separate the 40 mung bean seeds into two groups of 20.
2) Sterilize both mason jars using dish soap and water, dry thoroughly.
3) Mark one mason jar with a piece of colored tape.
4) Fill each mason jar with 100 mL of water.
5) Add one 3 mg melatonin capsule to the marked mason jar, stir until dissolved.
6) Put one group of 20 seeds into each jar and put on their sprouting lids.
7) Place the mason jars next to each other on a table (or other flat surface) and place the cardboard box on top of them such that no light reaches the mason jars.
8) Allow the seeds to grow for 24 hours.
9) Take the cardboard box off and remove the mason jars.
10) Take of the sprouting lids and separate the seeds from the fluid by pouring them through a strainer over the sink (separate strainers for each jar).
11) Repeat steps 4 - 10 nine more times, returning the jars to the same place each time.
12) Empty the unmarked jar into a strainer.
13) Dry and weigh each sprout from the strainer and record the measurements in the data table in the “No Supplement” column. (Be sure to zero the scale before weighing)
14) Repeat step 13 with the marked jar, recording in the 3 mg supplement table.
15) Analyze the data.

Data: Tables / Graphs

Table 1: Raw Data Table for the Masses of Mung Bean Sprouts Grown With or Without a Melatonin Supplement (grams)

Trial	No Supplement	3mg Melatonin Supplement
1	0.14	0.21
2	0.26	0.27
3	0.20	0.22
4	0.21	0.19
5	0.23	0.15
6	0.21	0.19
7	0.27	0.18
8	0.19	0.20
9	0.16	0.26
10	0.17	0.24
11	0.15	0.17
12	0.17	0.16
13	0.24	0.19
14	0.29	0.19
15	0.21	0.22
16	0.20	0.18
17	0.19	0.18
18	0.22	0.18
19	0.20	0.13
20	0.17	0.14

Table 2: Summative Data Table for the Masses of Mung Bean Sprouts Grown With and Without Melatonin Supplement (grams)

	No Supplement	3mg Melatonin Supplement
Mean	0.204	0.193
Median	0.200	0.190
Mode	0.200	0.190
Standard Deviation	0.040	0.037
Variation	0.002	0.001
n	20	20

Figure 1: Bar Graph of Mean Masses From No Supplement and 3mg Melatonin Supplement Groups

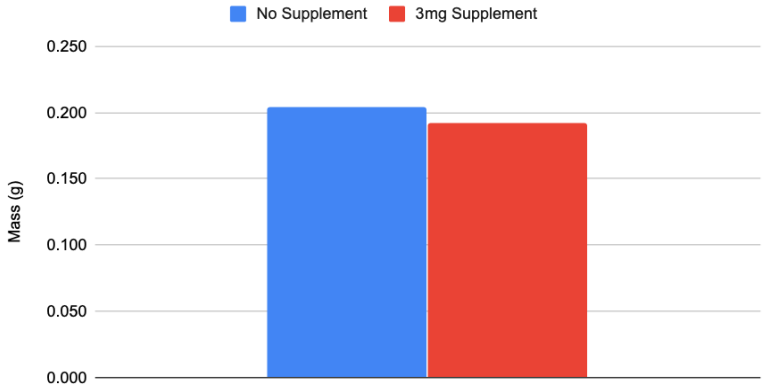
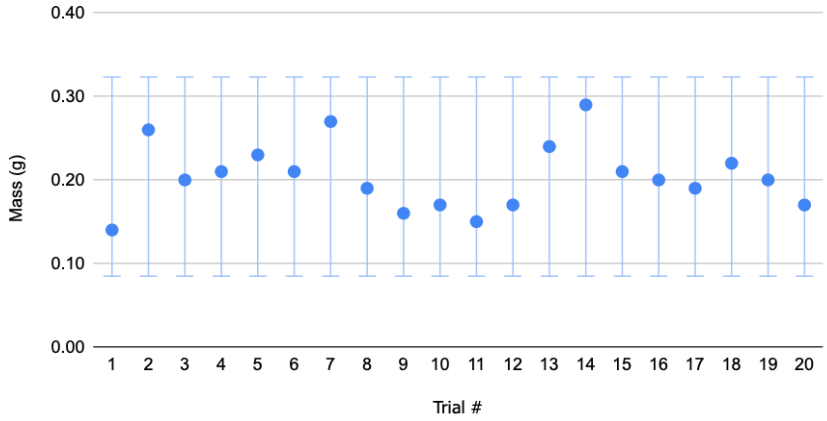
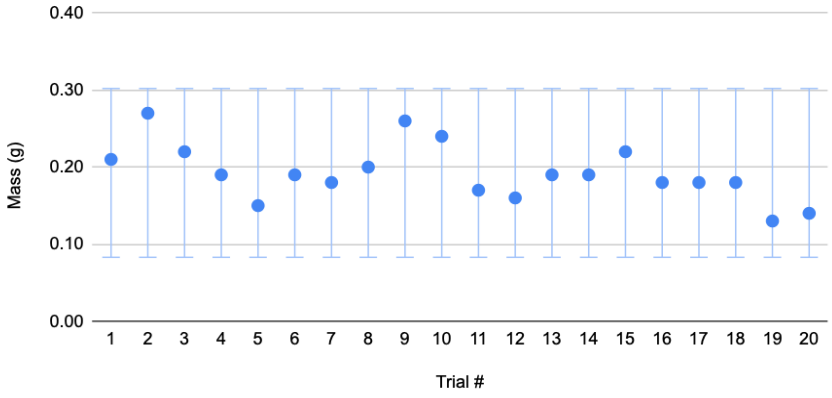


Figure 2: Scatter Plot of the Raw Data for the Masses of Mung Bean Sprouts with No Supplement with Y-Error Bars



The data in **Figure 2** contains no outliers.

Figure 3: Scatter Plot of the Raw Data for the Masses of Mung Bean Sprouts with 3mg Melatonin Supplement with Y-Error Bars



The data in **Figure 3** contains no outliers.

Inferential Statistics

A t-test was used to obtain the p-value of the data because a t-test quantifies the differences between the two sets of data. The type of t-test used was an independent 1-tailed t-test. The subjects in the experiment were independent from one another, so the t-test was independent. The t-test was used to find whether there was a significant increase in the mass of the mung beans, so it is one tailed.

The two sets of data used for the t-test were the raw data for the masses of mung bean sprouts grown with a melatonin supplement and the raw data for the masses of mung bean sprouts grown without a melatonin supplement.

The p-value obtained was 0.1736.

The level of significance used was 0.05.

Findings: Discussion of Statistical Results

Do not support the alternative hypothesis.

Fail to reject the null hypothesis.

The data do not support that mung beans grown in 100 mL of water with a 3 mg melatonin dose will have significantly higher growth (by mass) compared to mung beans grown in only 100 mL of water.

The p-value of 0.1736 is greater than the alpha value of 0.05, which means that more than 5% of the data was similar. Because the p-value exceeds the established alpha value, the alternate hypothesis cannot be supported by the data.

Findings: Discussion of Research Results

The results of this experiment show that the addition of melatonin to the mung beans during growth did not significantly increase their growth compared to the mung beans grown in only water. This is not the result that was expected prior to the experiment. Based upon information from other studies and sources, it was expected that the mung beans given the melatonin supplement would have significantly more growth (by mass). One variable which may have affected the results of the experiment was the length of the growth period. The allotted 10 day time frame, though used in similar experiments, may not have been enough time for the melatonin to reveal its full effects on the mung beans or enough time for the mung beans to process the melatonin. Another variable which may have affected results is the dosage of melatonin. The chosen 3 mg dosage may have provided an insufficient amount of melatonin molecules for the mung beans to be affected. On the other hand, the chosen 3 mg dose may have provided too much melatonin to the mung beans, which might alter its effectiveness. Besides those previously stated, there are no obvious variables that may have affected the results of the experiment, as the growing location and conditions of the two groups were kept identical, besides the addition of the supplement to the non-control group. From the data gathered in this study, the conclusion that melatonin does not significantly increase growth in mung bean sprouts is accurate.

Suggestions for Further Study

The experiment conducted in this study took place over a ten day period, which may have been insufficient time for the melatonin to have its full effects on the mung beans or for the mung beans to exhibit any effects the melatonin may have had. It is suggested that in further study, the growth period of the mung bean sprouts is increased. Additionally, the dosage of melatonin used for the non-control group can be adjusted to higher or lower amounts. It is suggested that in further study, different melatonin dosages from those given in this study are given to the non-control group. Another suggestion in this respect is the addition of more levels to the independent variable, allowing a larger variety of dosages to be tested against the control group.

Works Cited

Arnao, Marino B., Hernández-Ruiz, Josefa. “Melatonin: plant growth regulator and/or biostimulator during stress?” Trends in Plant Science, vol. 19, no. 12, December 2014, pp. 789 - 797.

This journal article describes multiple experiments on plants involving melatonin which were used to determine what effects melatonin has on the plants in terms of growth under stress.

Bauer, Brent A. “Pros and Cons of Melatonin.” *Mayo Clinic*, Mayo Foundation for Medical Education and Research, 10 Oct. 2017, www.mayoclinic.org/healthy-lifestyle/adult-health/expert-answers/melatonin-side-effects/faq-20057874.

This website provides information on the use of and potential side effects and risks of taking melatonin supplements which were used to assess the risk of using melatonin in the experiment and to describe melatonin’s use for humans.

“Fertilisers and the Environment.” NSW Department of Primary Industries, NSW Government, 1 Jan. 1970, www.dpi.nsw.gov.au/agriculture/soils/improvement/environment.

This website provides information on the environmental impact of the use of many types of fertilizers and specific ways of avoiding damage to the environment.

Hardeland, Rüdiger, et al. “Melatonin.” The International Journal of Biochemistry & Cell Biology, vol. 38, no. 3, Mar. 2006, pp. 313 - 316.

This journal article explains the presence of melatonin in a multitude of organisms and its behavior.

Janas, Krystyna Maria, and Małgorzata Maria Posmyk. “Melatonin, an Underestimated Natural Substance with Great Potential for Agricultural Application.” Acta Physiologiae Plantarum, vol. 35, no. 12, 2013, pp. 3285–3292., doi:10.1007/s11738-013-1372-0.

This journal article provides a comprehensive analysis of melatonin’s presence and roles in plant growth and served to validate the legitimacy of this experiment.

Moran, Maggie. “How to Grow Bean Sprouts Indoors.” *WikiHow*, WikiHow, 29 Mar. 2019, www.wikihow.com/Grow-Bean-Sprouts-Indoors.

This website provides detailed instructions on the cultivation of mung beans indoors and served as a model for the procedure of the experiment to help ensure that the mung beans were grown in the proper conditions.

N., Euphrasie. “The Effect of Caffeine on Plant Growth | Science Project.” *Education.com*, Education.com, 14 Apr. 2011, www.education.com/science-fair/article/effect-caffeine-plant-growth/.

This website provides a complete description of a similar experiment with the use of mung beans and the measurement of growth with an additive.

Reiter, Russell J., et al. “Melatonin in Plants.” Nutrition Reviews, vol. 59, no. 9, September 2001, pp. 286 - 290.

This journal article details melatonin’s presence in plants and summarizes many studies which have been done regarding it.