How Gibberellic Acid Affects the Growth of *Arabidopsis Thaliana*

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Abstract

Gibberellic acid is a growth hormone found in plants and fungi that works particularly on cell elongation and growth in height. The purpose of the experiment was to find the optimal amount of this growth hormone a plant would need, and if there is such a thing as “too much”. We predicted that there is a moderate concentration of gibberellic acid the plant can be exposed to in order to optimize its growth. In order to determine this, we measured the growth in length of six separate groups of *Arabidopsis thaliana* with varying concentration levels of GA after two weeks of being sedentary.

The significance of this experiment and findings can be found within the amount of hormone in the plants, meaning a hyperactive production of gibberellin could harm a plant just as much as a hypoactive one. A key finding of ours was that the highest concentration of gibberellic acid was not the highest growth yielding experimental group. In response to this, our conclusion is that there is a statistical significance in our 0.8 mL experimental group being the highest growing plant group. Overall, our results correctly matched our hypothesis.

Introduction

Arabidopsis thaliana is a flowering plant popular for use in biological modeling for its quick life cycle and ability to thrive without extensive management or specific needs (Meinke et al. 1998). Because of these unique qualities, *Arabidopsis thaliana* is an obvious choice for testing how certain amounts of plant hormones, chemicals that regulate plant cell processes, affect plant growth (Springer Nature 2019). One plant hormone known for encouraging seed germination by destroying RGL2, a protein-coding gene which suppresses germination in a seedling, is gibberellic acid (Piskurewicz et al. 2008).

Gibberellic acid is extremely common in biological use but especially in the agricultural industry with its unparalleled versatility, resilience in application, and ability to provoke seed germination. With this knowledge of gibberellic acid as a model organism and its effects on promoting seed growth, we can test the research question, What is the optimal amount of gibberellic acid introduced to *Arabidopsis thaliana* that will yield the most growth in the plant? While we know plant hormones can promote growth in *Arabidopsis thaliana*, if the plant is exposed to too much acid then the hormone could build up in the plant’s tissue and stunt its growth instead. This is important to understand while studying our research question in order for us to appropriately predict what quantities will be necessary to test as well as what could affect our results if the plants do not produce the growth you might expect. Therefore, we predicted there is a moderate concentration of gibberellic acid the plant can be exposed to in order to nurture its growth without overdoing it.

Materials and Methods

To discover the optimum amount of gibberellic acid an Arabidopsis *thaliana* plant can be exposed to in order to develop the most in length, we set up a thorough and well-organized experiment. First we obtained six sterile petri dishes and labeled the bottom of each dish with our names, date, section number, and most importantly, treatment type. Then we gathered six tubes of *Arabidopsis thaliana* seeds for each of the petri dishes. We proceeded to add 1mL of either gibberellic acid, sterile water, or a combination of the two depending on the treatment type to the seeds in their tubes.

Our six treatments were 0mL of gibberellic acid and 2mL of water (0uM/mL GA), .2mL of acid and 1.8 mL of water (28 uM/mL GA), .4mL of acid and 1.6mL of water (56 uM/mL GA), .6mL of acid and 1.4mL of water (84 uM/mL GA), .8mL of acid and 1.2mL of water (112 uM/mL GA), and finally equal parts of 1mL of acid and 1mL of water (140 uM/mL GA). Afterwards, we vortexed the microcentrifuge tubes and poured them out onto the corresponding dishes. Next, we spread the seed solutions around the surfaces using a sterile loop (using a new sterile loop for each plate), then placing the lids on. The final concentrations of gibberellic acid in each plate were 0uM/mL for the 0mL GA treatment, 1.4 uM/mL GA for the .2mL treatment, 2.8 uM/mL GA for the .4mL treatment, 4.2 uM/mL GA for the .6mL, 5.6 uM/mL GA for the .8mL treatment, and lastly, 7 uM/mL GA for the 1mL treatment.

After taping the dishes shut with surgical tape, we then placed the plates in a controlled environment consisting of adequate and equal access to sunlight, keeping them at room temperature, and leaving them taped and untouched as we made sure to observe and take photos of them over the two-week observation period. Once the two-week period was over, we brought the dishes back and took all of the seedlings out of the dishes.

In order to collect thorough and complete data of all the seedlings and their total lengths after the growing period, we placed the seedlings from each dish onto separate pieces of pink construction paper. Then, using Leica, we took pictures of each piece of paper with a ruler on it and uploaded them to Image J to acquire accurate measurements, in centimeters, of each individual seedling in its entirety in every treatment, as well as the average length of all the seedlings in each dish.

For analysis, we ran multiple T-tests to look for a statistical significance in our data. The tests we ran were between 0 mL and 1 mL, 0 mL and 0.8 mL, and 0.2 mL and 0.8 mL. 0.8 mL yielded our highest growth on average, 0.491 cm. 1 mL had our highest amount of gibberellic acid, 0 mL was our control, and 0.2 mL was our first experimental group. It seemed necessary to produce a T-test for all combinations because of our hypothesis that the optimal amount would be a moderate concentration of gibberellic acid. The type of T-test we ran was unpaired and 2-tailed. Unpaired because it was 2 different data sets with a different variable, and 2-tailed because we needed to see both ends of the data, in 2 directions, rather than just one side of the data.

Data and Results

T-test results (Table 1)

Experimental Groups	0ml to 1ml (control group and largest concentration)	0 ml to 8ml (control group and largest amount of growth)	0.2ml to 0.8ml (smallest amount of experimental group growth and largest amount of experimental group growth)
T-test Result (p value)	0.175606	0.01521	0.011184
Support or Rejection of the Null Hypothesis	Support	Reject	Reject

Figure 1

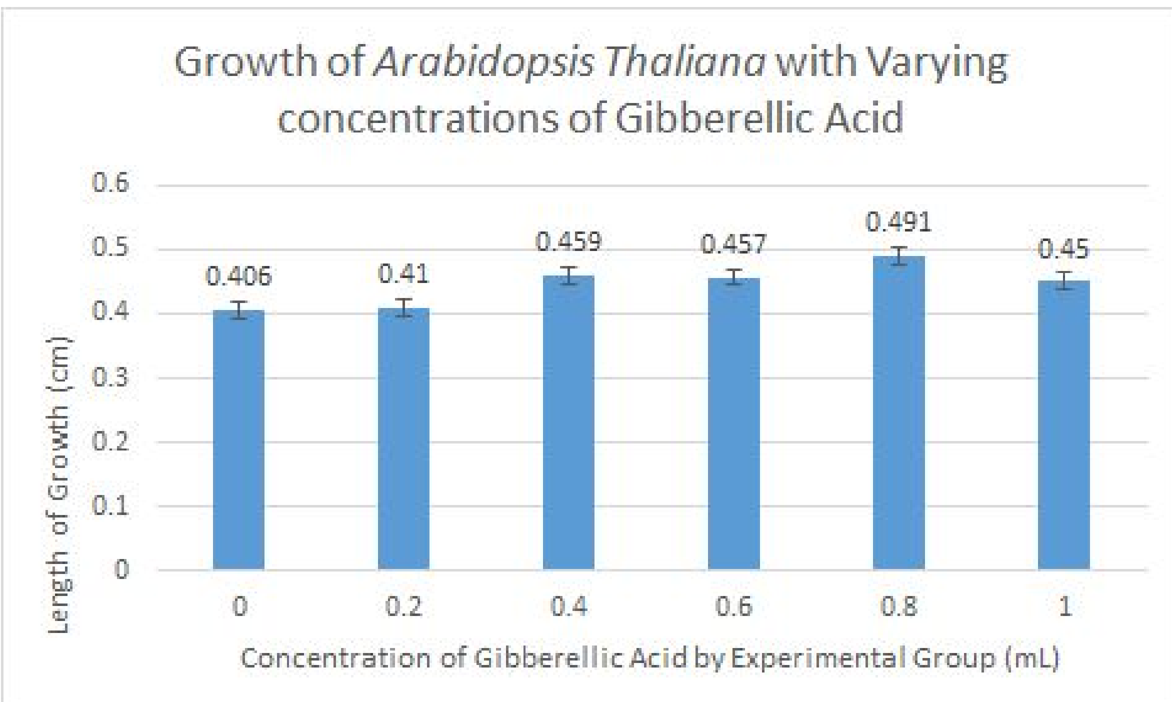

Figure 1: The figure above shows the average length of growth, in centimeters, of each sample group of *Arabidopsis Thaliana* after two weeks. As you can see, the .8mL GA treatment group (5.6 uM/mL) of *Arabidopsis Thaliana* produced the highest length in growth among the six treatment groups, .491cm. Due to the fact that the bar heights generally increase up until the .8mL group and then decrease at the 1mL group, we can infer that the growth of *Arabidopsis Thaliana* is maximized when given a moderate concentration of gibberellic acid and water, or

more specifically, between the concentrations of 2.8uM/mL (.4mL group) and 5.6uM/mL (.8mL group).

The sample size was 20 plants from each different concentration of gibberellic acid. The average growth for each concentration is expressed in the graph above, but is respectively from 0 mL to 1 mL: 0.406, 0.41, 0.459, 0.457, 0.491, and 0.45cm (Figure 1). Standard deviations are as follows, respectively: 0.168, 0.136, 0.105, 0.078, 0.089 and 0.085. We ran multiple T-tests to look for a statistical significance in our data. The tests we ran were between 0 mL and 1 mL, 0 mL and 0.8 mL, and 0.2 mL and 0.8 mL. 0.8 mL yielded our highest growth on average with 0.491 cm (Table 1). 1 mL had our highest amount of gibberellic acid, 0 mL was our control, and 0.2 mL was our first experimental group. The p values are as follows, respectively: 0.175606, 0.01521, 0.011184.

We deemed it necessary to produce a T-test for all combinations due to our initial hypothesis which states the optimal amount would be a moderate concentration of gibberellic acid. As stated previously, the type of T-test we ran was unpaired and 2-tailed.

Discussion and Conclusion

The results of the T-tests showed statistical significance except for one of the T-tests, which was to be expected. The 0 mL to 0.8 mL had a p value of 0.01521, showing a rejection of the null hypothesis. The 0 mL to 1 mL had a p value of .175606, supporting the null hypothesis, which was expected, as it was not the highest growth on average. The 0.2 mL to 0.8 also had a p value that rejected the null hypothesis, which came out at 0.011184. This type of analysis shows that the difference in the data is significant because of the p values being below .05, especially between the control and the highest growth yielding experimental group due to its extremely low p value.

In the end, results from this experiment were about as expected. The experimental group with the second highest concentration of gibberellic acid yielded the most amount of growth of *Arabidopsis thaliana.* Our hypothesis of an intermediate experimental group having the most growth appears correct. The lowest amount of growth came from the control, 0mL of GA and 2mL of water (0 uM/mL GA concentration), which was also predicted. Too high and too low of a concentration of gibberellic acid do not support an amount of growth hormone optimal for plant growth. Regardless of the amount of hormone added, the experimental groups had more growth overall than the control. An intermediate amount of gibberellic acid had the most growth, which was also consistent with our hypothesis. This then also begs the question of whether or a very high concentration of the hormone can be harmful to the plant.

According to our T-test between 0 ml and 1 ml, there was no statistical significance in growth, therefore a large concentration of gibberellic acid is not optimal for a plant's growth. As found in a study done by Riley, too high of a concentration will hinder growth in Arabidopsis

Thaliana (Riley JM. 1987). The high concentration overwhelms the plant's capacity to grow and uses the hormone to slow down growth.

The optimal range of GA we captured from our tests was found to be consistently within the 0.4-0.8mL GA mark, or more specifically, concentrations of 2.8-5.6uM/mL GA. This supports the idea, and fact, that gibberellin has regulatory abilities to control the amount of growth hormone the plant has available. These can also inhibit the amount of growth hormone needed due to the fact that too little of gibberellic acid will not produce enough growth for the plant to be at its tallest and too much of the acid will not allow the plant to grow as much as it could in length (National Center for Biotechnology Information).

In the future, we are hopeful that we can use the information we have compiled through our weeks of researching, experimenting, and analyzing to efficiently perform further tests to determine a more precise optimal amount of gibberellic acid to promote growth in *Arabidopsis Thaliana*. While we are confident after unpacking many articles and executing numerous tests that the optimal amount falls between the concentrations of 2.8 uM/mL GA and 5.6 uM/mL GA, using the limits we determined to find an even more precise measurement can put other researchers in a great position to advance their knowledge of Arabidopsis Thaliana growth, its qualifications to be a model organism in the biological field, and most importantly, how GA can affect other similar organisms' growth.

Citations

Bernaola L, Stout M, He J, Chen Q, Xin P, Yuan J, Ma Y, Wang X, Xu M, Chu J et al. 2019. Plant hormones - Latest research and news | Nature. Nature.com. https://www.nature.com/subjects/plant-hormones

"Gibberellic Acid." National Center for Biotechnology Information. PubChem Compound Database, U.S. National Library of Medicine. https://pubchem.ncbi.nlm.nih.gov/compound/Gibberellic-acid.

Meinke DW, Cherry JM, Dean C, Rounsley SD, Koornneef M. 1998. *Arabidopsis thaliana*: A Model Plant for Genome Analysis. Science. 282(5389): 662-682.

Piskurewicz U, Jikumaru Y, Kinoshita N, Nambara E, Kamiya Y, Lopez-Molina L. 2008. The Gibberellic Acid Signaling Repressor RGL_2 Inhibits *Arabidopsis* Seed Germination by Stimulating Abscisic Acid Synthesis and ABI_5 Activity. The Plant Cell. 20(10): 2729-2745.

Riley JM. 1987. Gibberellic Acid for Fruit Set and Seed Germination. CRFG Journal. 19(10-12).

Weinstein R. 2019. Maximizing Gibberellic Acid's Efficiency to Increase Growth. Sciencebuzz.com. https://www.sciencebuzz.com/maximizing-gibberellic-acid-efficiency-to-increase-growth/