# An Analysis of the Effect of Guayakí Yerba Mate on Plant Growth

Arya A., Adrienne C., Annica E., and Kyle G.

aryaa774@gmail.com, adri803cruz@gmail.com, annica0415@gmail.com, kylgms@gmail.com

**Abstract**

Despite the documented negative effects of caffeine on plant growth, gardening experts recommend watering plants with black coffee since coffee contains nutrients also found in plant fertilizers. The objective of our study was to determine whether varying dilutions of another common caffeinated drink, Guayakí Yerba Mate (GYM), would promote shoot growth in green onions. In this experiment, green onions were watered with different dilutions of GYM (25%, 50%, 75%, or 100% concentration) for our treatment groups and tap water for our control group. We hypothesized that if GYM is beneficial for plant growth, then the green onions placed in the 100% GYM solution would have the greatest shoot growth after a two-week period compared to the green onions in the diluted solutions and tap water. The mean shoot growth for each group was as follows: control = 24.2 cm, 25% = 10.13 cm, 50% = 4.96 cm, 75% = 4.12 cm, 100% = 3.02 cm. A one-way ANOVA test followed by a Tukey's multiple comparison test showed that the mean shoot growth of the control group was significantly greater than each of the four treatment groups ($p<0.0001$). In conclusion, our hypothesis was not supported, and we found that GYM negatively affected the growth of the green onion shoots due to the impairment of root growth and function.

## Introduction

Caffeinated beverages have risen in popularity and have become essential in our daily lives, prompting research on their physiological effects on humans and other organisms like plants. Although it has been observed that coffee and coffee grounds have been used to supplement plant growth (Ogden 25), a major component of coffee is caffeine, which has been shown to decrease plant growth (Smyth 125; Mohanpuria and Yadav 293). Therefore, why are methods such as pouring diluted coffee on plants recommended by gardening experts (Tarlton)? Further research suggests that it is potentially due to the nutrients present in coffee, such as calcium, potassium, nitrogen, and phosphorus, which are also commonly found in fertilizers (Grant). Perhaps the beneficial effects of these nutrients outweigh the adverse effects of coffee's

caffeine content. Furthermore, diluting coffee prior to using it to water plants could also decrease the adverse effects of caffeine. While the possible benefits of diluted coffee as a watering solution for plants has been well documented on gardening forums, this led us to wonder about the effect other common caffeinated beverages, such as Guayakí Yerba Mate (GYM), have on plant growth. In comparison, GYM has 150 mg of caffeine per 458 mL, while brewed coffee has approximately 185 mg of caffeine per 458 mL (Mayo Clinic; Weinfeld).

Further research on GYM yielded that, despite its high caffeine content, it also contains components which could positively affect plant growth. For example, GYM contains citric acid, which has been found to increase the root weight of plants and improve photosynthesis, both of which correspond to increased plant growth (Talebi et al. 1; Mallhi et al. 1). Furthermore, yerba mate leaves contain minerals, such as potassium, zinc, copper, manganese, iron, and nickel, which are required by plants for adequate nutrition (Heck and de Mejia 143; White and Brown 1). The leaves also contain amino acids, which can improve photosynthesis, transpiration, and the transportation of nutrients (Khan et al. 6). The purpose of our study is to compare the effects of different dilutions of GYM on plant growth when used as the watering solution. Specifically, we investigate whether any of the GYM dilutions would yield a greater green onion shoot growth over a two-week period when compared to a control group of tap water. Green onions were chosen to model plant growth as they are fast-growing and easy to maintain. Based on our research, we hypothesize that if GYM is beneficial for plant growth, then the green onions placed in 100% GYM will have the greatest shoot growth after two weeks compared to the green onions placed in the diluted solutions and tap water. Our null hypothesis ($H_0$) is that there is no statistically significant difference in the mean shoot growth between the different watering

solutions, while our alternate hypothesis ($H_A$) is that there is a statistically significant difference. We hope our study will elucidate whether other caffeinated beverages in addition to coffee could be used by home gardeners to enhance plant growth.

**Methods**

Bundles of green onions were purchased from local supermarkets in Richmond, B.C., with each group member requiring a total of fifteen green onions. The green onions were washed and then cut 4 cm from the bottom of the bulb, such that the cut pieces retained their roots. The GYM used in this experiment was the "Lemon Elation" flavour and two 458 mL cans were required. There were four treatment groups in our study – 25%, 50%, 75%, and 100% concentrated GYM solutions, and tap water as the control group. Each watering solution group had three green onion samples, which were each placed in a plastic cup that was labelled with the watering solution and sample number (1, 2, or 3). We used clear, medium-sized plastic cups to hold the different watering solutions and green onion samples.

Next, we created 300 mL of each GYM dilution. To create the 25% GYM solution, we measured 75 mL of GYM and added it to a plastic water bottle. We then measured 225 mL of tap water and added it to the same plastic water bottle. The solution was mixed well and the bottle was labelled "25% GYM solution". This process was repeated for the 50% and 75% GYM solutions with the necessary volumes shown in Table 1. Finally, we poured 300 mL of GYM from the can into a plastic water bottle and labelled it "100% GYM". In the end, each group member had four labelled water bottles.

| Treatment Group | Guayakí Yerba Mate (mL) | Water (mL) | Total (mL) |
|---|---|---|---|
| 25% | 75 | 225 | 300 |

| | | | |
|---|---|---|---|
| 50% | 150 | 150 | 300 |
| 75% | 225 | 75 | 300 |
| 100% | 300 | 0 | 300 |

**Table 1.** The amounts of Guayakí Yerba Mate and tap water needed to create 300 mL of 25%, 50%, 75%, and 100% concentrated GYM solutions. Measurements given in mL.

For the treatment groups (25%, 50%, 75%, and 100% concentrated GYM solutions), we measured and poured out 50 mL from the pre-made bottles into the respective plastic cups. The bottles containing the remaining 150 mL of watering solution were capped and placed in the fridge for the following week. For the control group, we added 50 mL of tap water into the respective cups. The solutions were all at room temperature when poured into the cups. Once the labelled cups were filled with their corresponding watering solution, we carefully placed one onion bulb into each of the cups with the roots facing downwards into the watering solution. We ensured that the bulbs were not fully submerged into the solution. The samples were then placed in an area where they had access to sunlight.

Growth was measured for each green onion sample every two days for a period of two weeks. When measuring, we removed the green onions from the solution one by one and measured the length from the bottom of the bulb (not including the root ends) to the top of the shoot using a ruler. In order to find total growth, 4 cm was subtracted from the measured value in order to account for the original length of the green onion (Figure 1A). If multiple shoots began to grow from the green onion, each individual shoot length was measured from the base of the shoot to the tip (Figure 1B). Then to calculate the total growth, the lengths of the additional shoots were added to the length of the pre-existing main shoot and 4 cm was subtracted to account for the original length of the green onion.

After one week, the green onion samples were carefully removed from the cups and the old solution was discarded. Another 50 mL of the respective solution was added to each cup. For the treatment groups, this came from the pre-made refrigerated bottles, which were warmed to room temperature before being added to the plastic cups. For the control group, water was taken from the tap. Once the solutions were replaced, the green onion samples were carefully placed back into the same plastic cup. After raw data were collected for two weeks, statistical analyses of the mean shoot growth between different watering solutions were conducted using a one-way ANOVA test, followed by a Tukey's multiple comparisons test. All statistical analyses and graphs were done using the GraphPad Prism (V9.0.2) statistics tool.

**Figure 1. (A)** Photo of a single shoot of a green onion sample grown in 25% concentrated GYM solution on day 7 of our experiment. Total growth was determined by subtracting 4 cm from the measured length of the single shoot to account for the original length of the green onion. The calculation for this green onion was as follows: 6.5 cm - 4 cm = 2.5 cm. **(B)** Photo of the main shoot and offshoot of a green onion sample grown in tap water on day 7 of our experiment. Total growth was determined by adding the length of the main shoot and offshoot, then subtracting 4 cm to account for the original length of the green onion. The calculation for this green onion was as follows: 9.2 cm + 9.7 cm - 4.0 cm = 14.9 cm.

## Results

In total, 60 green onions were analyzed among the five watering solution groups, with 12 green onions in each group (n = 12). The mean shoot growth for each watering solution group was as follows: Control = 24.2 cm, 25% concentrated GYM solution = 10.13 cm, 50% concentrated GYM solution = 4.96 cm, 75% concentrated GYM solution = 4.12 cm, 100% concentrated GYM solution = 3.02 cm, as shown in Figure 2. The general trend was that mean green onion shoot growth decreased as the GYM concentration of the watering solution

increased. To determine if our data fit the assumptions of an ANOVA, we performed a Shapiro-Wilk test and graphed the data on a QQ plot. Through these methods, we confirmed that the normality assumption of an ANOVA was met. A one-way ANOVA of the data was performed and yielded a p-value of $p<0.0001$. A significance level ($\alpha$) of 0.05 was chosen as this is the standard in biology. We determined which groups had a significant difference in mean shoot growth when compared to one another by performing a Tukey's multiple comparison. The results showed that the mean shoot growth for the control group was significantly greater than each treatment group ($p<0.0001$, Figure 2). Furthermore, the mean shoot growth of the 25% and 75% concentrated GYM solutions were found to be significantly different ($p = 0.0217$). The mean shoot growth of the 25% and 100% concentrated GYM solutions were also found to be significantly different ($p = 0.0041$). Lastly, we found that there were no significant outliers in our data based on our QQ plot and individual histograms.

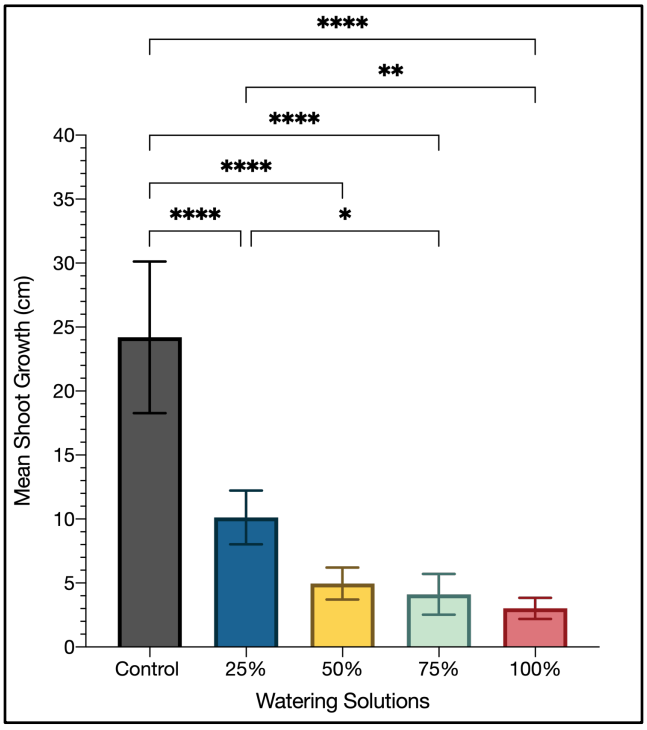



**Figure 2.** The mean green onion shoot growth (in cm) of each watering solution group. Treatment groups were a dilution composed of the percentage of GYM noted and the control group was tap water. Means for each watering solution group were as follows: Control = 24.2 cm, 25% = 10.13 cm, 50% = 4.96 cm, 75% = 4.12 cm, 100% = 3.02 cm. Each group had 12 green onion samples (n = 12). Whiskers represent 95% confidence intervals. Bars with asterisks above show Tukey's pairwise comparisons representing statistically significant differences. Asterisks correspond to the p-values noted: *p = 0.0217, **p = 0.0041, ****p<0.0001.

**Discussion**

The objective of our study was to investigate the effects of different dilutions of GYM on green onion shoot growth when used as the watering solution over a two-week period. Given that a one-way ANOVA of the data yielded a p-value of $p<0.0001$, we reject the null hypothesis ($H_0$) as the p-value is less than our α-value of 0.05. Instead, our data lends support to the alternate hypothesis ($H_A$) that there is a significant difference in mean shoot growth between the watering solution groups. Specifically, since the Tukey's multiple comparison test determined that the mean shoot growth of the control group was significantly greater than each treatment group, GYM is shown to not be beneficial for plant growth. As a result, our original hypothesis that the green onions placed in the 100% GYM solution would have the greatest shoot growth at the end of the two-week period, was not supported.

Though GYM contains components thought to enhance plant growth like citric acid, minerals, and amino acids, any beneficial effects were likely inhibited by the caffeine that is also present. A plant's health and ability to grow is directly influenced by the roots through which water and nutrients are taken up, thus unhealthy roots result in an unhealthy plant (Timmerman et al. 3). Throughout our study, we observed that the green onions placed in the GYM-containing solutions had thicker and shorter roots, which is indicative of an unhealthy plant. This can be explained by the fact that caffeine impedes shoot and root elongation by hindering the function of calcium and Rubisco in plants (Smyth 125). First, caffeine inhibits adenosine receptors that regulate intracellular calcium levels in plant cells. Exposure to caffeine causes a reduction in intracellular calcium, which is required for the proper functioning of the enzyme alpha-amylase. With regards to plant growth, alpha-amylase breaks down starch to release sugars that drive root

and shoot elongation (MacNeill et al. 4437; Jadhav et al. 597). Thus, by indirectly disrupting alpha-amylase function, caffeine hinders plant growth. Second, caffeine decreases the expression and activity of Rubisco, which is an important enzyme for photosynthetic carbon fixation (Yoshikawa 1571). Lower levels of Rubisco are known to induce early senescence and decrease root and shoot size (Mohanpuria & Yadav 294). As a result, caffeine is again described to impede plant growth. Overall, the negative effects of caffeine on plant growth are well documented in other studies. Subsequently, our findings are in line with the literature, as the high caffeine content of GYM could provide an explanation for the statistically significant decrease in mean shoot growth in the treatment groups when compared to the control group.

In our study, there was a source of variation that could have influenced our results as the location that the green onion samples were placed in during the two-week period differed among group members. Specifically, half of the samples were placed on a windowsill with direct sunlight and the other half were placed near a glass door with indirect sunlight. It is well documented that light intensity directly impacts plant growth and development, with plants subject to lower intensities displaying reduced growth (Gao et al. 1). Consequently, the green onions in direct sunlight on the windowsills received greater light intensities compared to the samples placed near the glass doors, which could have influenced their growth.

A potential source of error in our study could have been due to the difficulties measuring curved green onion shoots with a standard ruler. In order to measure the curved shoots, they had to be straightened out or measured in segments along straight stretches. Straightening out the green onions had the potential to damage them and impair their ability to grow. Additionally,

measuring the shoots in segments and summating them is not very accurate. If this experiment were to be repeated, a flexible tape measure could be used.

In the future, we would be interested in repeating our study using decaffeinated GYM to eliminate any of the effects of caffeine. We would then be able to determine if caffeine was the cause of the significant decrease in plant growth that was observed when GYM was present in the watering solution, or if it was perhaps due to another component of GYM. This study could further contribute to the evidence that caffeine negatively impacts plant growth that is documented in the literature.

**Conclusion**

In conclusion, we found that GYM negatively affected green onion shoot growth. As a result, our hypothesis that green onions grown in 100% GYM would have the greatest shoot growth after two weeks was not supported. This can most likely be attributed to the fact that GYM contains large amounts of caffeine, which has been well documented in the literature to decrease plant growth. Overall, our study demonstrated that GYM is not an appropriate watering solution for use by home gardeners on their plants, even when diluted with water.

**Acknowledgements**

We would like to thank our BIOL 342 professor, Dr. Celeste Leander, for providing us with the necessary information and continued support to develop and achieve our project. We would also like to thank our BIOL 342 Teaching Assistants, Jordan Hamden, Tessa Blanchard and Sofya Langman, for providing useful feedback throughout the progression of our project. Lastly, we would like to thank the University of British Columbia for the opportunity to take this

course and pursue our research ambitions. We would like to acknowledge that our work took place on the traditional, ancestral, and unceded territory of the Musqueam people.

## Works Cited

Gao, Song, et al. "Comparison of the effects of LED light quality combination on growth and nutrient accumulation in green onion (Allium fistulosum L.)." *Protoplasma* 2021, pp. 1-11, doi:10.1007/s00709-020-01593-y.

Grant, Amy. “Diluted Coffee For Plants: Can You Water Plants With Coffee.” *Gardening Know How*, www.gardeningknowhow.com/garden-how-to/soil-fertilizers/diluted-coffee-for-plants.htm.

Heck, Caleb I., and Elvira G. de Mejia. “Yerba Mate Tea (Ilex Paraguariensis): A Comprehensive Review on Chemistry, Health Implications, and Technological Considerations.” *Journal of Food Science*, vol. 72, no. 9, 2007, pp. R138-51, doi:10.1111/j.1750-3841.2007.00535.x.

Jadhav, Machindra, et al. "Study on effect of caffeine on growth of Vigna radiate L." *International Journal of Advanced Research*, vol. 4, no. 3, 2016, pp. 596-602.

Khan, Shumaila, et al. “Exogenous Application of Amino Acids Improves the Growth and Yield of Lettuce by Enhancing Photosynthetic Assimilation and Nutrient Availability.” *Agronomy*, vol. 9, no. 5, 2019, pp. 266, doi:10.3390/agronomy9050266.

MacNeill, Gregory J., et al. “Starch as a Source, Starch as a Sink: The Bifunctional Role of Starch in Carbon Allocation.” *Journal of Experimental Botany*, vol. 68, no. 16, 2017, pp. 4433–4453, doi:10.1093/jxb/erx291.

Mallhi, Zahid I., et al. “Citric Acid Enhances Plant Growth, Photosynthesis, and Phytoextraction of Lead by Alleviating the Oxidative Stress in Castor Beans.” *Plants*, vol. 8, no. 11, 2019, doi:10.3390/plants8110525.

Mayo Clinic. “Caffeine Content for Coffee, Tea, Soda and More.” *Mayo Clinic*, 2017, www.mayoclinic.org/healthy-lifestyle/nutrition-and-healthy-eating/in-depth/caffeine/art-20049372.

Mohanpuria, Prashant, and Sudesh K. Yadav. "Retardation in Seedling Growth and Induction of Early Senescence in Plants upon Caffeine Exposure is Related to its Negative Effect on Rubisco." *Photosynthetica*, vol. 47, no. 2, 2009, pp. 293-297, doi:10.1007/s11099-009-0045-0.

Ogden, Lisa. “Plants with Perk: Using Coffee Pot Leftovers Delivers a Java Jolt and Fertile Grounds.” *Barnyards & Backyards*, 2017, pp. 25-27.

Smyth, Douglas A. (Pequest River Laboratory, Belvidere, NJ). "Effect of Methylxanthine Treatment on Rice Seedling Growth." Journal of Plant Growth Regulation, vol. 11, no. 2, 1992, pp. 125-128, doi:10.1007/bf00198025.

Talebi, Majid, et al. “Foliar Sprays of Citric Acid and Malic Acid Modify Growth, Flowering, and Root to Shoot Ratio Of Gazania (Gazania Rigens L.): A Comparative Analysis by ANOVA and Structural Equations Modeling.” *Advances in Agriculture*, vol. 2014, 2014, pp. 1–6, doi:10.1155/2014/147278.

Tarlton, Amanda. “Here’s Why You Should Be Watering Your Plants With Coffee.” *Reviewed,* https://www.reviewed.com/home-outdoors/features/why-you-should-water-your-plants-with-coffee.

Timmerman, Amy D., et al. *Common Signs and Symptoms of Unhealthy Plants EXTENSION*., 2014.

Weinfeld, David. “Our Product Family.” *Guayaki Yerba Mate*, guayaki.com/our-product-family/.

White, Phillip J., and Powel H. Brown. “Plant Nutrition for Sustainable Development and Global Health.” *Annals of Botany*, vol. 105, no. 7, 2010, pp. 1073–1080, doi:10.1093/aob/mcq085.

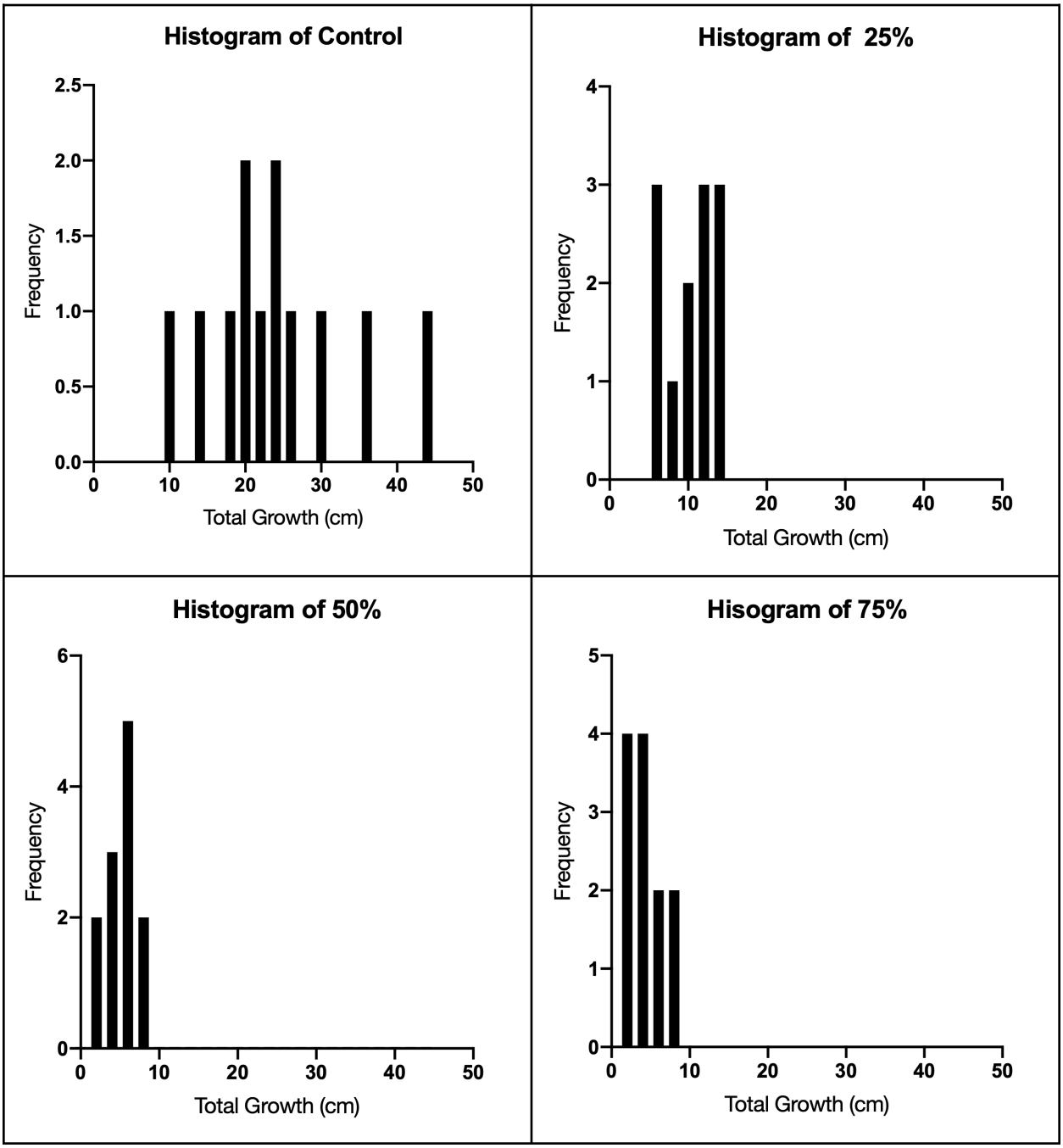
Yoshikawa, Masaaki. *Handbook of Biologically Active Peptides*. Edited by Abba J Kastin, 2nd ed., Oxford, Academic Press, 2013.

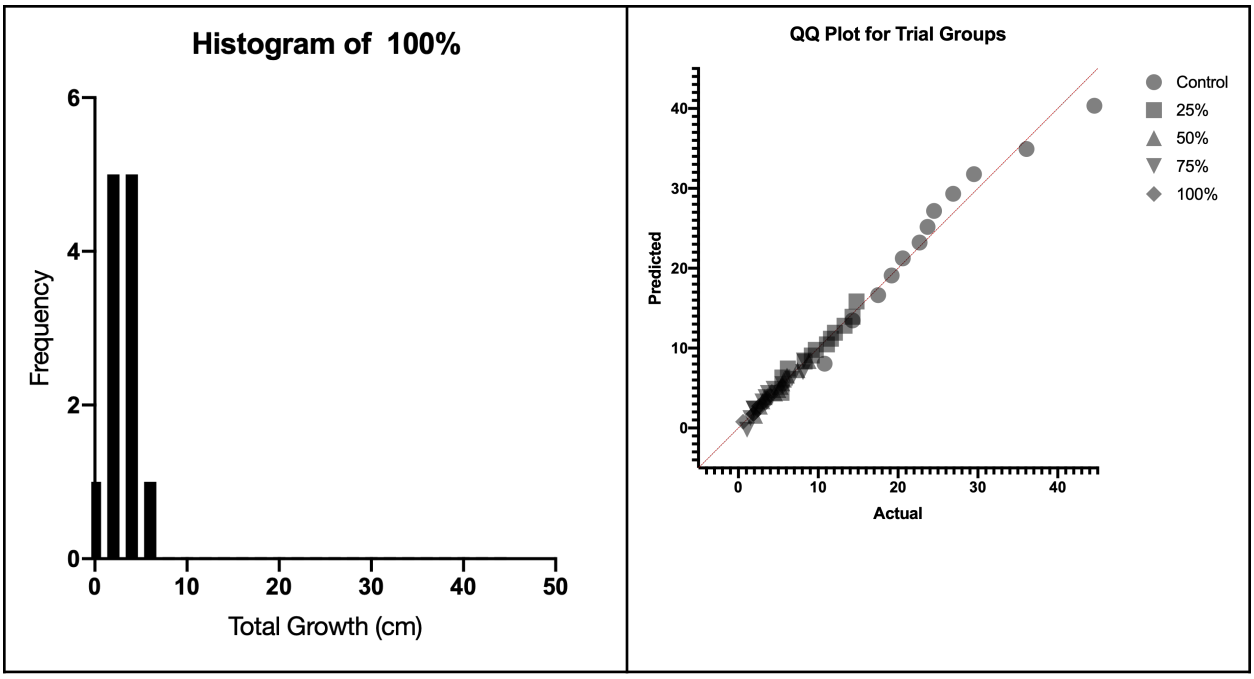
**Appendix A - Raw Data**

**Total green onion shoot length in centimeters (cm)**

| Control | 25% Concentrated GYM | 50% Concentrated GYM | 75% Concentrated GYM | 100% Concentrated GYM |
|---|---|---|---|---|
| 24.5 | 6.2 | 2.7 | 3.7 | 4.2 |
| 17.5 | 5.4 | 3.3 | 5.5 | 3.5 |
| 29.5 | 5.5 | 5.5 | 3.0 | 4.0 |
| 26.9 | 11.1 | 8.8 | 6.7 | 5.5 |
| 22.7 | 14.8 | 5.0 | 8.1 | 2.4 |
| 36.1 | 12.1 | 6.1 | 1.9 | 3.6 |
| 19.2 | 13.3 | 2.1 | 1.9 | 0.6 |
| 10.8 | 8.3 | 5.3 | 1.1 | 2.6 |
| 14.3 | 11.6 | 3.0 | 1.5 | 1.9 |
| 20.6 | 9.2 | 7.4 | 4.4 | 2.6 |
| 23.7 | 9.7 | 5.7 | 3.4 | 1.9 |
| 44.6 | 14.3 | 4.6 | 8.2 | 3.4 |

**Appendix B - Supplementary Graphs**




Histogram of 100%
Frequency
Total Growth (cm)
0
2
4
6
0
10
20
30
40
50
QQ Plot for Trial Groups
Predicted
Actual
Control
25%
50%
75%
100%