Maggie Lin, Isabel Yang, Isabelle Yung FLHS Science Research
Final Paper

The Effect of Ghost Shrimp (*Palaemonetes paludosus*) Aquaponics on the Cultivation of Watercress (*Nasturtium officinale*)

Abstract

Aquaponics is the integration of hydroponic plant production into recirculating fish aquaculture systems. In aquaponics, nutrients excreted from the fish are absorbed by the plants hydroponically. Ghost shrimp (*Palaemonetes paludosus*) is a freshwater crustacean that is primarily clear in color. Ghost shrimp produce nutrients not only through their feces, but also through their burrows. Ghost shrimps are edible and are rich in fat, protein, and calcium. They also contain traces of vitamin B & E, magnesium, and antioxidants. Watercress (*Nasturtium officinale*) is an excellent source of the antioxidants, vitamins A and C, as well as vitamin K. It is also a rich natural source of lutein and zeaxanthin, two carotenoid nutrients that are gaining attention for their ability to protect vision and support cardiovascular health. This experiment aims to test if various quantities (0 shrimp, 5 shrimps, 10 shrimps, 15 shrimps) of ghost shrimp would increase the quality and growth of watercress plants in an aquaponic system. Overall, the data illustrated that shrimp wastewater had a significant effect on the cultivation of watercress. Shrimp waste water significantly increased the fresh weight in watercress plants. In forthcoming works, the inclusion of more trials using larger numbers of ghost shrimp and extending the experiment for a longer period are possible methods to further research and find the best quantity of ghost shrimp that will be beneficial towards watercress growth.

Introduction

Aquaponics

Aquaponics is the integration of hydroponic plant production into recirculating fish aquaculture systems (Nelson, 2008). In aquaponics, nutrients excreted from the fish are absorbed by the plants hydroponically. As the hydroponic component acts as a biofilter, a separate biofilter is negligible (Roosta & Hamidpour, 2011). Furthermore, aquaponics is self-sustained and can be utilized in cities by not only turning waste into nutrients, but also a source of food production (Veludo *et al.*, 2012).

Aquaponics is the combination of aquaculture and hydroponics. Aquaculture is breeding, raising, and harvesting fish, shellfish, and aquatic plants and hydroponics is the cultivation of plants without the use of soil. Aquaponics surpasses conventional agriculture in various ways.

Conventional agriculture is defined as growing crops in soil, open air, with application of nutrients, pesticides, and herbicides. Conventional agriculture requires a high amount of water, large pieces of land, soil degradation and erosion, and it has inefficient water usage. As the population rapidly grows, an alternative method of sustainable agriculture is necessary. Aquaponics provides remedies for these issues. Aquaponics utilizes nutrient-rich water instead of chemical fertilizers; fertile land is not needed, so it can be utilized in locations where conventional agriculture is unfeasible; vertical farming is applicable, thus increasing the yield of an area unit; and if practiced in a controlled environment, it can be a sustainable food source (AlShrouf, 2017 & Barbosa, 2015).

Ghost Shrimp

Ghost shrimp (*Palaemonetes paludosus*) is a freshwater crustacean that is primarily clear in color (Robert, 2022). Ghost shrimp is an important bioturbation, commonly found in muddy and fine sandy intertidal and shallow subtidal coastal sediments, where it often creates dense monospecific populations. Ghost shrimp increases the organic matter content in the sediment and benthic metabolism by incorporating organic detritus, such as seagrass debris, in burrow chambers and consolidates the burrow walls with mucus (Papaspyrou *et al.*, 2005). Ghost shrimp produce nutrients not only through their feces, but also through their burrows.

Ghost shrimps are edible and are rich in fat, protein, and calcium. They also contain traces of vitamin B &E, magnesium, and antioxidants. However, they are not a common food source around the world. Although ghost shrimp is nutrient rich, it is not a sufficient nutrient source alone (UFK, 2021). Regardless, it can be a new food source added to one's diet. Nutrients in shrimps are important for plants as it provides beneficial microorganisms with rich compounds suitable for their growth.

Watercress

Watercress (*Nasturtium officinale*) is an excellent source of the antioxidants, vitamins A and C, as well as vitamin K. It is also a rich natural source of lutein and zeaxanthin, two carotenoid nutrients that are gaining attention for their ability to protect vision and support cardiovascular health. Watercress have been shown to have anticancer effects, as they promote the level of antioxidants in the blood and to protect DNA against damage (Tsoukanelis, 2007). Watercress is also rich in bioactive compounds, named polyphenols, glucosinolates and Phenethyl isothiocyanate (PEITC) (Clemente *et al.*, 2019).

Hypothesis

In this study, it was hypothesized that the quantity of 15 ghost shrimp would be most beneficial to watercress quality and growth. Since larger quantities of shrimp produce an increased amount of nutrients, the watercress will be fertilized further than compared to a lower quantity of shrimp.

Justification

By testing the effects of various amounts of waste produced by ghost shrimp on the growth rate of watercress, it is possible to generate a feasible and inexpensive method to hasten the growing process of watercress. Watercress plants are a part of many individuals' diets as they come with many health benefits. Because of the many benefits that come with consuming watercress, this study aims to determine a method to produce a way to grow watercress at a faster rate. Watercress plants contain great amounts of antioxidants, vitamins A and C, as well as vitamin K. Additionally, the consumption of watercress also provide a rich natural source of lutein and zeaxanthin; these carotenoid nutrients have been proven to gain attention for their ability to protect vision and support cardiovascular health. Furthermore, watercress has anticancer effects, as they promote the level of antioxidants in the blood, which protects DNA against damage (Tsoukanelis, 2007).

Utilizing shrimp in aquaponic systems have proved to be beneficial. As shrimp are one of the preferred seafood items globally, the usage of shrimp in aquaponic systems is logical. Shrimp aquaculture generates significant nutrient-rich wastewater. Generally, shrimp excretia is released into streams and rivers causing effluent environmental concerns. However, shrimp excretia can be a nutrient source for plants. Shrimp wastewater retains a similar nutrient supply as vermicompost and can be utilized in low nutrient demanding plants (Zheljazkov *et al.*, 2011). Furthermore, research states that inoculating probiotics in conjunction with pacific white leg shrimp aquaponics, has the efficiency of stabilizing water quality, cultivating microbial communities, and enhancing the health of shrimp and plants in the system (Chu & Brown, 2021). Although many types of shrimp were tested in aquaponics, ghost shrimp have not been tested before.

Although other studies have tested shrimp aquaponics with cultivation of lettuce (de Farias *et al.*, 2019), previous studies have not tested the cultivation of watercress with shrimp aquaponics. Furthermore, other studies have not utilized ghost shrimp, instead they used pacific white leg shrimp (Chu & Brown, 2021). Fish aquaponics is more commonly used, so further research on shrimp aquaponics is needed. When combining these research bases, it can be further explored on how shrimp aquaponics can be an effective method of cultivation.

Materials and Methods

Table 1- Experimental Design

Groups	Number of shrimps	Number of plants	Tank Volume (L)	Hydroponic Volume (L)	Feed (g)
a (control)	0	10 (2x5)	2	1	1
b	5	10 (2x5)	2	1	1
c	10	10 (2x5)	2	1	1
d	15	10 (2x5)	2	1	1

Aquaponic system design:

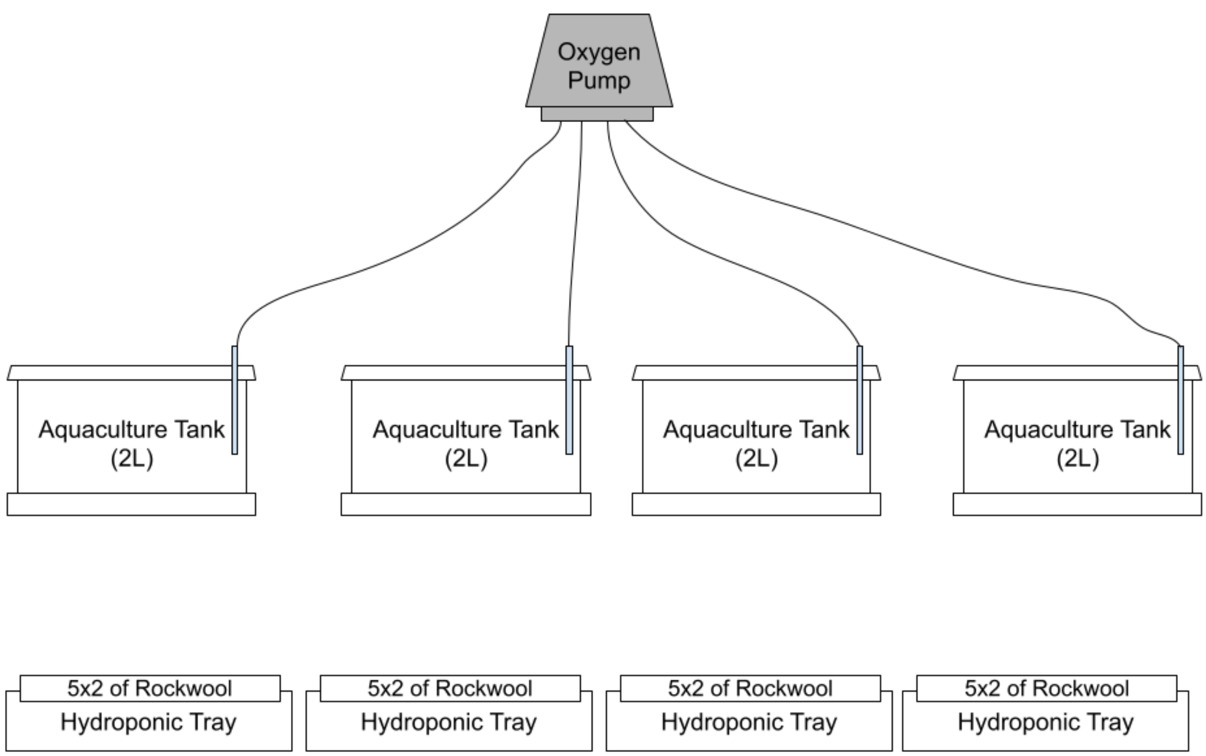

Figure 1- Diagram of Aquaponic System Design (Created by Isabelle Yung).

Four aquaponic systems were set up according to Figure 1. Each aquaponic system was equipped with a 2 L aquaculture tank. Each aquaculture tank was equipped with an oxygen pump, pumping oxygen into the tank. All tanks were kept in the dark to ensure a habitable water temperature.

Shrimp Culture:

Ghost shrimps were ordered from Carolina Biological Supply. After arrival, the ghost shrimps were quarantined for a 1-week period before being transported into the aquaponic systems.

Shrimp Habitat:

The water temperature was set to 21℃-26℃. Black sand was placed at the bottom of the tank. 2 L of deionized water was added to the tank. Every day in the afternoon, the ghost shrimps were fed 1 g of commercial shrimp feed.

Watercress Materials:

Groups a, b, c, and d were connected to tanks with shrimps according to Table 1. In each tray, watercress seeds were planted in a 2x5 rockwool. The rockwool acted as the hydroponic substrate for the plants. Germination of the plants was recorded during the 5-day germination period. The plants were taken out daily to assess their length and height after the germination period. On day 14 the post-experimental weight was assessed. The data collected was recorded on excel.

Control

Group a was connected to a tank without ghost shrimp according to Table 1. In each tray, watercress seeds were planted in a 2x5 rockwool. The rockwool acted as a hydroponic substrate for the plants. Germination of the plants was recorded during the 5-day germination period. The plants were taken out daily to assess their length and height after the germination period. On day 14 the post-experimental weight was assessed. The data collected was recorded on excel.

Measuring of Watercress

The height of the watercress was recorded daily after the 5-day germination period. The daily heights and germination process was recorded on excel. The post-experimental weight was weighed after 14 experimental days. The post-experiment weight is the weight of the plant after the plants have gone through the whole experiment.

Data Analysis

The watercress height (from rockwool to top of the plant), fresh weight, germination days, plant survival rate, and shrimp survival rate were measured throughout this experiment. These measurements were recorded and entered in an Excel spreadsheet. Based on the weight measurements observed, the mean measurement for each group was calculated. The group of watercress that has the most mass and length, is the most effective number of shrimps to use to cultivate watercress. Figures 2 and 4 represent the data recorded for plant height. The data was plotted into a line graph. The x axis represented the day number since sowing. The y axis represented the heights (in cm). The data's significance compared to the control group was calculated using ANOVA (Analysis of Variance) followed by Tukey HSD; the website utilized was https://astatsa.com/OneWay_Anova_with_TukeyHSD/. P-values less than 0.05 were significant. The significance between each group was calculated using Tukey HSD. Figures 3 and 5 represent the data recorded for fresh weight. The data was plotted into a bar graph. The x axis represented the different number of shrimp groups. The y axis represented the fresh weight (in grams). Standard deviation was calculated, and error bars were added by the standard function of excel. The data's significance compared to the control group was calculated using an unpaired, one-tailed t test by the standard function of excel. P-values less than 0.05 were significant.

Results

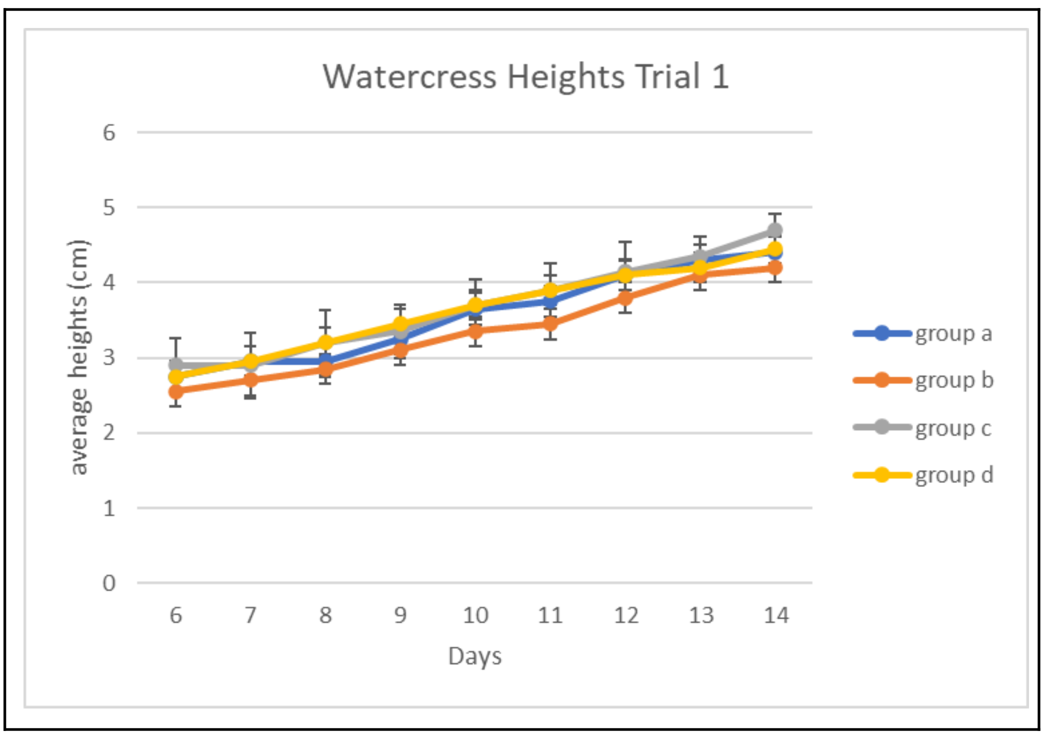

Figure 2- Watercress heights trial 1. Each line represents the average heights of each group. Only days 6-14 are depicted as there was a 5-day germination process. ANOVA followed up by Tukey HSD was conducted to determine the significance of each experimental group compared to the control group.

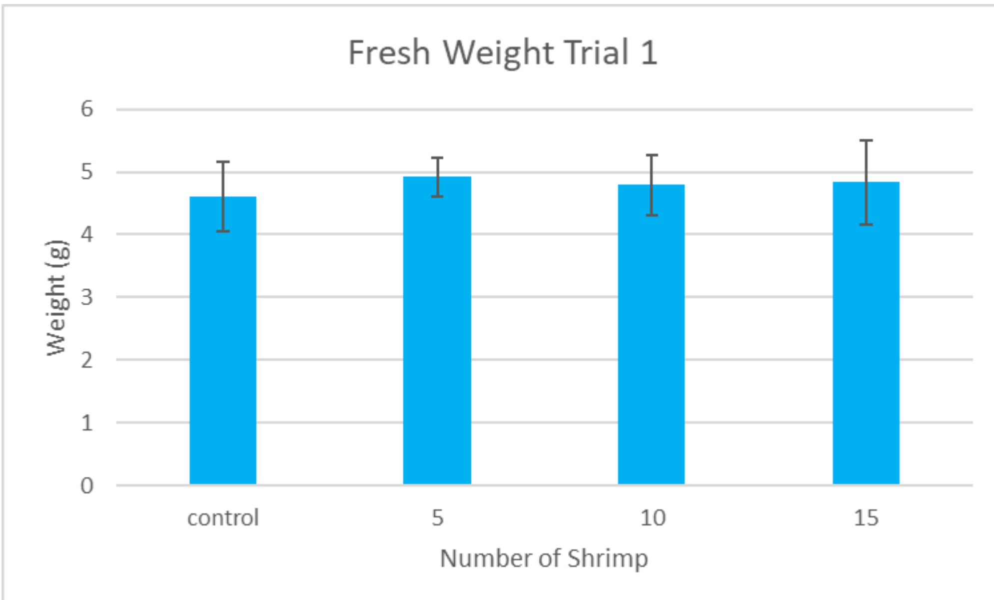

Figure 3- Fresh Weight Trial 1. Each bar represents the average weight of each watercress plant. Error bars represent standard deviation. An unpaired, one-tailed t-test was conducted to determine the significance of each experimental group compared to the control group.

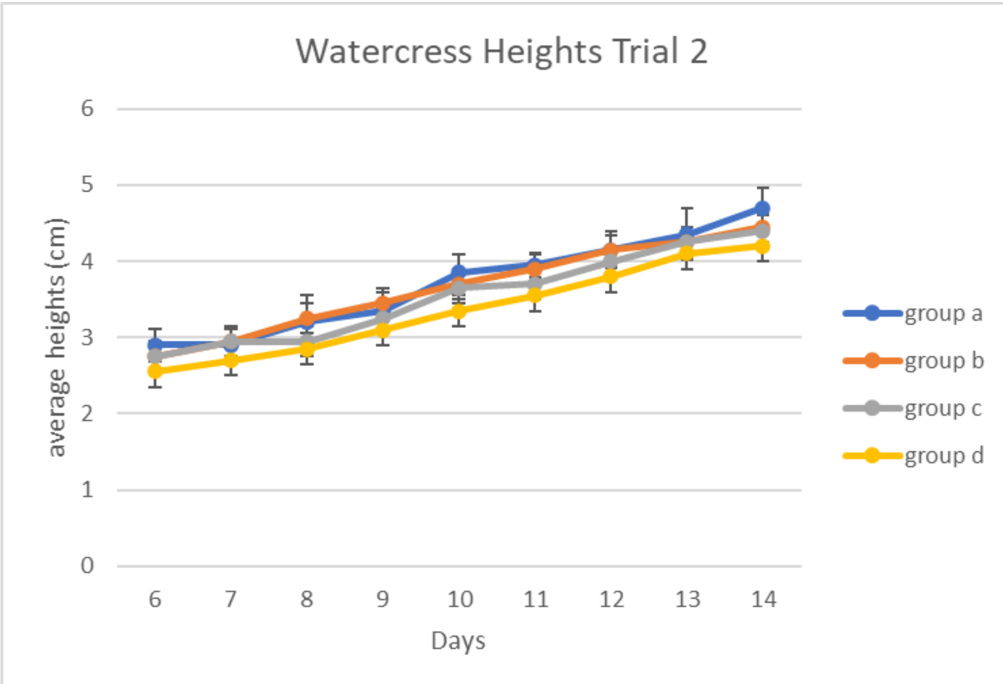

Figure 4- Watercress heights trial 2. Each line represents the average heights of each group. Only days 6-14 are depicted as there was a 5-day germination process. ANOVA followed up by Tukey HSD was conducted to determine the significance of each experimental group compared to the control group.

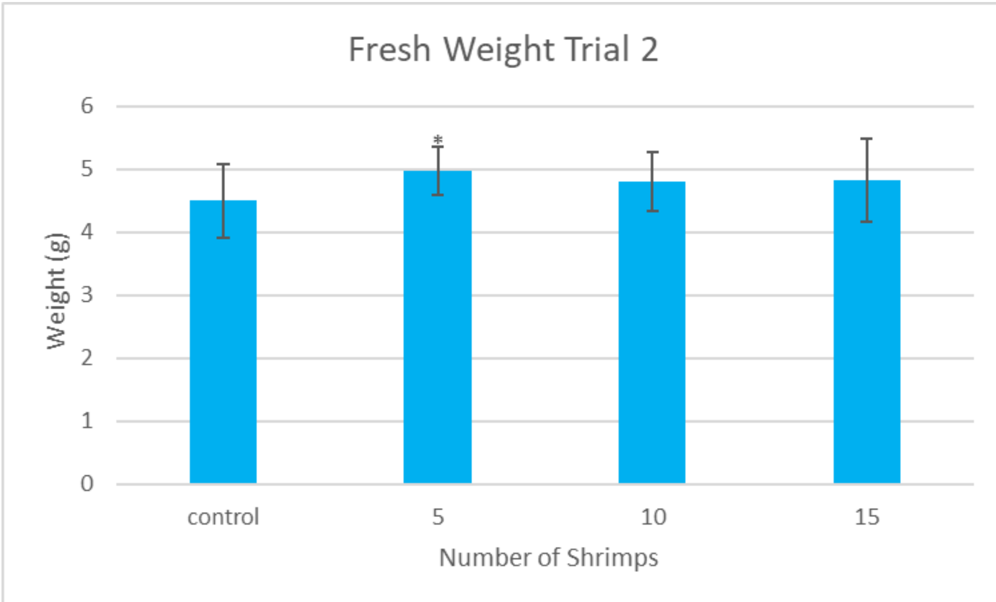

Figure 5- Fresh Weight Trial 2. Each bar represents the average weight of each watercress plant. Error bars represent standard deviation. * Represents $p<0.05$. An unpaired, one-tailed t-test was conducted to determine the significance of each experimental group compared to the control group.

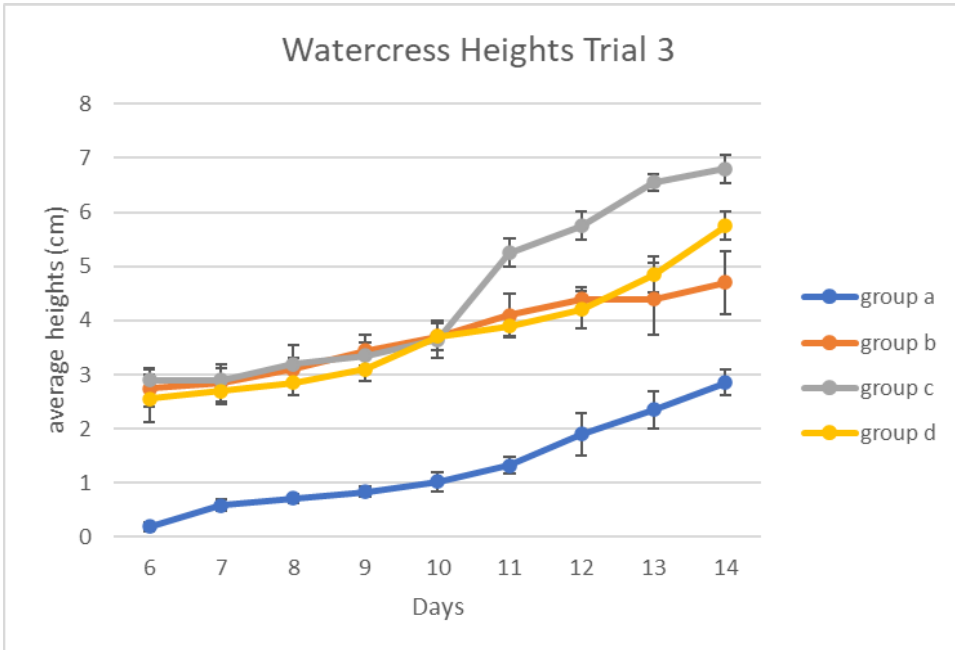

Figure 6- Watercress heights trial 3. Each line represents the average heights of each group. Only days 6-14 are depicted as there was a 5-day germination process. ANOVA followed up by Tukey HSD was conducted to determine the significance of each experimental group compared to the control group.

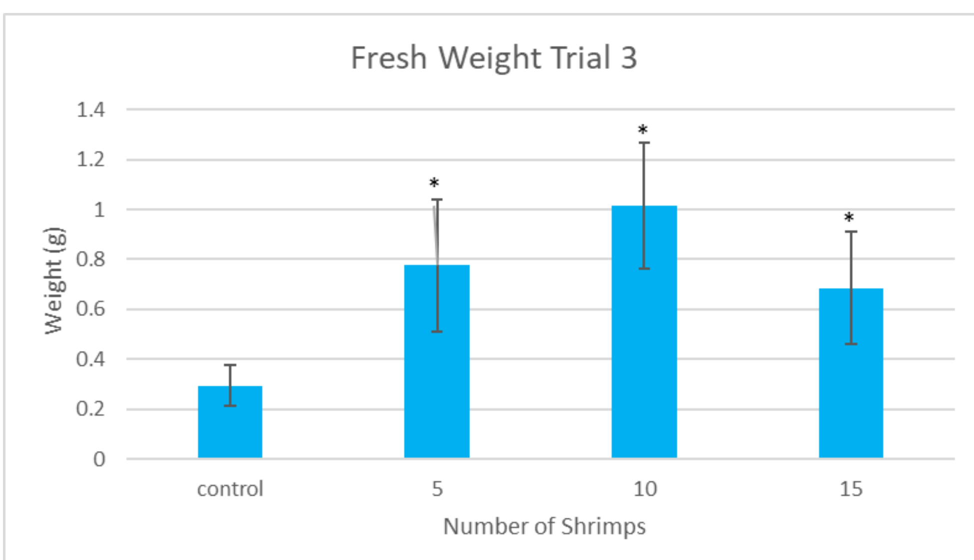

Figure 7- Fresh Weight Trial 3. Each bar represents the average weight of each watercress plant. Error bars represent standard deviation. * Represents $p<0.05$. An unpaired, one-tailed t-test was conducted to determine the significance of each experimental group compared to the control group.

Discussion

Plant Height

In both trials 1 and 2 there was no significance between any groups, $p>0.05$. In trial 1, group B had a p-value of 0.4510, group C had a p-value of 0.7007, and group D had a p-value of 0.8718 compared to group A. In trial 2, group B had a p-value of 0.8520, group C had a p-value of 0.5923, and group D had a p-value of 0.2527 compared to group A. In trial 3, group B had a p-value of 0.0000998 compared to group A, group C had a p-value of 0.0000852 compared to group A, and group D had a p-value of 0.00000814 compared to group A. Trials 1 and 2 showed insignificance to each other based on Tukey HSD calculations. However, it is evident in trial 3 that group B, C, and D are significant compared to the control group. The most significant being group D. Based on trials 1 and 2, it is concluded that shrimp wastewater had no effect on the plant height of watercress. However, in trial 3, it is concluded that the wastewater with the most amounts of shrimp (15 shrimp) had the greatest effect on the plant height of watercress.

Fresh Weight

In trial 1 there was no significance between any groups, $p>0.05$. In trial 1, group B had a p-value of 0.0647, group C had a p-value of 0.2104, and group D had a p-value of 0.2089 compared to group A. In trial 2, group B had a p-value of 0.0243, group C had a p-value of 0.1118, and group D had a p-value of 0.1356 compared to group A. It was found that group B was significantly different from group A, $p<0.05$. In trial 3, groups B, C, and D were found to be significant compared to the control. Group B had a p-value of 0.0000331815, group C had a p-value of 0.000000087334, and group D had a p-value of 0.0000570013. It is concluded that shrimp wastewater had minimal effect on the fresh mass of watercress.

Plant Survival Rate

All plants from all trials survived. The survival rate was 100%.

Ghost Shrimp Survival Rate

In trial 1, by the end of the experiment, a total of 12 shrimp amongst all groups were alive. The survival rate was 50%. In trial 2, by the end of the experiment 15 shrimp were alive. The survival rate was 73%. In trial 3, a total of 21 shrimp were alive. The survival rate was 90%. Shrimp death could be due to several factors. Such as fluctuating temperatures in the environment as well as the inability to feed on certain days.

Potential Sources of Error/Limitations

Among the two portions of our project, those being ghost shrimps and watercress, there are multiple errors and limitations that occurred. As the ghost shrimps are purchased from Carolina Biological Supply, there was a lack of consistency in life expectancy of the shrimp due to the lack of control over the environment of the vender and the conditions of the transportation. However, to compensate for this potential loss, 3 times the required number of ghost shrimps were purchased and only the healthiest most active shrimps were utilized.

Potential sources of error that do not currently have a solution is the survival rate of the ghost shrimp. Although some shrimp that were delivered were already dead, it was hard to maintain the surviving shrimp alive. Due to the demise of multiple shrimp every day, it was hard to stay true to the different amounts of shrimp needed in each tank according to our experimental groups. Further research is needed to figure out how to keep ghost shrimp alive.

Furthermore, when trials 1 and 2 were conducted, the shrimps were kept in a warm room, causing the shrimps to die easily. This caused us to have an inaccurate number of shrimps in each group due to the environment we conducted trials 1 and 2 in. However, trial 3 was conducted in a room that wasn't as warm, allowing us to have the accurate number of shrimps in each group.

Conclusion

Overall, the data shown in trial 3 illustrated that shrimp wastewater had a significant effect on the cultivation of watercress. This conclusion supported the hypothesis that the quantity of 15 ghost shrimp would be most beneficial to watercress quality and growth. In trial 3 groups B, C, and D were significant compared to the control group. This suggests that ghost shrimp can be utilized in an aquaponics system because their waste water supports plant growth.

Future Research

In forthcoming works, the inclusion of more trials using larger numbers of ghost shrimp and extending the experiment for a longer period are possible methods to further research. Increasing the experiment's time allows the researchers to collect further data and to observe changes in the growing watercress over a longer period. The use of larger shrimp quantities, such as 30 and 40, could display the significance of shrimp wastewater on the cultivation of watercress. Thus, resulting in finding a specific quantity of shrimp that is most effective in cultivating watercress the best.

Acknowledgements

We thank: Dr. D. Marmor, Mrs. N. Jaipershad, Dr. L. Wang, Dr. J. Cohen, Dr. X. Lin, Ms. A. Khemlani, Mr. Z. Liang, Ms. J. Zhu and Ms. DePietro.

Funding: FLHS Science Research Program

References

AlShrouf, A. (2017). Hydroponics, aeroponic and aquaponic as compared with conventional farming. *American Academic Scientific Research Journal for Engineering, Technology, and Sciences*, *27*(1), 247-255.

Barbosa, G.L., Gadelha, F.D.A., Kublik, N., Proctor, A., Reichelm, L., Weissinger, E., Wohlleb, G.M., & Halden, R.U. (2015). Comparison of Land, Water, and Energy Requirements of Lettuce Grown Using Hydroponic vs. Conventional Agricultural Methods. *International Journal of Environmental Research and Public Health,* **12**, 6879-6891. 10.3390/ijerph120606879

Chu, Y. T., & Brown, P. B. (2021). Evaluation of Pacific whiteleg shrimp and three halophytic plants in marine aquaponic systems under three salinities. *Sustainability*, *13*(1), 269.

Clemente, M., Miguel, M., Gribner, C., Moura, P. F., Rigoni, A. A. R., Fernandes, L. C., & Miguel, O. G. (2019). Watercress, as a functional food, with protective effects on human health against oxidative stress: A review study. *International Journal of Medicinal Plants and Natural Products*, *5*(3), 12-16.

de Farias Lima, J., Duarte, S. S., Bastos, A. M., & Carvalho, T. (2019). Performance of an aquaponics system using constructed semi-dry wetland with lettuce (Lactuca sativa L.) on treating wastewater of culture of Amazon River shrimp (Macrobrachium amazonicum). *Environmental Science and Pollution Research*, *26*(13), 13476-13488.

Nelson, R.L., 2008. Aquaponic Food Production. Nelson and Pade Inc. Press, Montello, WI, USA, 218 pp.

Papaspyrou, S., Gregersen, T., Cox, R. P., Thessalou-Legaki, M., & Kristensen, E. (2005). Sediment properties and bacterial community in burrows of the ghost shrimp *Pestarella* *tyrrhena* (Decapoda: Thalassinidea). *Aquatic Microbial Ecology*, *38*(2), 181-190.

Robert. (2022, January 14). *Ghost shrimp care [diet, lifespan, Breeding & More]*. Fishkeeping World

Roosta, H.R., & Hamidpour, M. (2011). Effects of foliar application of some macro- and micro-nutrients on tomato plants in aquaponic and hydroponic systems. *Scientia Horticulturae*, **129**, 396-402. 10.1016/j.scienta.2011.04.006

Tsoukanelis, A. (2007). Benefits for Cancer Protection, Vision, and Heart Health.

Veludo, M., Hughes, A., & Le Blan, B. (2012). Introduction to Aquaponics: A Key to Sustainable Food Production. *Survey of Aquaponics in Europe. Water*.

UFK (2021). Can You Eat Ghost Shrimps? Easy Ghost Shrimp Recipe Or A Fad? *Urban Fish Keeping*.

Zheljazkov, V. D., Horgan, T. E., Astatkie, T., Fratesi, D., & Mischke, C. C. (2011). Study on shrimp waste water and vermicompost as a nutrient source for bell peppers. *HortScience*, *46*(11), 1493-1496.