



Comparison of the effects of monochromatic light with white light on the growth and development of hydroponically grown *Allium fistulosum*

Pandey A

Submitted: March 31, 2024, Revised: version 1, June 9, 2024

Accepted: June 29, 2024

Abstract

The wavelength and intensity of light regulates the growth and development of plants and influences their physiology and morphology both above and below the ground. The emergence of hydroponic propagation of plants under controlled light conditions necessitates an evaluation of different factors for optimal plant growth through this technology. As such, to study the effects of light wavelength on the overall growth of hydroponically grown *Allium fistulosum*, or scallions, the stalk growth, pseudostem diameter, fresh and dry weight of stalks, dry weight of roots, number of secondary roots, and average length of secondary roots were measured under irradiation with four different light wavelengths; 472 nm (blue), 532 nm (green), 700 nm (red) and 400-700 nm (white). Results showed that the growth of stalks and biomass above the ground was significantly maximized for *Allium fistulosum* irradiated with white light while the root growth was significantly maximized when irradiated with red light. The growth of hydroponically cultivated *Allium fistulosum* at various stages in its lifecycle can hence be influenced using different combinations of light wavelength. These findings are of value in hydroponic agriculture to maximize yield.

Keywords

Botany, Plant physiology, Light wavelength, *Allium fistulosum*, Plant morphology, Root development, Shoot development, Scallions, Hydroponics, Biomass

Aarushi Pandey, The Woodlands College Park High School, 3701 College Park Dr, The Woodlands, TX 77384, USA. aarushipandey.007@gmail.com

1 Introduction

1.1 Background

Light is a vital component for oxygenic photosynthesis in plants, algae, and cyanobacteria and is a key abiotic factor that affects plant growth, morphology, and physiology (1). Photoreceptor proteins, such as UVR8, phototropins, cryptochromes, and phytochromes detect and decode light signals to regulate the development of plants, including germination, root development, and biomass growth (2). Exposure to light stimulates seed germination in species such as *Lactuca sativa*, while in others, such as *Allium sativum*, it has the opposite effect (3). Xu et al. found that the 8R1B1P1G wavelength combination maximized root growth in *C. Lanceolata* (4). In another study on the correlation between light and root sensitivity to salinity, Yokawa et al. found that root halotropism of *Arabidopsis* was less effective in light-exposed roots (5). The intensity of light during early stages of plant development also affect leaf morphology. In a study conducted on *Glycine max*, Feng et al. observed a positive correlation between light intensity and the thickness of leaves and their palisade tissue (6).

The rate of photosynthesis is also affected by the color of light; more photosynthetic reactions occur within the red spectrum compared to blue because red wavelengths are absorbed by photosystem II while blue wavelengths are absorbed by carotenoids, which are highly inefficient in transferring the energy to chlorophyll (7). A study on the effects of light color on the morphology of

Lindberg et al. found that red wavelengths amplified leaf biomass and stem length of *Impatiens walleriana*, *Solanum lycopersicum*, and *Salvia officinalis* while plants grown under blue wavelength irradiation produced shorter stems and smaller leaves (8). Ye et al. observed that *Anoectochilus roxburghii* plants irradiated with blue wavelength consistently produced higher concentrations of photosynthetic pigments (9).

As the effect of different wavelengths of light on various plant processes become increasingly clear, it is important to not only consider the immediate physiological responses but also to understand the long-term developmental changes induced by varying light conditions. This broader perspective is vital when considering the optimal lighting strategies for plant growth and development. For instance, Tomari et al. specifically examined the growth responses of green onions to different light irradiation methods (10), comparing traditional top-down lighting with sideward irradiation. They found that sideward irradiation significantly enhanced shoot fresh weight compared to top irradiation, highlighting an area of consideration for agricultural optimization.

The pH is a critical factor influencing plant growth and development, with significant, observable effects on both shoot and root mass. Gillespie et al. found that as pH decreased, plant growth was adversely affected, with spinach at a pH of 4.0 exhibiting stunted overall growth and severely inhibited root development (11). To achieve and stabilize

desired pH levels, various buffering compounds are employed, including tribasic sodium phosphate yielding a neutral to slightly basic aqueous solution and sodium hydrogen sulfate whose aqueous solution pH is slightly acidic. When used at appropriate concentrations, these buffers do not adversely affect plant growth, thereby ensuring that observed developmental changes are not because of buffer concentration.

This study compared the effects of different monochromatic wavelengths of light: 472 nm (blue), 532 nm (green), 700 nm (red) with the broader spectrum of white light: 400-700 nm, on three important aspects of plant morphology—the root system, the shoot system, and overall plant biomass.

2 Materials and Methods

2.1 Hydroponic system setup

2.1.1 *Air pump setup*

The air pump consisted of four 147 cm silicone hoses (divided into four 10 cm, 12 cm, and 125 cm sections), four regulator valves, four air stones, one air pump, and one power outlet. One end of each 10 cm silicone hose section was attached to each of the four outlets on the air pump and the other end of each hose was attached to the thinner ends of four regulator valves. Then, the other, thicker end of the regulator valves were attached to one end of each four 12 cm hose section cut earlier. The other end of each 12 cm hose was attached to one end of each check valve. The other end of each check valve was attached to each of the longer 125 cm silicone hoses. Finally, the other end of each 125 cm silicone hose was attached to four air stones which were used in each of the nutrient reservoirs. This air pump was plugged into a power outlet. The schematic diagram in Figure 1 shows the arrangement of various components after the final assembly of the air pump.

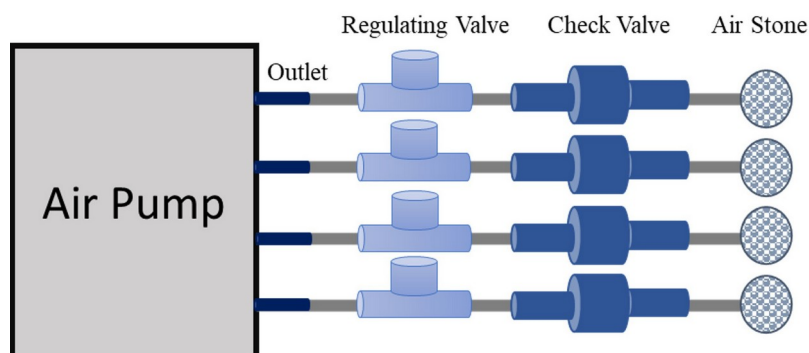


Figure 1. Schematic diagram of air pump

2.1.2 Nutrient reservoir setup

Each of the water reservoirs for the four hydroponic systems were filled with water, with 2 cm of space deliberately left at the top. One air stone was placed in each of the reservoirs and the air pump was powered for a duration of 8 hours to dechlorinate the water in the reservoirs. After the water was dechlorinated, pH readings were taken from each reservoir and 20 g of hydroponic nutrients were added and stirred for 15 minutes until completely dissolved. The pH was once again recorded, and pH buffers were added to adjust the pH of each reservoir, as needed, to a range of 5.3-5.8. The buffering solution utilized to raise the pH in this study was formulated with potassium carbonate and potassium silicate as its primary ingredients. It is an odorless water soluble liquid with a pH value of 12. While the precise concentrations of potassium carbonate and potassium silicate were not specified, they are typically found in the range of 0.1 M to 1 M. The buffering solution utilized to lower the pH in this study contained phosphoric acid, citric acid, and mono ammonium phosphate. This solution is also odorless, featuring a more acidic pH of 1.2. The concentrations of the acids and buffering agent in this solution are generally not precisely defined but are estimated to be substantial. Phosphoric acid concentrations usually span from 5% to 30% by weight, with molarity possibly ranging from 0.1 M to 1 M for citric acid and from 0.05 M to 0.5 M for mono ammonium phosphate. Initial measurements of Total Dissolved Solids (TDS) (ppm) and temperature (°C) were recorded for each reservoir. Finally, the lids of each

reservoir were secured and buoys were inserted in each slot.

2.1.3 Isolated lighting environment setup

Three remote-controlled RGB light bulbs were fixed in a socket splitter and connected to an extension hanging lantern cable; this process was repeated four times for each of the hydroponic systems. Four boxes were assembled to mimic a dark room by closing one end and leaving the other end open. Three openings of the size of each RGB light bulb were cut on the closed end of each box, and the light bulbs were subsequently inserted through the openings and secured using high-strength tape. Next, each of the extension hanging lantern cables were connected to a power outlet and the bulbs were tested for wavelength changing capabilities. In this study, a top irradiation setup was employed using LED lighting systems positioned directly above the green onions. This method was chosen to standardize the light exposure and simplify the experimental design, making it possible to focus on the effects of different light spectra on plant growth. While effective for these purposes, this setup did not explore the potential advantages of varying the direction of light exposure.

2.1.4. Preparation of *Allium fistulosum* for planting

Forty-eight planting sponges were soaked in pH adjusted nutrient solution for 2 minutes and set aside. The roots of all *Allium fistulosum* plants were gently washed using clean water. The saplings were positioned in the soaked planting sponges in such a way that the lower

end of each stem was fixed in the sponge and the roots remained outside to ensure that roots were able to absorb nutrients from the solution. Netting pots were subsequently placed in each of the twelve openings on the four reservoir lids, and the planting sponges with saplings were placed in them. In this study, all plants were uniformly trimmed to 5 cm from the base prior to light irradiation application in order to ensure standardized growth conditions across all experimental groups.

2.1.5. Drying

Extraneous material was removed, followed by washing with distilled water and patting dry with paper towels. Samples were pre-dried in a well ventilated area for 12 hours. They were then placed in an oven set to 82°C with the door slightly ajar and dried for 1 hour with turnings every 15 minutes to ensure even

drying. After cooling in a desiccator, the material was weighed. This process of oven drying was repeated until constant weight.

2.1.6. Final assembly

The four hydroponic reservoirs were placed next to each other near a power outlet and covered with the lighting boxes to ensure they were isolated from each other and external light sources. To complete the final assembly, the extension hanging lantern cables were plugged into a surge protector and the surge protector was plugged into a smart switch, which was programmed to power on at 7 AM and power off at 7 PM to ensure consistency in light exposure duration. The air pump was then powered to run for the entire duration of the experiment. The final system appeared similar to the schematic diagram depicted in Figure 2.

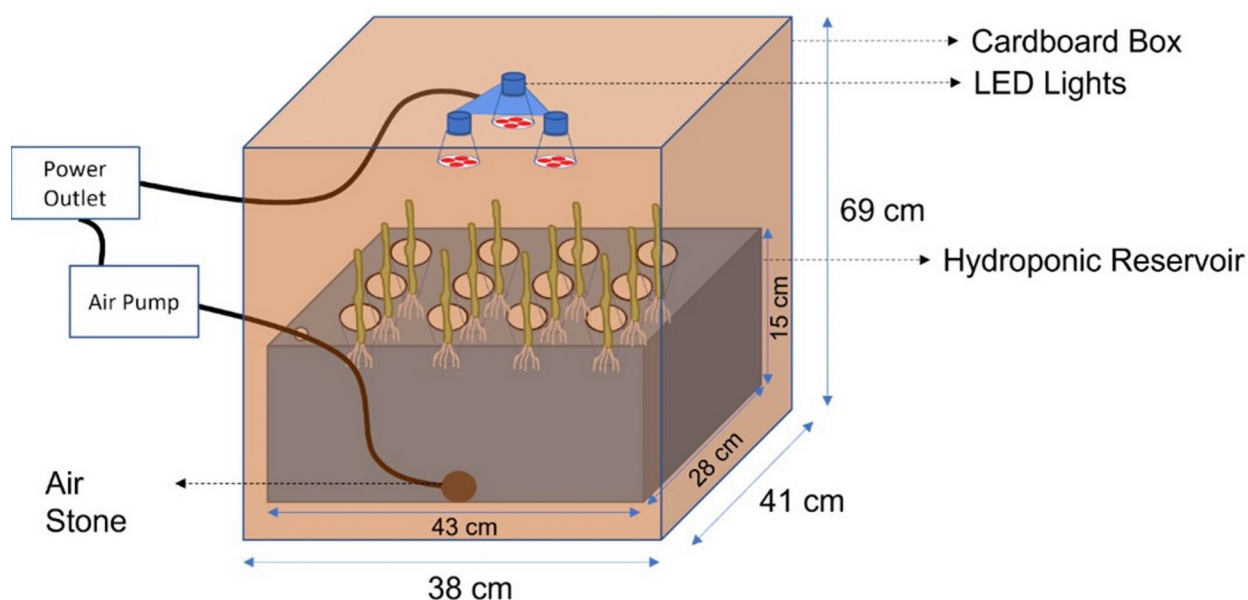


Figure 2. Schematic diagram of hydroponic system

2.2 Experimentation

The pH of the nutrient solutions was maintained to remain in the range of 5.3-5.8, the temperature was maintained to remain in the range of 20-25°C, and the TDS remained in the range of 3000-4500 ppm. Since the *Allium fistulosum* was regrown following trimming rather than from new bulbs, the experiment was run for a relatively short duration of ten days. This duration was chosen because regrowth from trimmed stalks, utilizing the existing root system, is significantly faster compared to growing from seeds. The ten-day period allowed for the effective observation and measurement of rapid early-stage regrowth. *Allium fistulosum* in each of the four reservoirs was exposed to lights from the RGB bulbs (different wavelengths) for a fixed duration of 12 hours each day. The heights and growth of each stalk in each reservoir was measured all for all ten days of each round of experimentation. The TDS and pH were also measured to ensure that these factors remained within their prescribed levels.

2.3 Data collection and statistical significance

On the tenth day of each trial, final measurements of stalk growth (cm), pseudostem diameter (cm), the fresh weight of the main stalk (g), fresh weight of all the stalks (g), dry weight of all the stalks (g), dry weight of the roots (g), number of secondary roots, and lengths of the secondary roots (cm) were recorded. The fresh and dry weights were recorded using a digital weighing scale, $d=0.01$ g. After data for all dependent variables was collected, a one-factor analysis of variance (ANOVA) test was conducted for each

dependent variable. These tests were done to determine whether the differences in the mean values of each dependent variable across the four groups were statistically significant. A p-value of < 0.05 was considered significant.

3 Results

The null hypothesis stated that there was not a significant difference between the mean growth of *Allium fistulosum* grown under the four different wavelengths. The alternative hypothesis stated that the color of light irradiation did influence the mean growth of *Allium fistulosum*. This section focuses on analyzing the observations made during experimentation.

3.1 Raincloud Plots of Measured Variables

Under irradiation with white light, the median total growth (Figure 3), median fresh weight of the main stalks (Figure 4), median fresh weight of all stalks (Figure 5), median dry weight of all stalks (Figure 6), median pseudostem diameter (Figure 8), and median root length (Figure 9) were maximum. However, median secondary root count (Figure 10) was the greatest under red light illumination. Median dry root weight (Figure 7) was maximum under both red and white light illumination. Under red light exposure, median total growth (Figure 3), median fresh weight of the main stalks (Figure 4), median fresh weight of all stalks (Figure 5), median dry weight of all stalks (Figure 6), and median root length (Figure 9) were minimum. Finally, median secondary root count (Figure 10) was lowest under blue and green light exposure, and median dry root weight (Figure 7) and median pseudostem

diameter (Figure 8) were lowest under green light exposure. In summary, for hydroponically grown *Allium fistulosum*, longer and thicker shoots as a higher biomass was observed for plants grown under white light compared to those grown under red and blue light illumination, whereas plants grown under red light were observed to have higher levels of root development compared to those grown under blue and green light illumination.

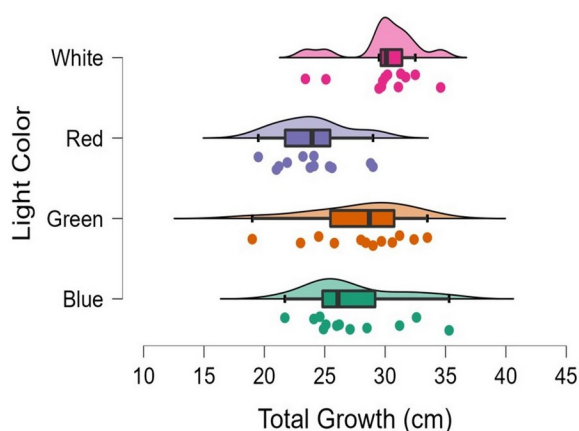


Figure 3. Total growth, Raincloud plot

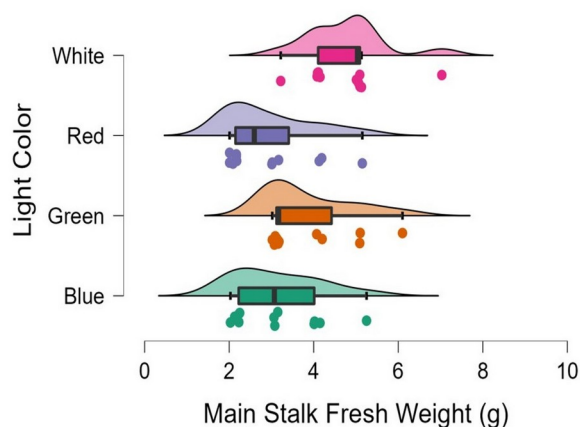


Figure 4. Fresh weight of main stalks, Raincloud plot

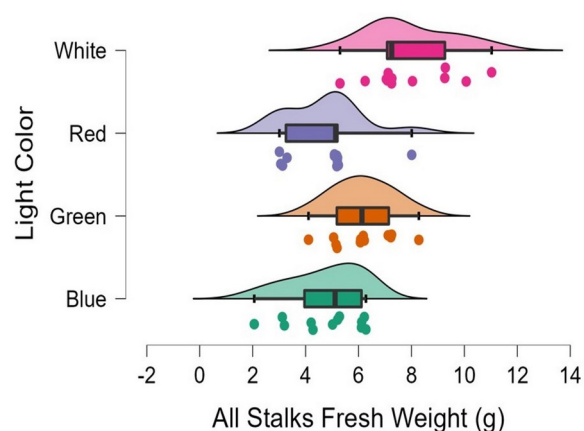


Figure 5. Fresh weight of all stalks, Raincloud plot

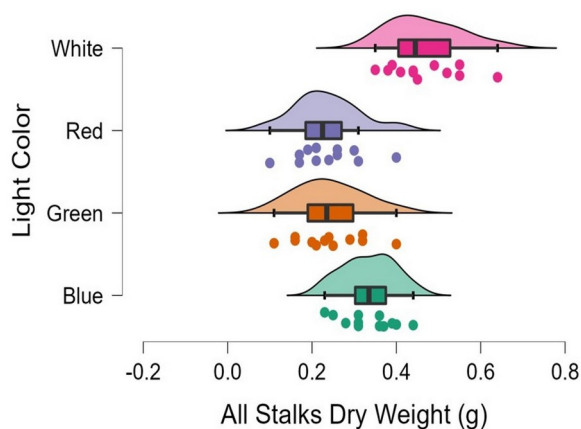


Figure 6. Dry weight of all stalks, Raincloud plot

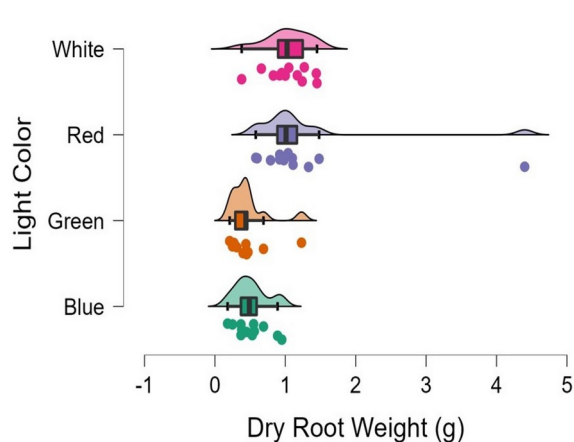


Figure 7. Dry root weight, Raincloud plot

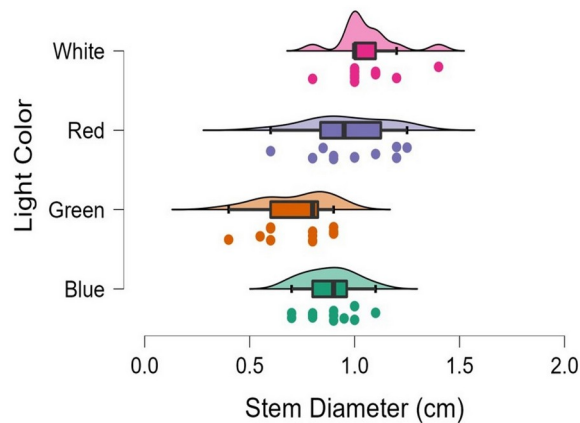


Figure 8. Pseudostem diameter, Raincloud plot

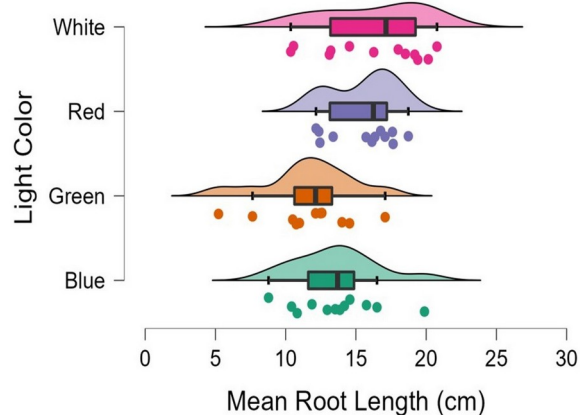


Figure 9. Mean root length, Raincloud plot

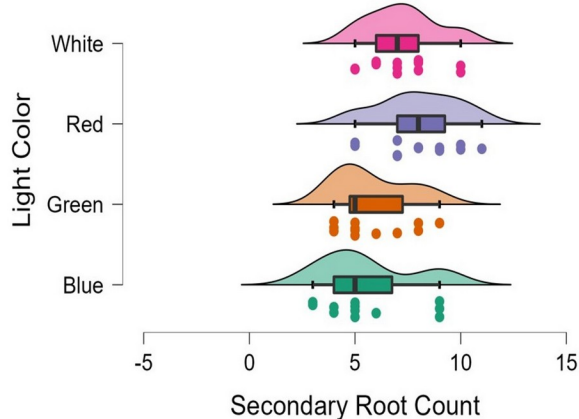


Figure 10. Secondary root count, Raincloud plot

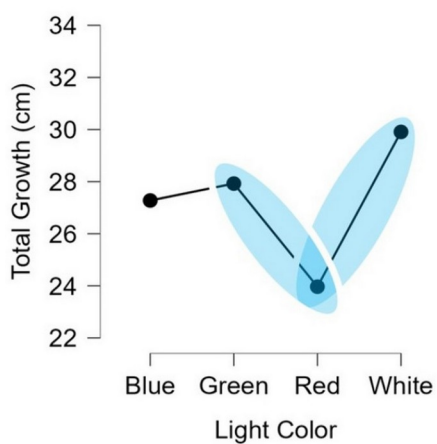


Figure 11. Total growth, descriptive plot

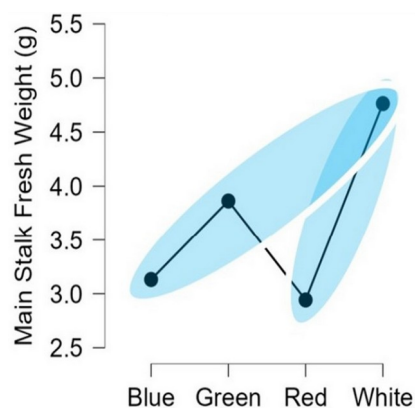


Figure 12. Main stalk fresh weight, descriptive plot

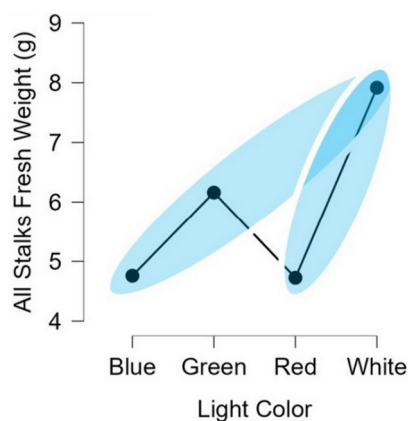


Figure 13. All stalks fresh weight, descriptive plot

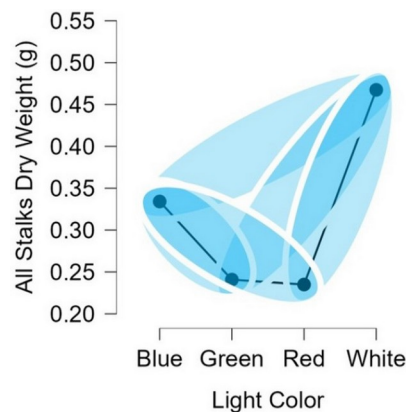


Figure 14. All stalks dry weight, descriptive plot

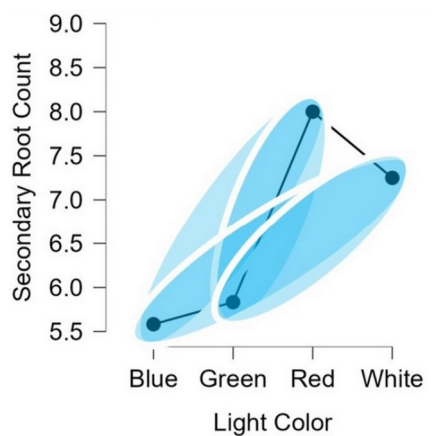


Figure 15. Secondary root count, descriptive plot

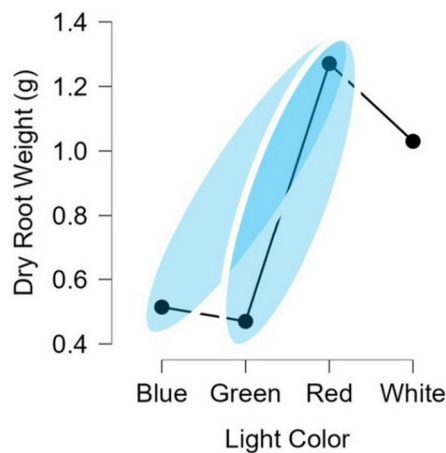


Figure 16. Dry root weight, descriptive plot

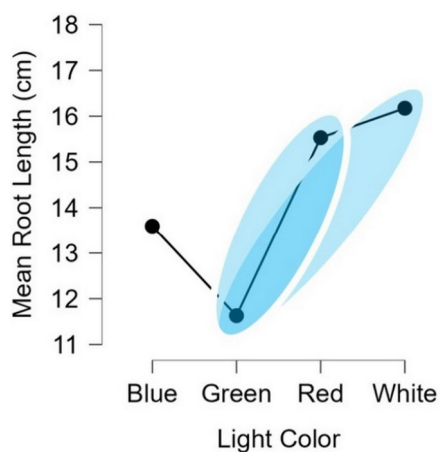


Figure 17. Mean root length, descriptive plot

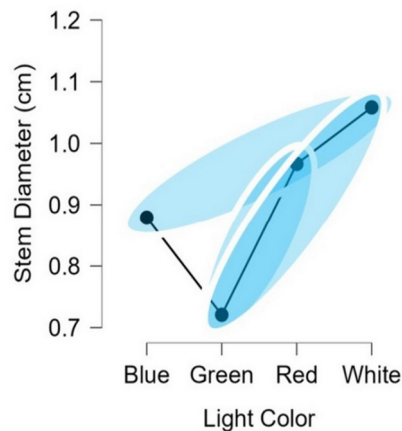


Figure 18. Pseudostem diameter, descriptive plot

3.2 Tests of Statistical Significance

Table 1 shows the results of a one-factor ANOVA for total growth, and Table 2 shows the summary statistics of Tukey post-hoc

comparisons of the mean total growth. Figure 11 visually depicts the mean total growth of *Allium fistulosum* upon exposure to the four different light wavelengths.

Table 1. Total Growth (cm) ANOVA

Cases	Sum of Squares	Degrees of Freedom	Mean Square	F	p
Between Groups	219.622	3	73.207	5.745	0.002
Within Groups	560.681	44	12.743		

Table 2. Total Growth (cm) Post-Hoc Comparisons

Wavelength pairs		Mean Difference	SE	t	p _{Tukey}
Blue	Green	-0.650	1.457	-0.446	0.970
	Red	3.308	1.457	2.270	0.121
	White	-2.633	1.457	-1.807	0.284
Green	Red	3.958	1.457	2.716	0.045*
	White	-1.983	1.457	-1.361	0.530
Red	White	-5.942	1.457	-4.077	0.001**

* $p < 0.05$, ** $p < 0.01$

Table 3 shows the results of a one-factor ANOVA for main stalk fresh weight, and Table 4 shows the summary statistics of Tukey post-hoc comparisons of the mean main stalk fresh

weight. Figure 12 visually depicts the mean fresh weight of the main stalks of each *Allium fistulosum* upon exposure to the four different light wavelengths.

Table 3. Main Stalk Fresh Weight (g) ANOVA

Cases	Sum of Squares	Degrees of Freedom	Mean Square	F	p
Between Groups	24.642	3	8.214	7.874	< 0.001
Within Groups	45.900	44	1.043		

Table 4. Main Stalk Fresh Weight (g) Post-Hoc Comparisons

Wavelength pairs		Mean Difference	SE	t	p _{Tukey}
Blue	Green	-0.732	0.417	-1.755	0.309
	Red	0.189	0.417	0.454	0.969
	White	-1.633	0.417	-3.915	0.002**
Green	Red	0.921	0.417	2.208	0.137
	White	-0.901	0.417	-2.160	0.150
Red	White	-1.822	0.417	-4.369	< 0.001***

** p < 0.01, *** p < 0.001

Table 5 shows the results of a one-factor ANOVA for fresh weight of all stalks, and fresh weight of all the stalks of each *Allium fistulosum* upon exposure to the four different post-hoc comparisons of mean fresh weight of light wavelengths.

Table 5. All Stalk Fresh Weight (g) ANOVA

Cases	Sum of Squares	Degrees of Freedom	Mean Square	F	p
Between Groups	81.801	3	27.267	13.363	< 0.001
Within Groups	89.784	44	2.041		

Table 6. All Stalk Fresh Weight (g) Post-Hoc Comparisons

Wavelength pairs		Mean Difference	SE	t	p _{Tukey}
Blue	Green	-1.402	0.583	-2.404	0.091
	Red	0.034	0.583	0.059	1.000
	White	-3.158	0.583	-5.414	< 0.001***
Green	Red	1.436	0.583	2.462	0.080
	White	-1.756	0.583	-3.011	0.022*
Red	White	-3.192	0.583	-5.473	< 0.001***

* p < .05, *** p < .001

Table 7 shows the results of a one-factor ANOVA for dry weight of all stalks, and Table 8 shows the summary statistics of Tukey post-hoc of the mean dry weight of all stalks. Figure 14 visually depicts the mean dry weight of all the stalks of each *Allium fistulosum* upon exposure to the four different light wavelengths.

Table 7. All Stalk Dry Weight (g) ANOVA

Cases	Sum of Squares	Degrees of Freedom	Mean Square	F	p
Between Groups	0.425	3	0.142	23.569	< 0.001
Within Groups	0.265	44	0.006		

Table 8. All Stalk Dry Weight (g) Post-Hoc Comparisons

Wavelength pairs		Mean Difference	SE	t	p _{Tukey}
Blue	Green	0.093	0.032	2.948	0.025*
	Red	0.099	0.032	3.132	0.016*
	White	-0.133	0.032	-4.211	< 0.001***
Green	Red	0.006	0.032	0.184	0.998
	White	-0.227	0.032	-7.158	< 0.001***
Red	White	-0.233	0.032	-7.342	< 0.001***

* $p < .05$, *** $p < .001$

Table 9 shows the results of a one-factor ANOVA for secondary root count, and Table 10 shows the summary statistics of Tukey post-hoc comparisons of secondary root count. Figure 15 visually depicts the mean secondary, or lateral, root count of each *Allium fistulosum* upon exposure to the four different light wavelengths.

Table 9. Secondary Root Count ANOVA

Cases	Sum of Squares	Degrees of Freedom	Mean Square	F	p
Between Groups	47.833	3	15.944	4.417	0.008
Within Groups	158.833	44	3.610		

Table 10. Secondary Root Count Post-Hoc Comparisons

Wavelength pairs		Mean Difference	SE	t	p _{Tukey}
Blue	Green	-0.250	0.776	-0.322	0.988
	Red	-2.417	0.776	-3.116	0.016*
	White	-1.667	0.776	-2.149	0.154
Green	Red	-2.167	0.776	-2.793	0.037*
	White	-1.417	0.776	-1.826	0.275
Red	White	0.750	0.776	0.967	0.769

* $p < .05$

Table 11 shows the results of a one-factor ANOVA for dry root weight, and Table 12 shows the summary statistics of Tukey post-hoc comparisons of the mean dry root weight. Figure 16 visually depicts the mean dry weight of the roots of each *Allium fistulosum* upon exposure to the four different light wavelengths.

Table 11. Dry Root Weight (g) ANOVA

Cases	Sum of Squares	Degrees of Freedom	Mean Square	F	p
Between Groups	5.435	3	1.812	5.585	0.003
Within Groups	13.947	43	0.324		

Table 12. Dry Root Weight (g) Post-Hoc Comparisons

Wavelength pairs		Mean Difference	SE	t	p _{Tukey}
Blue	Green	0.044	0.238	0.186	0.998
	Red	-0.757	0.233	-3.324	0.011*
	White	-0.516	0.233	-2.219	0.134
Green	Red	-0.801	0.238	-3.369	0.008**
	White	-0.560	0.238	-2.356	0.101
Red	White	0.241	0.233	1.036	0.730

* $p < .05$

Table 13 shows the results of a one-factor ANOVA for mean root length, and Table 14 shows the summary statistics of Tukey post-hoc comparisons of the mean root length. Figure 17 visually depicts the mean length of the roots of each *Allium fistulosum* upon exposure to the four different light wavelengths.

Table 13. Mean Root Length (cm) ANOVA

Cases	Sum of Squares	Degrees of Freedom	Mean Square	F	p
Between Groups	144.761	3	48.254	5.005	0.005
Within Groups	414.589	43	9.642		

Table 14. Mean Root Length (cm) Post-Hoc Comparisons

Wavelength pairs		Mean Difference	SE	t	p _{Tukey}
Blue	Green	1.964	1.296	1.515	0.437
	Red	-1.937	1.268	-1.528	0.430
	White	-2.581	1.268	-2.036	0.191
Green	Red	-3.901	1.296	-3.010	0.022*
	White	-4.545	1.296	-3.507	0.006**
Red	White	-0.644	1.268	-0.508	0.957

* $p < 0.05$, ** $p < 0.01$

Table 15 shows the results of a one-factor ANOVA for mean pseudostem diameter, and the stalks of each *Allium fistulosum* upon exposure to the four different light wavelengths. Table 16 shows the summary statistics of Tukey post-hoc comparisons of the mean pseudostem diameter. Figure 18 visually

Table 15. Pseudostem Diameter (cm) ANOVA

Cases	Sum of Squares	Degrees of Freedom	Mean Square	F	p
Between Groups	0.743	3	0.248	9.766	< 0.001
Within Groups	1.115	44	0.025		

Table 16. Pseudostem Diameter (cm) Post-Hoc Comparisons

Wavelength pairs		Mean Difference	SE	t	p _{Tukey}
Blue	Green	0.158	0.07	2.436	0.085
	Red	-0.088	0.07	-1.346	0.539
	White	-0.179	0.07	-2.756	0.041*
Green	Red	-0.246	0.07	-3.782	0.003**
	White	-0.338	0.07	-5.192	< .001***
Red	White	-0.092	0.07	-1.418	0.500

* $p < 0.05$, ** $p < 0.01$, *** $p < .001$

4 Discussion

The purpose of this study was to test if there was a significant difference in the shoot growth, root complexity, and biomass of *Allium fistulosum* grown indoors hydroponically irradiated with blue, green, red, and white wavelengths of light. Measurements of the daily growth (cm) of the main stalks of each plant, pH of the nutrient solution, TDS (ppm), and temperature (°C) for each reservoir

were recorded every day for a period of ten days. The final measurements were taken on the tenth day for the total growth of the main stalks, pseudostem diameters of the stalks (cm), total fresh weight of the main stalks, total fresh weight of all the stalks, total dry weight of all the stalks, total number of secondary roots, total dry weight of all the roots, and total lengths of all the secondary roots.

One-factor analysis of variance (ANOVA) tests were conducted for each of these dependent variables to determine if there were statistically significant differences in their mean values across four groups of plants grown under different wavelengths of light. The results of ANOVA tests for each dependent variable produced a p-value of less than 0.05, which was the set significance interval. The null hypothesis stating that the difference, if any, between the mean growths, root complexity, and biomass of *Allium fistulosum* exposed to red, blue, green, and white light was not significant was therefore rejected.

The ANOVA tests conducted for each of these dependent variables are described below. It was observed that there was a significant difference in the mean total growth (cm) of the *Allium fistulosum* grown under different light colors (Table 1). This suggested that the color of light had a significant effect on the shoot development of hydroponically grown plants. Furthermore, a Tukey post-hoc test was used to compare the pairwise mean differences and it was found that the mean growth under red light was significantly lower than that under both green and white light (Table 2). From these

results, it was concluded that the shoots of hydroponically grown *Allium fistulosum* grew larger when exposed to white and green light compared to those exposed to red light (Figure 11).

One-factor ANOVA tests were also conducted for each of the variables measuring the plant biomass (fresh weight of main stalk, fresh weight of all stalks, dry weight of all stalks). A significant difference was observed in the mean values of the fresh weight of the main stalks (Table 3), fresh weight of all the stalks (Table 5), and dry weight of all the stalks (Table 7). This suggested that the color of light had a significant effect on the biomass of hydroponically grown *Allium fistulosum*. Tukey post-hoc tests found that compared to red and blue light, *Allium fistulosum* grown under white light had significantly greater main stalk fresh weights (Table 4), total stalk fresh weights (Table 6), and total stalk dry weights (Table 8). The mean fresh weights of all the stalks were also found to be significantly higher when grown under white light compared to when grown under green light (Table 6). Furthermore, the Tukey post-hoc tests also found that the mean difference in the dry weights of all the stalks was greater when grown under blue light compared to both red and green light (Table 8). From these results, it was concluded that white light produced a higher biomass in hydroponically grown *Allium fistulosum* compared to that grown under red and blue light (Figure 12, 13) and plants grown under red and green light had significantly lower mean total stalk dry weights

compared to plants grown under blue light (Figure 14).

One-factor ANOVA tests also indicated significant differences in the mean values of the dry root weights (Table 11), secondary root counts (Table 9), and root length (Table 13) across the four light treatments. This suggested that the color of light had a significant effect on the root development of hydroponically grown *Allium fistulosum*. However, unlike the shoot growth and biomass, the Tukey post-hoc test found that compared to blue and green light, *Allium fistulosum* grown under red light presented with significantly greater dry root weights (Table 12) and secondary root counts (Table 10). The plants grown under red light also had a significantly greater mean root length compared to the plants grown under green light (Table 14). Additionally, the plants grown under white light had a significantly greater mean root length compared to the plants grown under green light (Table 14). From these results, it was concluded that red light produced lengthier roots in hydroponically grown *Allium fistulosum* compared to those grown under blue and green light (Figure 15, 16, 17).

The roots of the plants grown under red light appeared to be the longest and healthiest while root rotting was observed for the plants grown under green light after the fifth day of experimentation. The health of roots can be determined by their appearance. Healthy roots are generally white, while rotting roots are generally darker in color.

Based on these results and observations, the hypothesis, which stated that *Allium fistulosum* hydroponically grown under the white light treatment would have the greatest overall growth, was partially supported. This was because *Allium fistulosum* hydroponically grown under white light presented with significantly greater mean shoot growth and biomass, compared to *Allium fistulosum* grown under red and blue light. However, there were no significant differences observed in the mean shoot growths for plants treated with white and green light. The results also showed that *Allium fistulosum* had a significantly higher root growth when grown under red light compared to when grown under blue and green light. It was concluded that; in terms of growth, biomass or length; the root system of *Allium fistulosum* responded better to red light exposure while the shoot system responded better to white light exposure.

4.1 Limitations

4.1.1 Environmental fluctuations

While recording the daily growth measurements, the plants in all the systems were exposed to outside light for approximately fifteen minutes every day, which could have affected the growth of the plants. The ambient temperature inside the boxes was not the same and this could have influenced the growth of the plants as well. The pH and TDS fluctuated within the various nutrient solutions daily, and it could have also altered the growth of the plants. While taking the measurements of the root lengths and root dry weights, plant number 9 in the hydroponic

system exposed to green light was damaged. Consequently, these readings for plant 9 under green light were missing from the data set that was analyzed.

4.1.2 *Trimming of Allium fistulosum*

In this study, the standard trimming of all green onion plants prior to treatment was intended to minimize initial variability and mimic conditions typical in agricultural management. However, the universal application of trimming does present a limitation in the experimental design: without untrimmed controls, it is challenging to disentangle the effects of the treatments from those induced by the trimming itself. While trimming generally promotes regrowth in green onions, the specific interactions between this regrowth and our experimental treatments remain unclear. The responses observed post-treatment could be partly influenced by the plants' recovery and adaptation to trimming; which in turn may be wavelength dependent. Given the regenerative nature of green onions, the impact of trimming can also extend to root system development and overall plant vigor, which in turn affects the plant's response to environmental stressors and treatment applications.

4.1.3 *Observational limitations of comparing monochromatic light with white light*

The study's experimental design, which utilized uniform light intensities for different spectral treatments, may not have accurately reflected the nuanced effects that varying wavelengths have on plant physiological processes. The impact of white, red, blue, and green light on plant growth and development

demonstrated distinct and varied responses. The effects of red, blue, and green monochromatic lights on plants are not simply cumulative when compared to the effects of white light, which includes a mixture of these wavelengths (12). Moreover, past experiments conducted by Gao et al. found that, when compared with the white light spectrum, monochromatic light significantly inhibited the growth and development of *Allium fistulosum* (13).

Thus, it is crucial to recognize the limitations in comparing white light with monochromatic light wavelengths. White light spans a broad spectrum of wavelengths, whereas monochromatic light is restricted to a narrow band. For example, the physiological impact of blue light at 430 nm with a specific luminosity cannot be directly equated to the broader range of wavelengths present in white light, even if the overall luminosity is adjusted proportionally.

To address this limitation, future studies should consider comparing white light supplemented with varying ratios of individual colored lights, as this approach allows for a more precise evaluation of the specific contributions of each wavelength to plant growth and development. This method can help isolate the effects of each wavelength within the context of the broader light spectrum present in natural white light, providing a clearer understanding of the interactions between different light qualities and their combined effects on plant physiology.

This confounding factor is increased when the effect of overhead illumination is applied to root structure that grows underwater. The absorption coefficient of red light (0.004 m^{-1}) is two-orders -magnitude lesser than that of blue light (0.4 m^{-1}). This implies that the light exposure intensity of the root system is significantly different when exposed to overhead white, red or blue light; thereby making any comparison almost meaningless. Nevertheless, this factor was not taken into account for the purposes of this study due to the mathematical complexity involved and the resultant digression from the main objectives of the study. Future experiments will need to keep the intensity of light that underwater root systems are exposed to, constant. This can perhaps be accomplished by installing underwater LEDs’.

4.1.4 *Limited observations on nutritional sulfur-containing compounds and antioxidants*

The experimental design, employing subsets of white light, may not have captured the full spectrum of effects that specific light wavelengths have on sulfur-containing compounds and antioxidant content in plants. While the approach in this study provided valuable preliminary observations, it lacked the detailed mechanistic insights that could be achieved by employing a more targeted light supplementation strategy. Thus, these findings, while indicative of some trends, should be viewed as preliminary. They underscore the need for further research that employs a more nuanced lighting strategy to identify the specific effects of different light qualities on plant metabolic pathways.

4.1.5 *Mineral Content*

Previous studies conducted by Jasoni et al. used nutrient solutions varying in pH, composition, and concentration to determine the effect of these variations on the biomass and mineral content of hydroponically grown *Allium fistulosum*, finding that changes in one did not necessarily correlate with changes in the other (14). Thus, while this study found that white light produced the greatest biomass, it is not necessary that white light also produced the highest mineral content, a factor that is valued by agricultural producers and consumers. This underscores the need for further experimentation to determine the combination of light wavelengths that maximizes both biomass production and mineral content.

4.1.6 *Focus on top irradiation*

Finally, while this study provided valuable insights into the effects of spectral variations while directly placed below a radiation source, it is limited by not considering the effect of different irradiation directions. Sideward irradiation, as shown to be effective in other studies, could potentially lead to different growth responses, which were not explored in this study’s setup (10). This limitation highlights the need for a broader examination of light direction in future studies to fully understand its implications on plant morphology and productivity.

4.2 *Applications and perspective*

This study’s results may help construct optimal combinations of light to improve the quantity and quality of *Allium fistulosum* produced in large scale commercial hydroponic farms.

However, it should be noted that the greatest biomass may not necessarily correlate with the greatest mineral, alkaloid, antioxidant or nutritional content, which may be prized by consumers.

In the future, more studies should be performed to understand the effects of light color and intensity on the qualitative aspects (taste, aroma, nutritional value, etc.) of hydroponically grown *Allium fistulosum*. Additionally, the effects of light color and intensity on the nutrient absorption by *Allium fistulosum*, and the correlation between light colors, TDS, pH, and different types of nutrients should also be studied to understand the ideal growing conditions for commercial hydroponic cultivation of this crop.

Furthermore, to better isolate the effects of the treatments, future studies should include both trimmed and untrimmed plants as comparative groups. This approach would a more accurate assessment as to whether the observed benefits are due solely to the treatments or are also influenced by enhanced growth from trimming.

Future research should also employ variable light intensities tailored to each wavelength, thereby enabling a more precise evaluation of how light quality influences plant biological responses, including their impact on plant sulfur containing compounds and antioxidant content. Subsequent research should include detailed quantitative analyses of mineral and

antioxidant levels under varied lighting conditions. Such studies would provide valuable insights into the mechanistic effects of light on plant physiology and help validate the empirical models suggested by observational studies.

Finally, to build on the findings of this study, future research should explore the effects of different irradiation directions, such as side or combined top and side irradiation, on green onions. Investigating these variables could provide deeper insights into the optimal environmental conditions for enhancing growth and yield. Such studies could employ a comparative analysis of top versus sideward irradiation, potentially leading to more refined and effective agricultural practices.

5 Conclusion

The growth of hydroponically grown *Allium fistulosum* exposed to different overhead light wavelengths was significantly influenced by the wavelength of the light. Longer and thicker shoots and a greater biomass was observed for plants grown under white light irradiation compared to those grown under red and blue light illumination, whereas plants grown under overhead red light were observed to have higher levels of root development compared to those grown under blue and green light illumination, although in this case, the penetration of light of different wavelengths through the water medium was not taken into account.

6 References

1. Liu, Y., Xiuxia, R., Jeong, B.R. (2019). Supplementary Light Source Affects Growth, Metabolism, and Physiology of *Adenophora triphylla* (Thunb.) A.DC. Seedlings. *BioMed Research International*, 6283989. <https://doi.org/10.1155%2F2019%2F6283989>
2. Paik, I., Huq, E. (2019). Plant photoreceptors: Multi-functional sensory proteins and their signaling networks. *Seminars in Cell & Developmental Biology*, 92, 114-121. <https://doi.org/10.1016/j.semcdb.2019.03.007>
3. Lambers, H. (n.d.). Afterripening, stratification, and temperature effects. *Britannica*. <https://www.britannica.com/science/seed-plant-reproductive-part/Afterripening-stratification-and-temperature-effects>.
4. Xu, Y., Liang, Y., Yang, M. (2019). Effects of Composite LED Light on Root Growth and Antioxidant Capacity of *Cunninghamia lanceolata* Tissue Culture Seedlings. *Scientific Reports*, 9, 9766. <https://doi.org/10.1038/s41598-019-46139-2>
5. Yokawa, K., Fasano, R., Kagenishi, T., Baluska, F. (2014). Light as stress factor to plant roots case of root halotropism. *Frontiers in Plant Science*, 5, 718. <https://doi.org/10.3389/fpls.2014.00718>
6. Feng, L., Raza, M.A., Li, Z., Chen, Y., Khalid, M.H.B., Du, J., et al. (2019). The Influence of Light Intensity and Leaf Movement on Photosynthesis Characteristics and Carbon Balance of Soybean. *Frontiers in Plant Science*, 9, 1952. <https://doi.org/10.3389/fpls.2018.01952>
7. Samuoliene, G., Virsile, A., Brazaityte, A., Jankauskiene, J., Sakalauskiene, S., Vastakaite, V., et al. (2017). Blue light dosage affects carotenoids and tocopherols in microgreens. *Food Chemistry*, 228, 50-56. <https://doi.org/10.1016/j.foodchem.2017.01.144>
8. Lindberg, H., Runkle, E. (2014). Growing Seedlings Under LEDs: Part Two. *Greenhouse Grower*. <https://www.greenhousegrower.com/production/plant-culture/growing-seedlings-under-leds-part-two/>.
9. Ye, S., Shao, Q., Xu, M., Li, S., Wu, M., Tan, X., Su, L. (2017). Effects of Light Quality on Morphology, Enzyme Activities, and Bioactive Compound Contents in *Anoectochilus roxburghii*. *Frontiers in Plant Science*, 8. <https://doi.org/10.3389/fpls.2017.00857>

10. Tomari, Y., Ishikawa, A., Maharjan, G., Watanabe, H. (2024). Examining Spectral Photon Flux Density and Lighting Position and Direction for Green Onion Production in a Plant Factory Using LEDs. *Eco-Engineering*, 36(1), 17-24. <https://doi.org/10.11450/seitaikogaku.36.17>
11. Gillespie, D.P., Papio, G., Kubota, C. (2021). High Nutrient Concentrations of Hydroponic Solution Can Improve Growth and Nutrient Uptake of Spinach (*Spinacia oleracea* L.) Grown in Acidic Nutrient Solution. *HortScience*, 56(6), 687-694. <https://doi.org/10.21273/HORTSCI15777-21>
12. Gao, S., Liu, X., Liu, Y., Cao, B., Chen, Z., Xu, K. (2021). Comparison of the effects of LED light quality combination on growth and nutrient accumulation in green onion (*Allium fistulosum* L.). *Protoplasma*, 258(4), 753-763. <https://doi.org/10.1007/s00709-020-01593-y>
13. Gao, S., Liu, X., Liu, Y., Cao, B., Chen, Z., Xu, K. (2020). Photosynthetic characteristics and chloroplast ultrastructure of welsh onion (*Allium fistulosum* L.) grown under different LED wavelengths. *BMC Plant Biology*, 20, 78. <https://doi.org/10.1186/s12870-020-2282-0>
14. Kane, C., Jasoni, R. L., Peffley, E. P., Thompson, L.D., Green, C.J., Pare, P., Tissue, D. (2006). Nutrient Solution and Solution pH Influences on Onion Growth and Mineral Content. *Journal of Plant Nutrition*, 29(2), 375-390. <http://dx.doi.org/10.1080/01904160500477028>