Mung Bean Germination Under Select Wavelengths

Rashmi Hundal, Michael Liang, and Kents Mutuc

Abstract

Mung beans (*Vigna radiata*) are an inexpensive, sustainable, and essential source of nutrients. Developing optimal methods in germination, such as determining light wavelengths that produce the healthiest seedlings, likely will correlate to protein content and metabolites, and overall a more nutritious bean. This is necessary as many impoverished areas across the world lack accessibility to nutrient-dense ingredients such as animal meat. This experiment had three mung bean groups limited to either red (λ= 680 nm), blue (λ= 410 nm), green (λ= 520 nm), and white light wavelengths. One treatment consisted of a no light control group. The mung beans were germinated under these various light conditions to determine if there would be a significant difference in their final radicle length after one week of light exposure. Statistical analysis showed that there was no significance between the red, blue, green, or white light treatment groups in terms of radicle length. The no light control group displayed significantly greater amounts of radicle growth ($p<0.0001$).

Keywords: mung beans; germination; wavelengths; radicle growth

Introduction

Mung beans (*Vigna radiata*) are a nutrient-rich legume that is high in economic value, especially in third-world countries. Regarding cultivation, mung beans are an essential crop harvested in Africa, South America, and Asia (Birhanu et al., 2018). Not only is *V. radiata* important as a food staple in developing countries, it is also a significant financial resource. Mung bean harvest and exportation is a means of income for many poverty-stricken areas around the globe, as many of these countries have the necessary growing conditions such as warm climates and short periods of rain (Birhanu et al., 2018). Additionally, they also have several nutritional benefits which make them fundamental to various dishes. For example, *V. radiata* is found to be an ideal source of complex carbohydrates, protein, vitamin B, and numerous minerals (Muinos, 2021).

Germination is found to cause variations in the nutrient composition of mung beans. According to Mubarak (2005), during the process of germination, enzymatic activity in legumes will remove non-metabolic components, which in turn, will increase the nutritional value of the mung bean. In developing countries, animal meat is costly which makes it inaccessible for most people; therefore, many communities will rely on legumes such as mung beans as their main source of nutrition (El-Adawy et al., 2003). Since mung beans are important for people's diets worldwide, it would be beneficial to determine optimal conditions for the germination of *V. radiata*, such as the necessary light conditions, which is a parameter of interest.

Bu et al. (2016) demonstrated how light exposure has a positive effect on seed germination in herbaceous species, however, they did not indicate which specific wavelengths of light were the main contributors. It is hypothesized that the radicle growth of *V. radiata* is dependent on multiple wavelengths due to their different roles in plant development. If this hypothesis holds true, the presence of various wavelengths should allow the white light group to outperform the other treatment groups since it contains all wavelengths of light. To execute this, a study was done between 5 different treatments to determine if there are significant differences in final radicle length.

Methods

To observe a difference in germination, 100 mung beans were limited to either red light (λ= 680 nm), blue light (λ= 410 nm), green light (λ= 520 nm), no light, or white light for one week. Our experimental control was the no light group. To prepare the treatment boxes, a 14 cm x 10 cm hole was cut out in the center of four boxes and covered with acetate filter paper or white paper towels (Figure 1). Each filter paper only allows for specific wavelengths of light to pass through. Multiple sheets of white paper towels were used for the white light treatment box to decrease the light intensity so that it matched that of the other groups. The fifth box was left uncut to block out almost all wavelengths of light to represent darkness.

Figure 1. The set up of the five treatment groups from left to right: no light, red light, blue light, green light, and white light. All treatment groups were kept at room temperature. The box without a lamp represents the dark control group. All lamps are situated in the top right corner of each box to ensure a similar angle of light exposure to all seeds. A light intensity of 35 ± 10 Lux was held constant for all groups.

Treatment groups consisted of 4 replicates, each containing 5 seeds. To prepare each replicate, a paper towel was dampened with 2 mL of tap water and placed into resealable baggies, then *V. radiata* seeds were oriented as shown in Figure 2. Each seed was soaked in tap water for 12 hours before being placed into the resealable baggies, allowing for growth to begin straightaway or preempt growth in some cases. It was important that each bag was sealed to maintain the dampness of the paper towel and prevent evaporation.

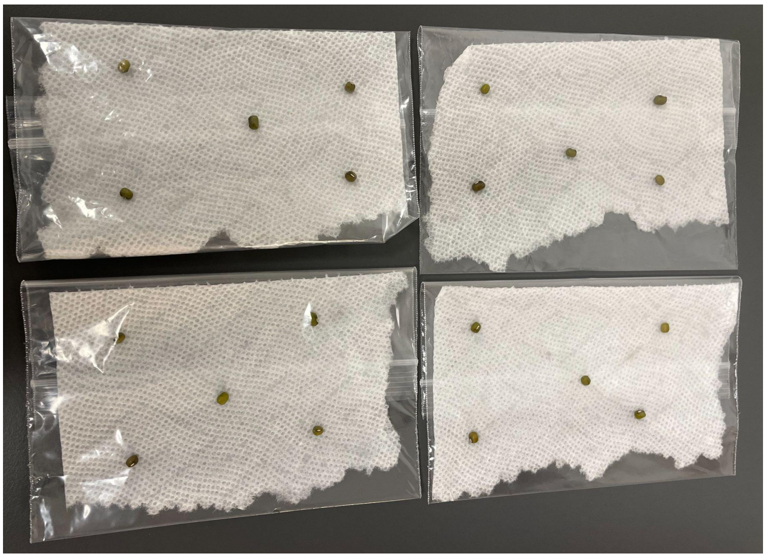

Figure 2. The placement of seeds in each replicate. This arrangement was consistent throughout all treatment groups. Every replicate was kept at room temperature. Each light treatment group had 4 replicates, as arranged above.

Each box represents a treatment group that contains 4 bags, resulting in 20 *V. radiata* seeds per box. To keep the light intensity consistent between all treatment groups, the Light Meter mobile app was used. The height and angle of the lamps were adjusted until each box interior gave a reading of 35 ± 10 lux, excluding the no-light treatment group. Using an automated timer, the boxes were exposed for 8 consecutive hours of light each day. Measurements were taken on the fourth and seventh day to allow for noticeable growth.

Two methods were used to obtain the length of each root. For the first day of the experiment, a string was placed along the root and measured using a ruler; always measuring from the base of the radicle to the tip. The two remaining days involved measuring with ImageJ as radicle growth became too complex to use string. Pictures of each treatment group were imported to the software and the radicle was measured by scaling the image and applying a segmented line tool. After measuring, the seeds and paper towels were re-sprayed with tap water and placed back inside each of the boxes. In terms of our statistical analysis, the mean root length for each treatment group was calculated using Google Sheets. Using an alpha of 0.05, significance was tested between the 5 treatment groups with a one-way Analysis of Variance (ANOVA) that was performed using GraphPad Prism 9. These results were then analysed further using a Tukey-Kramer statistical test.

Results

After the growth period, the data was examined with a one-way ANOVA and a Tukey-Kramer test. One seed in the white light treatment showed no signs of germination after seven days of illumination, and was discarded as an outlier. All four treatments were significantly shorter in length when compared to the no-light group (71.97 mm, $p<0.0001$, SE = 7.41), as shown in Figure 3. Comparisons between the red (30.16 mm, SE = 2.39), blue (23.58 mm, SE = 1.90), green (21.14 mm, SE = 1.95), and white (23.34 mm, SE = 1.26) groups were insignificant (p>0.05). These four treatments had thinner primary roots than the no-light group, but also considerable branching near their seed bases. The average radicle length for green was about 72% less than the average for the no-light group. Although the average radicle length of the red group was 25% longer than that of the blue, white, and green groups, it was not statistically significant.

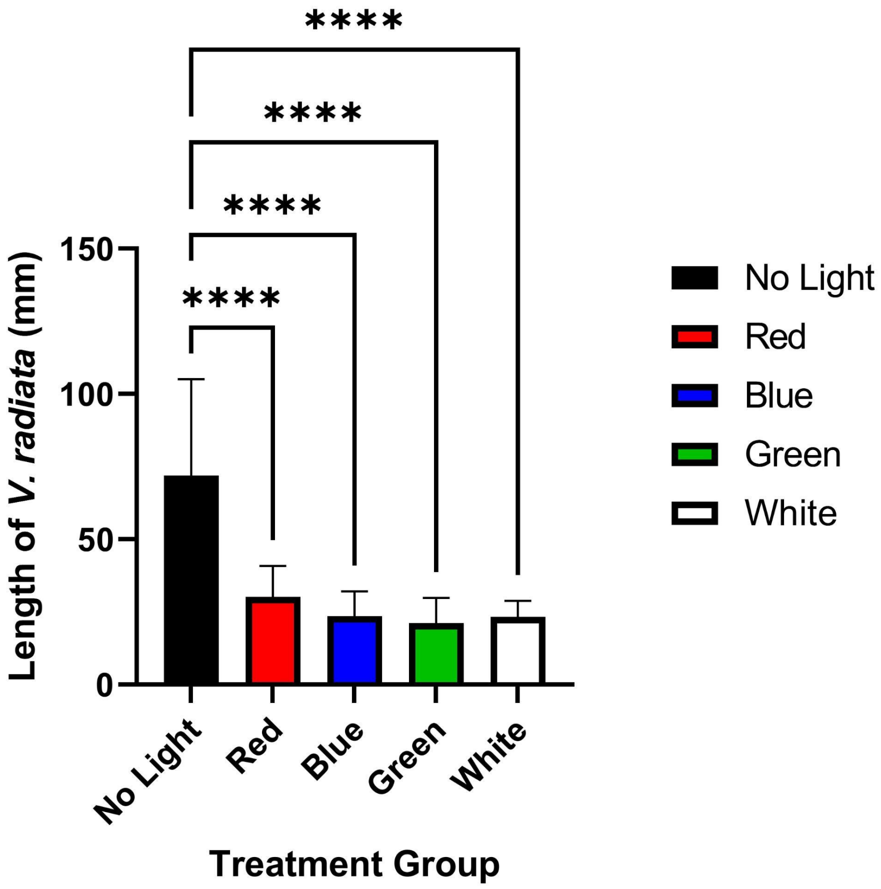

Figure 3. The average radicle length of *V. radiata* at different wavelengths after a week. This graph uses 95% confidence intervals. The asterisks denote a significant difference between treatments. Using an alpha of 0.05, the no light treatment significantly outperformed the others (p<0.0001). Differences between Red (n= 20), Blue (n= 20), Green (n= 20), and White (n= 19) were insignificant (p>0.05).

Discussion

Since Bu et al. (2016) demonstrated how the light had a positive effect on seed germination, similar results were expected. However, Tukey-Kramer analysis revealed that growth in the no-light treatment exceeded the other groups by a substantial amount. The red group did slightly outgrow the blue, white, and green treatments, however, this discrepancy is insignificant. Since the seeds perform better in response to darkness, the initial hypothesis can be refuted. There is no statistical significance proving that combinations of wavelengths induce greater radicle lengths in mung beans. Therefore, the null hypothesis cannot be rejected. Based on the results, *V. radiata* seeds can successfully germinate without direct illumination. Rather, it appears that exposure to light hindered the growth of the radicle.

According to the research of Alabadí et al. (2004), many plants experience skotomorphogenesis, which is when seedlings turn pale from a lack of light and rapidly grow in response. During this phase, they contain fewer chloroplasts and reduce the expression of light-regulated genes. Skotomorphogenic development is maintained until the plant reaches the soil surface where light is abundant. Therefore, it is likely that the no-light group was undergoing this process. This makes sense since their average radicle lengths were much longer than the rest of the treatments. The seedlings transition to photomorphogenesis once they have reached the soil surface, which is when exposure to light disables skotomorphogenesis. This evidence coincides with the findings of Silva-Navas et al. (2015) which suggest how root illumination supposedly shortens the root length and promotes the growth of lateral roots. Therefore, it is likely that our red, blue, green, and white treatment groups were in the process of transitioning from skotomorphogenesis to photomorphogenesis. As a result of the different wavelengths of light, all four groups exhibited stunted root growth and an increase in lateral roots. It is proposed that skotomorphogenesis evolved as a control mechanism to ensure photomorphogenesis does not occur until adequate light conditions are met. Hence, keeping the seedlings in this state can significantly increase their growth.

This experiment was repeated twice using the same procedure. During the first week, the paper towels had been found to be completely dried out over the weekend, and the mung beans exposed to the light had been burnt. This was partly due to a lack of water, and because the sides of the resealable baggies were cut open for easier transfer of the mung beans in and out of the bags for measurements and photos. An attempt to revive the mung beans with approximately 2 mL of water per replicate was made, but on the next measuring day it was observed that almost all of the mung beans had mold growth. A potential reason for mold growth is that the initial lack of water had caused the beans to expire over the weekend, and the addition of water gave the mold an ample opportunity to grow. These beans were discarded as future growth would only be observed in the mold. However, from the data we collected from this trial, it was evident that the no light control group was still outperforming the other light treatments, which supports the findings of the final trial and analysis.

One source of error is a bias in selecting *V. radiata* beans to place into each group. Since the beans were soaked in water overnight to allow them to quickly start germination during the experiment, some of the beans selected had initial growth. Researchers were more inclined to choose unopened beans when creating the replicate bags, which prejudices against initially successful beans. As a result, the *V. radiata* radicle lengths were negatively affected and may have generated larger values otherwise, but every group was exposed to this bias; this effect may not be evident in the final results. The effects of this source of error can be reduced by subtracting any initial growth from the final growth before statistical analysis.

Future directions to take this experiment are to analyze the nutritional value and protein content of the beans after germination under specific wavelengths or to observe how a longer period of light exposure would affect the beans.

Conclusion

Though the initial hypothesis indicated all different wavelengths being necessary for optimal radicle growth, key findings suggest that neither red, green, or blue wavelengths showed significantly higher radicle growth than white light. These results reason that none of the wavelength-exclusive groups are advantageous for radicle growth, rather, no light appears to produce the best results for seed germination of *V. radiata*. Overall, this means the initial hypothesis could be rejected, because no light exposure was observed to produce greater growth of the seedling. Since mung beans are so accessible across the world to many communities, it is important to grow them with techniques that will increase their nutritional value. This experiment starts with the first stage, which is at the level of breaking seed dormancy. Future analyses of the seedlings can be done to determine if there are any differences in the protein content from exposure to specific light wavelengths.

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Data Availability

Raw data on radicle measurements are available on request from the authors.

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