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Article

Effects of Different Spectra from LED on the Growth, Development and Reproduction of *Arabidopsis thaliana*

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Abstract: Light is the major source of energy for plants and as such has a profound effect on plant growth and development. Red and blue lights have been considered to best drive photosynthetic metabolism and are beneficial for plant growth and development, and green light was seen as a signal to slow down or stop. In this study, *Arabidopsis thaliana* (*Arabidopsis*) was used to investigate the effects of red, blue and green lights on the growth and development of plants from seed germination to seeding. Results demonstrated that red light showed a promotion effect but blue light a prohibition one in most stages except for the flowering time in which the effect of each light was just reversed. When mixed with red or blue light, green light generally at least partially cancelled out the effects caused by each of them. Results also showed that the same number of photons the plant received could cause different effects and choosing the right combination of different color of lights is essential in both promoting the growth and development of plants and reducing the energy consumption of lighting in plant factory.

Keywords: *Arabidopsis thaliana*; light quality; photomorphogenesis; LED; plant growth

1 Introduction

In the past few decades, studies have shown that light is a very important environmental factor in controlling plant growth and development, including germination, seedling development, flowering, seeding and so on [1]. It is increasingly used as an environment-friendly method to manage horticultural crops, especially in closed plant factories. There are three main parameters to be considered in plant lighting process: light quantity, quality, and duration. Each of these parameters may have different effects on plant growth and development. As an energy provider, light is used in photosynthesis by chlorophylls a and b and carotenoids as accessory light harvesting pigments. However, as a signaling source, it is perceived through a suite of at least four photoreceptors-cryptochrome and phototropin which perceive the light in blue and ultraviolet-A regions, phytochrome that monitors the red (Pr) and far-red (Pfr) regions of the light spectrum, and the ultraviolet-B photoreceptor [2].

As a popular model organism in plant biology and genetics, the influence of light on the growth and development of *Arabidopsis* has also been extensively studied. Illumination with UV-A was found to be

able to reduce the photosynthetic pigment content, the maximum photochemical quantum yield of photosystem II (*Fv/Fm*), and the effective quantum yield of photosystem II [3]. However, the preillumination of the *Arabidopsis's* leaves with red light could significantly reduce in the inhibitory effect of the UV-radiation on the photosystem II activity in the wild-type plants. The responses of regulatory genes to different light sources were investigated and UV-A light was found to be able to decrease the transcript level of all the structural genes compared to the dark control [4]. Research also found that flowering in *Arabidopsis* was accelerated by a reduced ratio of red light to far-red light (*R/FR*) in *Arabidopsis* and the *Bla-6* ecotype did not flower significantly earlier in response to low *R/FR*, but was still able to display other features of shade avoidance [5]. The changes in light intensity or quality induced changes in the redox state of the photosynthetic electron chain that acts as a trigger for compensatory acclimation response [6]. It was also found that decreasing the *R/FR* ratio significantly increased petiole elongation and leaf area expansion of Columbia *Arabidopsis*'s seedlings [7].

Despite of this, most of the researches on the characteristics of *Arabidopsis* were mainly focused on gene or cell level. As a complex multicellular eukaryote, *Arabidopsis* was the first plant to have its genome sequenced with about 135 mega base pairs [8], and is a popular tool for understanding the molecular biology of many plant traits, including flower development, morphogenesis and light sensing. Much work has been done to assign functions to its 27000 genes and the 35000 proteins they encode [9]. *Arabidopsis* was also used extensively in the study of the genetic basis of phototropism, chloroplast alignment, stomatal aperture, and other blue light-influenced processes [10]. These traits respond to blue light, which is perceived by the phototropin light receptors, and cryptochrome as another blue light receptor of *Arabidopsis*, which is especially important for light entrainment to control the plants' circadian rhythms [11].

Nevertheless, although it is regarded as a model plant, we found that studies on the properties of *Arabidopsis* were mainly focused on the cell or gene level and very few were on the morphological properties of it grown under different qualities of light. At the same time, recent advances in LEDs make them possible to be used under customized-tuned wavelengths for plant growth [12]. Based on these observations, this study investigated the growth and development of *Arabidopsis* under different LED light qualities to investigate morphological responses of *Arabidopsis*. The experimental results showed that in most of the cases, red and blue lights had an opposite effects on the growth and development of *Arabidopsis* and green light would generally at least partially cancel out the effects caused by either red or blue light.

2 Materials and Methods

2.1 Culture Methods

The cultivar of wild-type *Arabidopsis thaliana* ecotype *Columbia* was used in this study. *Arabidopsis thaliana* seeds were sterilized with 0.1% Triton X-100 and 50% ethanol for 5 min before rinsing three times in water and being stored at 4°C for 72 h. 21 glass culture dishes, which were divided into seven groups (Φ = 80 mm, numbered 1-1, 1-2, 1-3, 2-1, 2-2, 2-3, …, 6-1, 6-2, 6-3, CK-1, CK-2, CK-3, respectively), were used and filled with roseite then covered by two-layer filter papers. 40 seeds were sowed in each of these glass culture dishes. These glass culture dishes were then placed in a plant growth shelf (with several identical chambers) with different light spectra at 80 μmol/(m^2•s) photosynthetic photon flux density (*PPFD*) for one week at a 16 h light/8 h dark photoperiod. A constant 24°C room temperature and a 70% relative humidity were maintained.

Seven days after sowing the seeds, seedlings were transplanted to a flowerpot filled with a 3:2:1 mixture of nutrition soil, roseite and perlite. Then the seedlings were shifted to their own plant growth shelf chambers under the illumination of 140 μmol/(m^2•s) *PPFD* and different light qualities at a 16 h light/8 h dark photoperiod, 24°C room temperature, and a 70% relative humidity.

2.2 Plant Growth Chambers and Light Treatments

Although there are varies of plant growth chambers available in the market, most of them do not take the light uniformity nor the adjustment of light quality into consideration. Nonuniform distribution of light in a plant growth chamber will lead to an inaccurate data collection [13]. Therefore, it is necessary to carefully design the plant growth chamber with high light uniformity to ensure the high-accurate manipulation of plant growth. We chose a type of waterproof LED bar which has three adjustable wavelengths at the same time (Red: 660 nm, Green: 525 nm, Blue: 446 nm) as the light source in the plant growth chamber. What made it special was that the three LED chips with different colors were installed in a single chip frame, and in every chamber of the cabinet it could achieve higher light uniformity when compared with the exiting chambers. Fig. 1 shows the spectra for red, green and blue LEDs acquired by the hand-held spectrometer HR-350 from HiPoint Corporation (Taiwan, China) as shown in Fig. 1D.

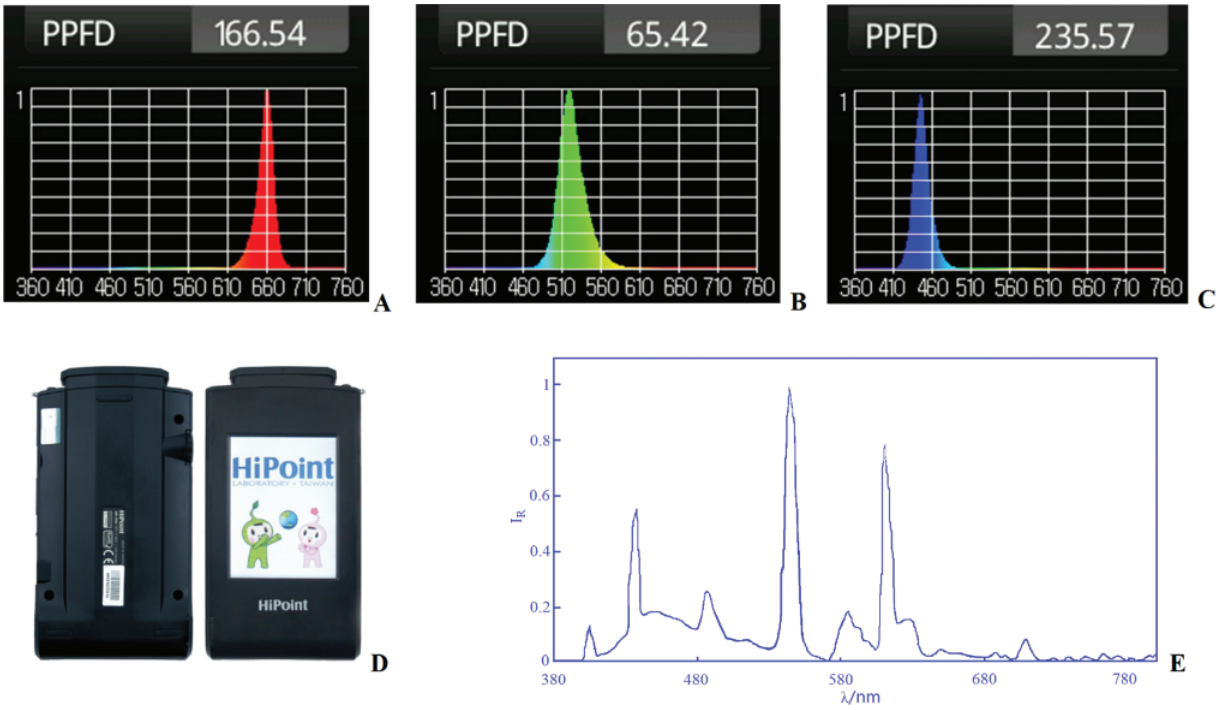

Figure 1: The light spectra of LEDs and fluorescent lamps. A: The light spectra for red LED; B: The light spectra for green LED; C: The light spectra for blue LED; D: Illustration for the hand-held spectrometer HR-350; E: The light spectra for white fluorescent lamps

As shown in Tab. 1, six groups of treatments were administered with LED light, with peak wavelengths of red, green and blue were set at 660 nm, 525 nm and 446 nm, respectively, and the CK group was administered with 6212K Philips white fluorescent lamps whose spectra are shown in Fig. 1E. The *PPFD* used in our experiments was set at 80 μmol/(m^2•s) in germination procedure and 140 μmol/(m^2•s) after transplanting and measured by the hand-held spectrometer HR-350. In all of the treatments, the duration of light and the total *PPFD* were kept constant, but the proportions of R, G and B were different at each treatment. Under the same light intensity, the spectra of different treatment groups are made up of different proportions of three-color light. For example, treatment 3 is composed of 6 parts of red light, 1 part of green light and 3 parts of blue light. During the germination procedure, the intensity of the red, green, and blue light is 48 μmol/(m^2•s), 8 μmol/(m^2•s), and 24 μmol/(m^2•s), respectively, with a total

Table 1: The proportions of R, G, B and *PPFD* of light treatments used in this study

Treatment		1	2	3	4	5	6	CK
R:G:B		10:0:0	8:2:0	6:1:3	3:1:6	0:2:8	0:0:10	White
PPFD μmol/(m^2•s)	Germination	80 μmol/(m^2•s)						
	After transplanting	140 μmol/(m^2•s)						

intensity of 80 μmol/(m^2•s); and the intensity of the red, green, and blue light after transplanting is set to 84 μmol/(m^2•s), 14 μmol/(m^2•s) and 42 μmol/(m^2•s), respectively, with a total intensity of 140 μmol/(m^2•s). Fig. 2 shows the illustrations for experiment.

2.3 Growth Parameters Measurement

When measuring the growth parameters, ten samples in each group are randomly chosen. The plants are dug out from the cultivation soil, washed, and then the surface moisture is blotted with filter paper. The lengths of the main roots and hypocotyls are measured using the WinRHIZO Root Analysis System from Regent Instruments Inc. (Quebec, Canada) in conjunction with an EPSON Expression 11000XL scanner (Nagano-ken, Japan). The leaf length and height of *Arabidopsis* are measured by electronic vernier calipers. The height of the *Arabidopsis* is measured with flower on the day of the first flower of plant. The total leaf length means the sum of leaf length and petiole length. The seed weight is measured by electronic balances. In the end, the average of the measured values of the ten samples is taken as the final measured value of the corresponding parameter.

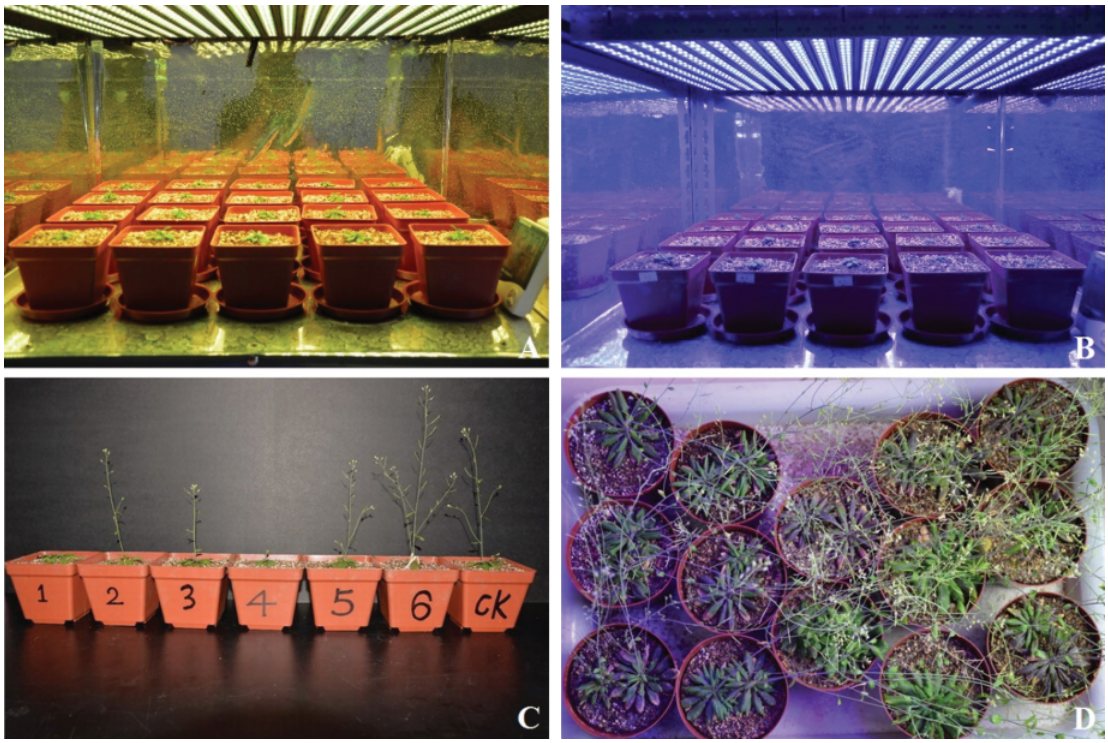

Figure 2: The illustration of experiments. A: illustration of light environment for treatment 2; B: illustration of light environment for treatment 4; C: contrasting photo for all treatments four weeks after transplanting; D: reproduction photo of *Arabidopsis* thaliana in experiment

2.4 Chlorophyll Fluorescence Analysis

Chlorophyll fluorescence measurements were performed using a PAM-2500 fluorimeter manufactured by WALZ Corporation (Forchheim, Germany). The whole plants were dark adapted for 20 min prior to illumination exposure and the fourth true leaf was selected as the subject of measuring. The calculations of *Fv/Fm*, *qP*, *Fv'/Fm'*, *Y(NPQ)* and *Y(II)* were referred to [14] methods. *Chla* (*Chlorophyll-a*), *Chlb* (*Chlorophyll-b*) and carotenoid contents were measured and calculated based on the absorption characteristics of chlorophyll extract solution for visible spectrum, *Beer-Lambert* Law, and the corresponding equations. During the measurement, the first step was to choose some mature leaves and remove all petioles. Then they were cut into pieces and 0.1 g leaves were put to a centrifuge tube (10 ml) filled with mixed solution (ratio of the volume of acetone: absolute ethyl alcohol: deionized water = 4.5:4.5:1) to 10 ml which kept in dark for 48 h. These solutions were then diluted with the mixed solution to 25 ml, and the original mixed solution was set as the reference in the measurement. The specific absorption coefficients of the diluted solution under the 663 nm, 645 nm, 440 nm illumination were measured by a UV-2700 ultraviolet spectrophotometer from Shimadzu Corporation (Kyoto, Japan) and denoted as OD_{663}, OD_{645} and OD_{440}, respectively. Finally, the OD_{663}, OD_{645} and OD_{440} were put into the following four equations [15] to work out the final results of contents of *Chla*, *Chlb* and carotenoid in mg/g, where V and W represent volume of the solution (ml) and raw weight of the sample (g), respectively. The measurements of all physiological indexes of the plant were repeated three times; data were recorded and analyzed by SPSS Statistics 22.0 and curves were plotted by Origin 9.0.

$$P_{\text{Chla}}(\text{mg}) = \frac{(12.7 \times OD_{663} - 2.69 \times OD_{645}) \times V}{(1000 \times W)} \tag{1}$$

$$P_{\text{Chlb}}(\text{mg}) = \frac{(22.9 \times OD_{645} - 4.68 \times OD_{663}) \times V}{(1000 \times W)} \tag{2}$$

$$P_{\text{Chl(a+b)}}(\text{mg}) = \frac{(20.21 \times OD_{645} + 8.02 \times OD_{663}) \times V}{(1000 \times W)} \tag{3}$$

$$P_{\text{Carotenoids}}(\text{mg}) = \frac{(4.7 \times OD_{440} - 0.27 \times (20.21 \times OD_{645} + 8.02 \times OD_{663})) \times V}{(1000 \times W)} \tag{4}$$

3 Results

3.1 Effects of Different Spectra on Arabidopsis Germination

Effects of different light qualities on the germination of *Arabidopsis* were investigated using the plant growth cabinet with different ratios of R, G, and B at 24°C for a week with 16 h light and 8 h dark and the results were shown in Tab. 2. Compared to the CK, the germination rates in groups 1 and 2 were slightly higher and rose by 2.5%, 1.11% and those in groups 3, 5, 6 were decreased by 1.94%, 0.28%, and 7.5%, respectively. It was obvious that the germination rate in group 6 was the lowest. Between groups 1 and 2, the germination rate of the latter was 1.39% lower, and for groups 5 and 6, the germination rate of the former was 7.22% higher than that of the latter. Germination rate in group 3 was 1.94% lower than that in group 4. From the results given in Tab. 2, we can see that different spectra had different influence on the germination rate of *Arabidopsis*. Red light could promote seed germination and blue light had an opposite effect, whereas green light may not only weaken the promotion effect of the red light, but also weaken the inhibition of the blue light on seed germination.

Table 2: Seed germination rate of *Arabidopsis*

Treatment	Percentage Germination/%
1	98.61 ± 0.96a
2	97.22 ± 1.92ab
3	94.17 ± 0.84c
4	96.11 ± 0.48bc
5	95.83 ± 0.84bc
6	88.61 ± 1.27d
CK	96.11 ± 0.49bc

(Note: Data in the table are the Means ± SD; The different normal letters indicate significant differences among treatments acquired by Duncan's test, $p < 0.05$ and the following tables and figures have the same setting; n = 3.)

3.2 Effects of Different Spectra on Seedling Main Root Length and Hypocotyl Length

Roots as an important nutritive organ could provide plants with essential nutrients and moisture in the process of plant growth and development, and the main root length is an important reference for the evaluation of plant health conditions. Especially for seedlings, the development degree of roots would directly affect the survival rate after being transplanted. Fig. 3 shows the seedling's main root length and its hypocotyl length after illuminated by different quality of light. We can see from Fig. 3A that, compared to the CK, the main root length in groups 1 and 2 were increased by 61.85% and 37.04% ($p < 0.05$), respectively. In contrast, those in groups 3, 4, 5, and 6 were decreased by 35.80%, 43.21%, 50.25%, and 58.89% ($p < 0.05$), respectively.

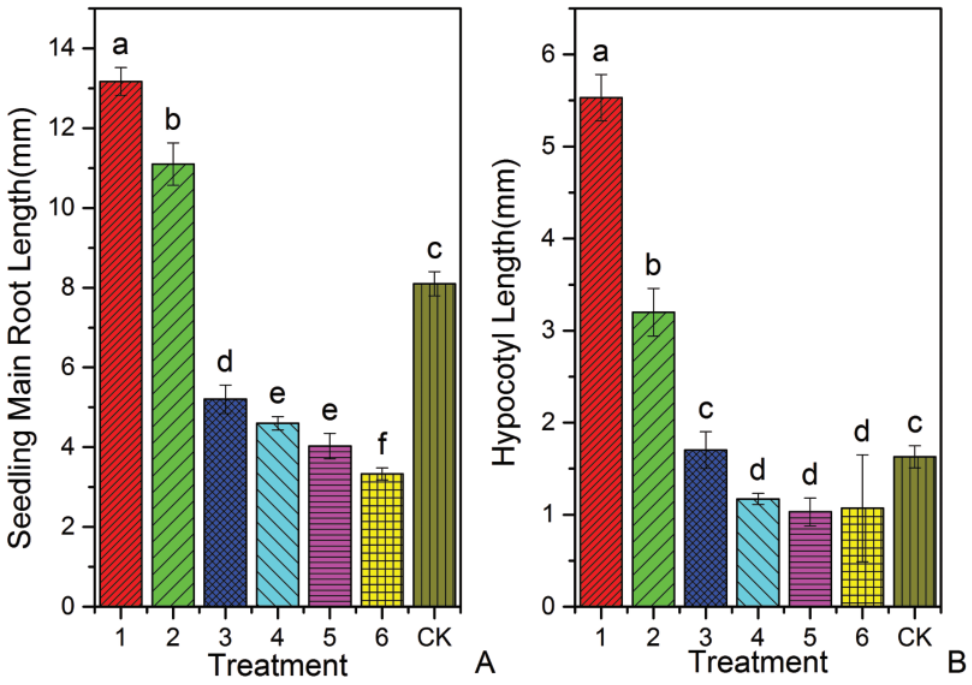

Figure 3: The effect of different spectra on the seedling's main root length and the hypocotyl length of *Arabidopsis* (n = 3)

Among the four groups 1, 2, 5 and 6, seedling's main root length in group 2 was about 15.33% shorter than that in group 1 and that in group 5 was about 21.02% higher than that in group 6. This indicates that red light had contributed to the elongation of seedling's main roots of *Arabidopsis*, and blue light had an opposite effect that inhibited the roots' elongation. Nevertheless for green light, it had a mixed effect on promoting or inhibiting roots' development. It had a restraint effect when green light was mixed with the red light, whereas there was a promotion effect on root elongation when it was mixed with the blue light. When green light was mixed with both the red and blue lights, their trend was the same as it was mixed with single red or blue, that is, the higher the content of red, the longer the main root.

From Fig. 3B, we can find that the hypocotyl length in groups 1 and 2 increased by 239.26%, 96.32%, respectively whereas that in groups 4, 5, and 6 decreased by 28.22%, 36.81%, and 34.36%, respectively, when compared with the CK ($p < 0.05$). Between groups 1 and 2, the hypocotyl length of the later was 42.13% shorter. But there were no significant differences among groups 4, 5, and 6 and between group 3 and the CK. On the other hand, the hypocotyl length in group 1 increased by 416.82% when compared with group 6. From these observations, it indicated that red light could promote hypocotyl elongation of *Arabidopsis* as it did for the main roots, and blue light had an opposite effect on it. Nevertheless, green light would inhibit the hypocotyl growth when mixed with the red light, but there was no significant effect on the hypocotyl growth when combined with the blue light. When green light was mixed with both the red and blue lights, their trend was also the same as it was mixed with single red or blue, that is, the higher the content of red, the longer the hypocotyl.

3.3 Effects of Different Spectra on Contents of Chlorophylls A, B and Carotenoid of Arabidopsis

When seedlings were cultivated for 25 days after transplanting, the contents of *chlorophyll a* (*Chla*), *chlorophyll b* (*Chlb*), *chlorophylls a+b* (*Chl(a+b)*), and carotenoid were measured and shown in Fig. 4A. The changes of the *Chla*/*Chlb* ratios under different lights are shown in Fig. 4B. The variation of contents of chlorophylls and carotenoid under different lights when compared to the CK was shown in Fig. 4C. When compared to CK, we found that the contents of *Chla* and *Chlb* in groups 1, 2, and 6 decreased by 22.68%, 14.79%, 7.89% and 22.84%, 20.37%, 14.51%, respectively, whereas the contents of *Chla* and *Chlb* in groups 4 and 5 increased by 8.02%, 17.92% and 6.79%, 8.83%, respectively. Nevertheless, the contents of carotenoid decreased by 16.67%, 9.17% for groups 1 and 2, and increased by 15.83%, 16.67%, and 57.5% for groups 4, 5, and 6, respectively. Between groups 1 and 2, the content of *Chla* of the latter increased by 11.66% compared with the former, while there were no significant differences on contents of *Chlb* and carotenoid.

When compared to group 6, contents of *Chla* and *Chlb* in group 5 significantly increased by 28.03%, 38.99%, respectively. But quite the opposite, the content of carotenoid in group 5 dramatically decreased by 25.93%. Between groups 3 and 4, the contents of *Chla*, *Chlb* and carotenoid of the latter increased by 13.57%, 6.13%, and 13.93%, respectively. However, we can find that the ratio of the content of *Chla* to the total content of *Chl(a+b)* (*Chla*/*Chl(a+b)*) was relatively stable, which was about 71.28% ± 1.35%. From the above analysis we can see that red and blue lights had a similar effect in inhibiting the accumulation of *Chla* and *Chlb*, and red light had a stronger inhibition effect than blue light. However, red light had an inhibition and blue light had a promotion on the formation of carotenoid. In particular, this kind of inhibition effect of red light on carotenoid was weaker than the formation of *Chla* and *Chlb*, and the content of carotenoid increased by more than half when grown under blue light irradiation. There were obvious differences on the formation of *Chla*, *Chlb* and carotenoid when green light was mixed. When mixed with red light, green light could promote the accumulation of *Chla* or weaken the inhibition of red light, whereas it had no significant effect on the formation of *Chlb* and carotenoid. When combined with blue light, green light could cause a significant promotion on the contents of *Chla* and *Chlb*, and significantly inhibit the formation of carotenoid. This indicates that green light could promote

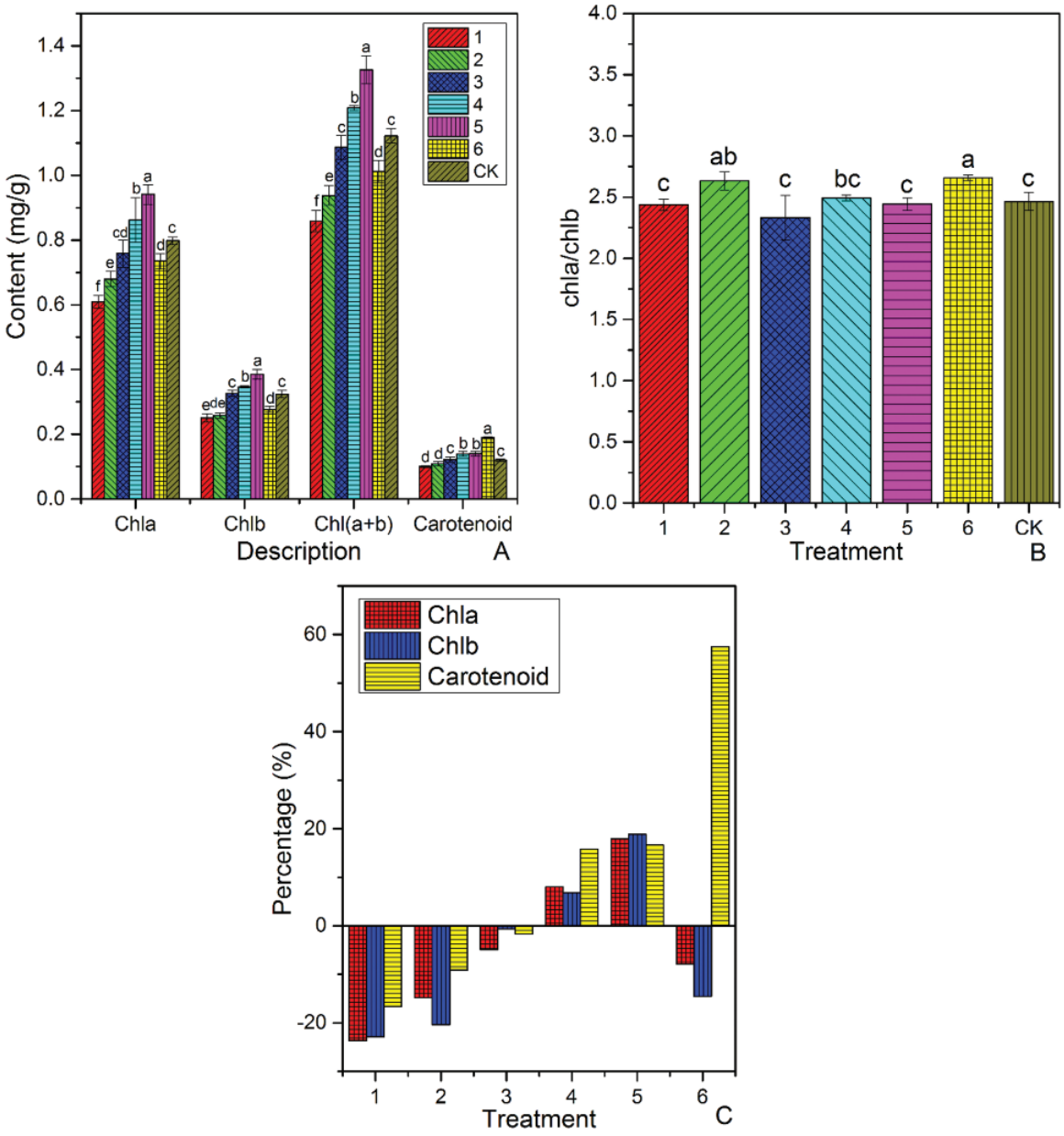

Figure 4: The effect of different spectra on contents of *Chla*, *Chlb* and carotenoid of *Arabidopsis*. A: the changes of contents under different lights; B: the changes of the *Chla*/*Chlb* ratios under different lights; C: the variation of contents compared to the CK under different lights (n = 10)

the formation of *Chla*, and it could promote the formation of *Chlb* and carotenoid only when combined with blue light. When green light was mixed with both red and blue lights, their trend was the same as it was mixed with single red or blue again.

3.4 Effects of Different Spectra on Leaf Length of Arabidopsis

Leaf is one of the most important organs for plants to absorb solar energy. Fig. 5A shows the distributions of petiole and leaf length of *Arabidopsis* grown under different light qualities. In this figure

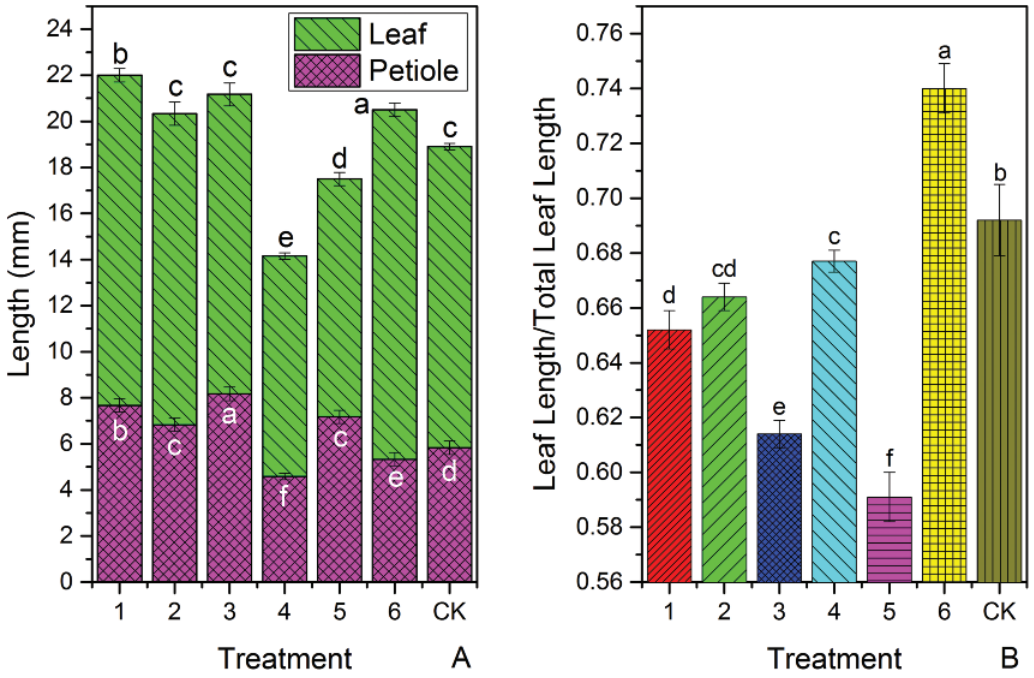

Figure 5: The leaf length of *Arabidopsis* grown under different lights after 25 days (n = 10)

the leaf and petiole lengths were measured from the fourth pair of true leaves. When compared to the CK, except for groups 4 and 6, the petiole lengths in groups 1, 2, 3, and 5 increased by 31.56%, 17.15%, 40.14% and 22.98%, respectively. For groups 4 and 6, they decreased by 21.44%, 8.58%, respectively. Between groups 1 and 2, the petiole length of the latter was 10.95% shorter than that of the former, and that in group 5 was about 34.52% longer than that in group 6. As for the leaves, the lengths in groups 1 and 6 increased by 9.56%, 15.98%, and those in groups 4 and 5 decreased by 26.76%, 21.02%, respectively when compared to the CK. While there were no obvious differences between groups 2 and 3 when compared with the CK. Between groups 1 and 2, the latter's leaf length was 5.79% shorter than the former, and the leaf length in group 5 was about 31.91% shorter than that group 6.

Figure 5B shows the ratio of leaf length to total length of leaf of the *Arabidopsis* grown under different light qualities. From this figure we can see that group 6 was the only one in which the ratio was 6.94% higher than that of the CK. The ratio in group 5 was the lowest which was 14.60% lower than that of the CK. Of groups 1, 2, 3 and 4, their ratio decreased by 5.78%, 4.05%, 11.27% and 2.17%, respectively compared to that of the CK. Compared with group 1, the ratio in group 2 slightly increased by 1.20%. Between groups 5 and 6, the ratio of the former was 20.14% lower.

3.5 Effects of Different Spectra on the Chlorophyll Fluorescence Parameters of Arabidopsis

Chlorophyll fluorescence analysis is a unique tool in reflecting the properties of photosynthetic process through light absorption, transmission, distribution, dissipation and conversion. Let *Fm* represent the maximum chlorophyll fluorescence yield when *photosystem II* (*PSII*) reaction centers are closed by a saturation pulse and *Fo* denotes the basic chlorophyll II fluorescence yield recorded with low measuring light intensities. Then, *Fv = Fm-Fo* is the variable fluorescence. The maximum photochemical quantum yield of *PSII Fv/Fm* is a measurement index on *PSII* photoinhibition. Fig. 6 shows the changes of chlorophyll fluorescence parameters of *Arabidopsis*, which were measured by the PAM-2500 fluorimeter manufactured by WALZ Corporation (Forchheim, Germany) when they were transplanted for 25 days, under different lights, including maximum photochemical quantum yield of *PSII* (*Fv/Fm*), photochemical quantum yield of

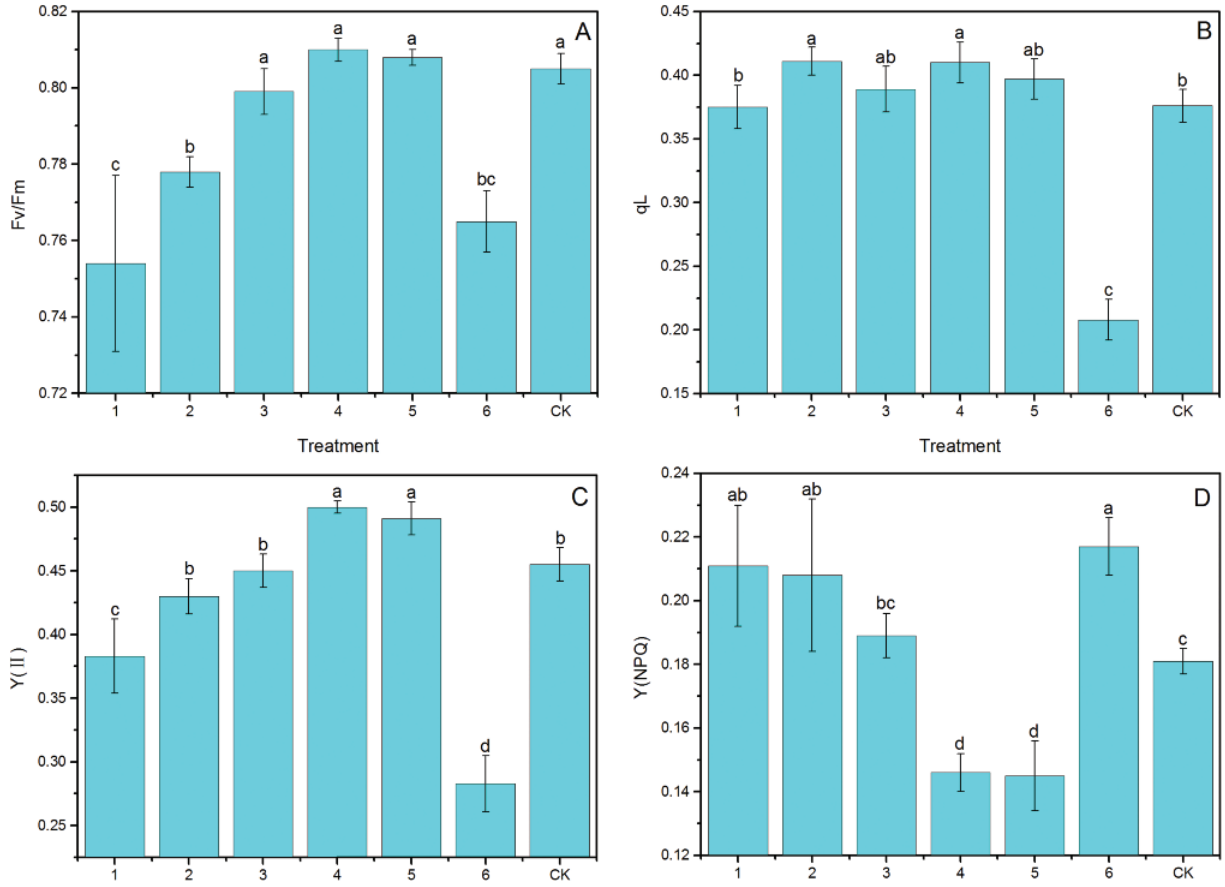

Figure 6: The changes of chlorophyll fluorescence of *Arabidopsis* grown under different lights after 25 days (n = 10)

PSII (*Fv'/Fm'*), effective photochemical quantum yield of *PSII* (*Y(II)*), quantum yield of non-photochemical energy conversion in *PSII* (*Y(NPQ)*), coefficient of photochemical fluorescence quenching (*qL*), coefficient of non-photochemical fluorescence quenching (*qN*), coefficient of photochemical fluorescence quenching (*qP*), and electron transport rate (*ETR*) etc.

Compared to the CK, the ratios of *Fv* to *Fm* (Fig. 6A) in groups 3, 4 and 5 had no significant difference, while the ratios in groups 1, 2 and 6 decreased by 6.34%, 3.35%, and 4.97%, respectively. Similar to the above results is that the ratios of *Fv'/Fm'* (*Fv' = Fm'-Fo'*, *Fm'* represents maximum chlorophyll fluorescence yield when *PSII* reaction centers are closed by a strong light pulse, *Fo'* denotes minimum chlorophyll fluorescence yield) in groups 3, 4 and 5 had no clear difference compared to that of the CK, and the ratios in groups 1, 2 and 6 decreased by 9.86%, 6.23%, and 5.07%, respectively. Therefore, the changes of *Fv/Fm* and *Fv'/Fm'* are quite similar. Comparing the *Fv/Fm* and *Fv'/Fm'* in groups 1 and 2, we found that adding green light to red resulted in the ratio to increase by 3.18% and 4.02%, respectively. When green light was added to blue, the ratios increased by 5.62% and 8.09%, respectively. Therefore, the amount of increase was more obvious by adding green light to the blue light than to the red light. As for the *qL* (Fig. 6B) and the *qP*, they showed similar trends under different lights. The value of *qL* in groups 2 and 4 had increased by 9.31% and 9.04%, and that of *qP* in group 4 increased by 6.98% compared with the CK. But the values of *qL* and *qP* in group 6 decreased by 44.68% and 34.45%, respectively. Comparing the values of *qL* and *qP* in groups 1 and 2 and groups 5 and 6, we found that adding green light to either red or blue light would

increase the values of both *qL* and *qP* (increased by 9.60%, 8.14% and 90.87%, 60.42%, respectively) but the amount of increase was much more obvious by adding it to blue light than to red light.

Considering *Y(II)* (Fig. 6C) and *ETR*, compared to the CK, the values of them in groups 4 and 5 increased by 9.89%, 7.91% and 9.75%, 7.70%, respectively. At the same time the values of them in groups 1 and 6 decreased by 37.86%, 37.80% and 16.05%, 15.82%, respectively. When green light was added to red, the values of *Y(II)* and *ETR* increased by 12.27% and 12.40%, respectively. When it was added to blue, they increased by 73.50% and 73.31%, respectively. The *ETR* showed similar tendency as *Y(II)*. Therefore, the amount of increase was much more obvious by adding the green light to blue than to red. However, the ratios of *ETR* to *Y(II)* were magically identical irrespective of the light quality. When comparing the results in groups 1 and 2, we found that the values of both *Y(NPQ)* (Fig. 6D) and *qN* did not have a significant difference after adding the green light. However, when adding the green light to the blue, *Y(NPQ)* decreased very significant.

3.6 Effects of Different Spectra on the Height of Arabidopsis

Fig. 7A shows the changes of height of *Arabidopsis* grown under different lights. It can be found that there were significant differences on the height of *Arabidopsis* among groups 1 to 6 compared to the CK. The

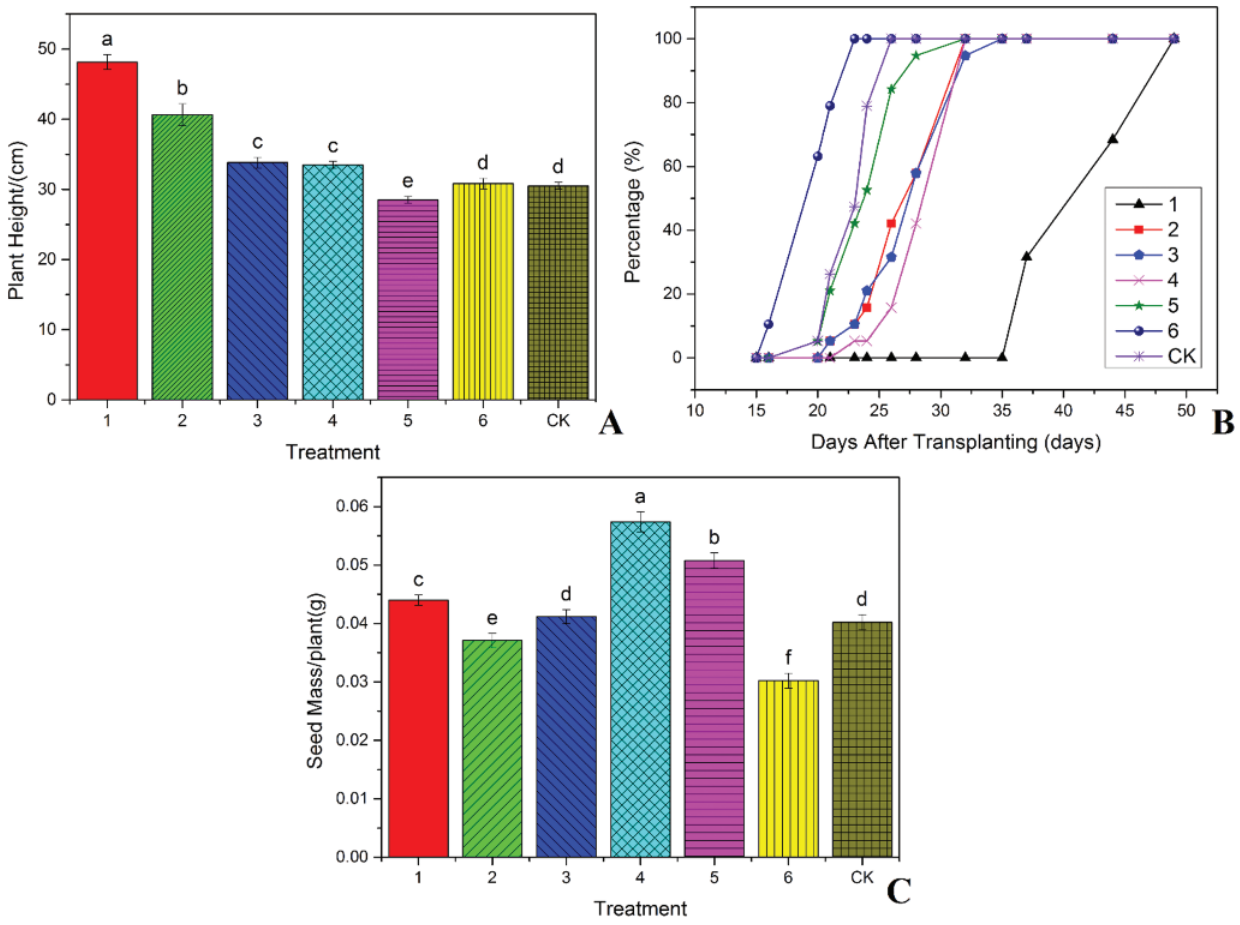

Figure 7: Illustration of plant height, flowering time and seed weight in experiment. A: The distribution of plant height of *Arabidopsis* grown under different lights; B: Differences of flowering time of *Arabidopsis* grown under different lights; C: The distribution of seed weight of *Arabidopsis* grown under different light qualities (n = 10)

height in groups 1, 2, 3, and 4 increased by 57.93%, 33.34%, 10.92%, and 9.84%, respectively. The height in group 5 was about 6.56% lower than that of the CK, but there was no significant difference between group 6 and the CK. This indicated that red light could promote the growth, while blue light had no obvious effects on this. When green light was mixed with red or blue light, it would inhibit the height of *Arabidopsis* which decreased by 15.57%, 7.56%, respectively. The inhibition effect was stronger when mixed with red light than with blue light. This indicated that *Arabidopsis*'s growth and development could be inhibited by green light.

3.7 Effects of Different Spectra on the Flowering Time of Arabidopsis

Flowering is one of the most important processes in higher plants' lifecycle. Flowering in many plants is a photoperiodic phenomenon. From Fig. 7B, it can be seen that *Arabidopsis* grown under different lights showed very different in flowering times. Group 6 was the earliest to blossom in which the first flower appeared on the 14th day after transplanting and group 1 was the latest in which it spent 35 days for the first flower to appear. Compared with the CK, the *Arabidopsis* in group 6 flowered 4 days in advance, and group 1 flowered 17 days later. The flowering time in group 2 was 11 days earlier than that in group 1, and group 5 was 4 days later than group 6. While there were no obvious differences between groups 3 and 4 on flowering time. Thus, red light would cause a great delay on flowering, and blue light would significantly accelerate flowering.

3.8 Effects of Different Spectra on the Seed Weight of Arabidopsis

Seeding marks an end of a plant's lifecycle and is also one of the most important processes in angiosperm's lifespan. We can see from Fig. 7C that apart from group 3, all the other groups had obvious differences from the CK. Seed weight in groups 1, 4 and 5 increased by 9.45%, 42.79%, and 26.37%, respectively. That in groups 2 and 6 decreased by 7.71% and 24.88%, respectively. When green light was added to the red, the seed weight in group 2 decreased by 15.68% compared with that in group 1, but when it was added to the blue, that in group 5 increased by 68.21%. Based on the results, it indicated that red light alone could promote seed production of *Arabidopsis*, but the effect of promotion was not obvious. While blue light alone could inhibit the seed production and the inhibition effect was stronger than that of the red light.

4 Discussion

The effects of lights on *Arabidopsis*'s germination, seedling's main root length, hypocotyl length, leaf length, flowering time, seed weight, and contents of chlorophyll and carotenoid were investigated through the mediation of various light qualities. In germination stage, red light could increase germination rate and blue light would decrease the rate. For green light mixed with red and green light, it may not only weaken the promotion of red light on germination, but also weaken the inhibition of blue light on germination. In the seedling stage, the situation was similar to the case in germination. The Kim HH's research [16] discovered that under certain *PPFD* a small amount of green light incorporated in red and blue light can enhance plant growth, but it also shows that the incorporation of a large amount of green light can significantly inhibit plant growth. The results of a small amount of green light for enhancing growth are different from this article. It may be due to the fact that green light can better penetrate the plant canopy and potentially increase plant growth by increasing photosynthesis from the leaves in the lower canopy, which is mentioned in that article. However, in our research, the plant is *Arabidopsis* and almost all the leaves can receive light uniformly. Moreover, our research uses three narrow-spectrum monochromatic LEDs to form the irradiated light, which is different in spectrum from other experiments. The monochromatic green light has been found the stimulation function on stem elongation [17,18]. That result is acquired based on a short-time radiation of monochromatic green light whereas our research uses a small amount green light mixed with red and blue light for a relatively long time in that study item.

Thus, there is some difference in the result. Furthermore, those articles mention that seedlings grown under green, red, and blue lights together are longer than those grown under red and blue alone, which also means the green light mixed with red and blue lights will influence the procedure of seedlings. The Folta's research also studies the growth regulation function of light [19]. The study shows the higher red light proportion can increase petiole length, which is consistent with the results of this article. For chlorophyll and carotenoid contents in the *Arabidopsis*, which were measured 25 days after transplanting, we found that red and blue lights could both slightly inhibit the formation of *Chla* and *Chlb*. While blue light could accelerate the formation of carotenoid, red light had an opposite effect on this. Green light could promote the formation of chlorophylls when mixed with either blue or red light but didn't had so much influence on the formation of carotenoid.

For the chlorophyll fluorescence analysis results, our experiments showed that the trend of *Fv/Fm*, *Fv'/Fm'*, *Y(II)*, and *ETR* are the same, in which the value of these parameters dramatically increased when green light was added to the blue but only slightly when it was to the red. In addition, the trend of *qL* and *qP* are similar, in which the value of them dramatically increased when green light was added to the blue but only very slightly when it was to the red. Finally, the behavior of *Y(NPQ)* and *qN* are similar, in which the value of them decreased when green light was added to the blue but did not change much when it was to the red. Therefore, red or blue light alone could cause *ETR* and *Fv'/Fm'* an obvious decline, which may be caused by photoinhibition and then an exception occurs in *Arabidopsis* photosynthesis process, which leads to electron transport more difficult in the changes of contents of *Chla* and *Chlb* grown under these light qualities. A further finding was that no matter how the quality of light changed, the ratio of *ETR* to *Y(II)* was very stable and stayed at 53.36 ± 0.05. This demonstrated that there is a dynamic equilibrium mechanism under different light qualities. The Hogewoning's research [20] uses cucumber as studying target and concluded that blue light during growth is qualitatively required for normal photosynthetic functioning. The research shows the addition of blue light can increase the ratios of *Fv/Fm* and *chla/chlb*, which is consistent with the experimental results in our article. But the increase of *chla/chlb* in our article is not obvious. This may be due to the light in our article mixed with a small amount of green light. Moreover, the far-red light is proved to be needed for efficient photochemistry and photosynthesis [21]. However, due to the limitation of experimental device, our research does not involve the spectrum of the far-red light and we will continue to do the research in the future.

Flowering was well known as a photoperiodic phenomenon in many plants that is mainly a response to the duration of light, while our experiments showed that different spectra also had significant effects on the flowering time. Monochromatic red light obviously delayed the flowering time, and monochromatic blue light significantly accelerated flowering. When green light was mixed with red light, it could weaken the inhibition of red light as well as the promotion of blue light on flowering. The effect of LEDs on wheat with different spectral quality and intensity is also studied [22]. The LED spectrum in that article has some differences from our article. However, the monochromatic blue light in that paper can delay the flowering time of wheat, which is contrary to the results in our paper. This may be caused by different genes of different plant species, or it may be caused by the planting environment and different spectra, and further research is needed in the future. In this paper, blue light accelerates flowering, but provides the least weight of the seeds. That may be due to the rapid flowering, which leads to insufficient organic accumulation in the early stage of the plant.

5 Conclusion

We had investigated the growth and development of *Arabidopsis* under the same fluence rate and duration of illumination but enhanced blue, red, and green lights. Morphological responses of *Arabidopsis*, such as germination, seedling development, flowering, seeding etc., biochemical contents, such as chlorophylls and carotenes, and photosynthetic properties were tested with different light

qualities. The results showed the following features: 1) When red or blue light was used alone, they generally had opposite effects on tested parameters, i.e., if one was enhancing a result, the other would be eliminating it; 2) When a small amount of green light was mixed with red or blue light, it generally eliminated the effects caused by the red or blue light alone. From the findings above, we conclude that green light may mediate the effects caused by red or blue light alone. As the *PPFD*s and the durations of illumination used in our experiments are all the same, the results showed that the same number of photons could have different effect on plants. Therefore, the appropriate choice of the right combination of different color of lights is essential in both promoting the growth and development of plants and reducing the energy consumption of light in plant factory.

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