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Effect of RGB LED Pulse Lights in Photomorphogenesis of *Brassica chinensis*

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Abstract. The study aims at evaluating the effects of combination of red (R) green (G) and blue (B) LED lights on *Brassica chinensis* under pulse lighting treatment (1 h dark 15 minutes lights) during the day. *Brassica Chinensis* were grown with different photon flux density (PFD) at 100 μmol $m^{-2}s^{-1}$ as treatment 1 (T1) and 50 μmol $m^{-2}s^{-1}$ as treatment 2 (T2). The combination of RGB LEDs ratio was 4:3:3 and plants were hydroponically cultured at ±28°C (day/night), 45% relative humidity, and 400 ppm CO_2 level inside control environment room for 25 days (5 days after sowing). Results showed that *Brassica chinensis* grown under RGB with pulse lighting treatment at 100 μmol $m^{-2}s^{-1}$ promote plant growth and photomorphogenesis.

Keywords: *Brassica chinensis*, LED, Photomorphogenesis.

1. Introduction

Light is not only a primary energy source for photosynthesis but also a vital regulator for numerous processes in plants. The lights intensity and quality are essential for plant growth, morphogenesis and other physiological responses [1]. Combination of red (R) and blue (B) LED light was an effective light source for plant growth and development, and the light spectra, intensities, and durations can easily be controlled by growers in artificial growing environments. Plants appear green because the plant reflects green light. Therefore, green light has been thought to be of no use for plant growth, particularly for photomorphogenesis and photosynthesis. Plants grow normally under sunlight or combined artificial red and blue light [2], but irradiation with green light induces stem elongation [3].

Among the various environmental factors, light is one of the most important variables affecting growth and development of many plant species such as, potato [4], and grape [5], strawberry [6]. Red and blue LEDs were used because red (650–700 nm) waveband is known to be involved in the photosynthetic and blue (450–500 nm) waveband in the photomorphogenic and phototropic responses of plants. It is well known that action spectra have action maxima in the B and R ranges [7]. The absorption of red or blue light by plant leaf pigments was about 90%, and that of green light was about 70–80% [8].

Plant physiological reactions to green light and the effects of green light on plant growth have been investigated, but there is no report of plants being cultivated under pulse lighting treatment in combination of RGB. In this study, two experiments using RGB light emitting diode (LED) with pulse lighting treatment and different light intensities were used to investigate the effect of RGB on *Brassica chinensis* growth and photomorphogenesis.

2. Material and Methods

2.1. Plant Materials and Lights Treatment

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Seeds of *Brassica chinensis* were germinated in sponge cubes (3cm x 3cm) and hydroponically grown for 5 days under a light intensity 100μmol $m^{-2}s^{-1}$ photon flux density (PFD) for 12 h under RB lights. Uniform-sized seedlings of *Brassica chinensis* at the 3-leaf stage were individually raised and mounted into a Styrofoam plate with fifteen holes, and placed in a container (55.5cm x 42cm x 13cm) filled with complete nutrient solution adjusted to pH 6 and an electrical conductivity of 2.5 mS cm^{-1}. The air temperature, relative humidity, and CO_2 levels for all treatments were respectively maintained throughout the experiment at ±28**°C** (day and night), 45% and 400 ppm. Plants were harvested at 30 days after transplant. Combination RGB LED with 4:3:3 array ratios was used as a light treatments with similar pulse lighting system (1 h light 15 minutes dark). The difference intensity expressed as photosynthetic PFD of 100 μmol m^{-2} s^{-1} for treatment 1 (T1) and PFD of 50 μmol m^{-2} s^{-1} for treatment 2 (T2) which was measured daily above the plant canopy and maintained by adjusting the distance of the LEDs to the plant canopy.

2.2. Plant Growth Measurement and Chlorophyll

If Measurements including stem height (SH), leaf number (LN), plant fresh weight (FW), plant dry weight (DW), moisture content (MC), and leaf area (LA). SH and LN were recorded on the plants in two replicates every weeks. The LA (cm^2) was measured with LI-3100 leaf area meter (LICOR) of every plant. For MC value, plant tissue samples were dried in a drying oven for 48 h at 65 ^oC before weighing and MC was calculated as in (1). To examine chlorophyll (chl) content, leaves were weighed to 0.1g FW and added into bottle containing 20 ml solution of 80% acetone. The bottle was kept in the dark room until the leaves changed their color to white. Optical density was measured with a UV 3101PC Scanning Spectrophotometer at 663 nm for chlorophyll a and at 645 nm for chlorophyll b. Concentrations of chl a and chl b was determined from the following equations:

$$MC = \frac{FW - DW}{FW} \times 100 \quad (1)$$

$$\text{Chl a (mg } g^{-1}) = [(12.7 \times A663) - (2.6 \times A645)] \times \text{ml acetone / mg leaf tissue} \quad (2)$$

$$\text{Chl b (mg } g^{-1}) = [(22.9 \times A645) - (4.68 \times A663)] \times \text{ml acetone / mg leaf tissue}$$

$$\text{Total Chl} = \text{Chl a} + \text{Chl b}$$

2.3. Gas Exchange Measurement

Measurements of net photosynthesis (μmol CO_2 $m^{-2}s^{-1}$), leaf stomatal conductance (mol H_2O $m^{-2}s^{-1}$), and transpiration rate (mol H_2O $m^{-2}s^{-1}$) of 2 replicate for each treatment were monitored using a Portable Photosynthesis System Li-6400XT (LICOR, USA). To assess the trade-off between CO_2 uptake and water loss, instantaneous water-use efficiency (WUE) was calculated as ratio between photosynthetic rate and transpiration rate (μmol CO_2/μmol H_2O). Diurnal measurements of gas exchange were taken from 0900 h to 1500 h on the fifth youngest fully expanded leaf of four plants in each replicate for T1 and T2. Statistical assessment was done on gas exchange parameters at between 1100 to 1200 h, which was presumed to be the diurnal period when photosynthetic rates would be maximal [9].

2.4. Statistical Analysis

Statistical analyses were conducted with statistical product and service solutions for Windows, version 16.0 (SPSS). All measurements were evaluated for significance by an analysis of variance (ANOVA) followed by the least significant difference (LSD) test at the $p < 0.05$ level [10].

3. Result

3.1. Plant Growth, Morphology and Pigment Contents

Results of the photomorphogenesis measurements of *Brassica chinensis* influenced by pulse light treatments shown in Table 1, and plants showed distinct growth responses for both treatments. Plants FW were shown the greatest weight when grown under T1 and followed by T2. Also for plant DW, plant that grown under T1 shown high value for T1 and plant FW and DW that grown under T1 and T2 was no

significant difference analyzed by SPSS. The values of LA were high in the order of plants grown under T1 and the parameters under both were not significantly different. In addition, normal appearances with stem height (SH) and leaf numbers (LN) of the *Brassica chinensis* plants were observed (Fig. 1). Moisture content higher was produced under T2, with the mean 30 samples of fresh plant weight of 18.54g and dry weight of 22.24 g, and moisture contents of 88.52%.

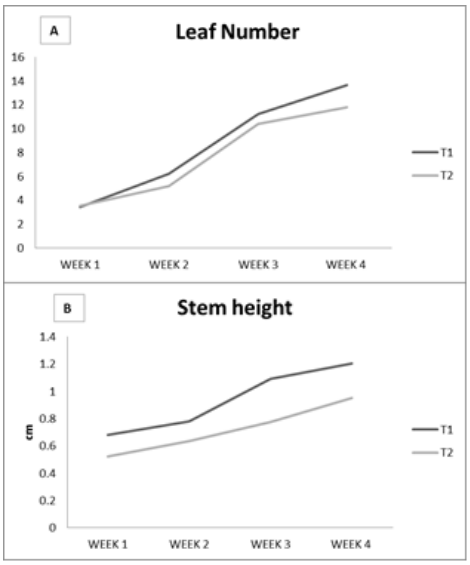

Fig. 1: Effects of RGB light-emitting diodes on (A) leaf number and (B) stem height of *Brassica chinensis* in weekly irradiation at T1 (100 µmol m^{-2} s^{-1}) and T2 (50 µmol m^{-2} s^{-1}).

Table 1: Influence of different intensities and photoperiod from a RGB (4:3:3) LED on LA, FW and DW, MC, chl a, chl b ant total chl of *Brassica chinensis* in 30 days.

T	Parameter						
	FW (g)	***DW (g)***	***MC (%)***	***LA (cm2)***	***Chl a (mg g^{-1})***	***Chl b (mg g^{-1})***	***Total Chl***
T1	26.65[a]	26.91[a]	87.41[a]	3.83[a]	2.62[a]	8.57[a]	3.48[a]
T2	18.54[a]	22.24[a]	88.52[a]	3.25[a]	1.81[b]	9.69[a]	2.78[a]

[a] and [b] shown significant different using LSD test at $P < 0.05$ probability level.

3.2. Chlorophyll Content

Chl a contents of *Brassica chinensis* leaves for both treatments showed significant difference while chl b and total chl showed no significant difference at $p > 0.05$ analayzed by SPSS. However, the higher chl a was under T1, chl b under T2 and for total chl under T1 (Table 1).

3.3. Gas Exchange

The diurnal mean leaf photosynthesis rates of *Brassica chinensis* under RGB (4:3:3) LED in pulse lighting treatment are shown in Fig. 2. Plants grown under T1 gave higher mean value in weekly at 1.203, 2.55, 5.37, 6.11 µmol CO_2 m^{-2} s^{-1}. The diurnal mean leaf transpiration rate of *Brassica chinensis* under RGB (4:3:3) LED in pulse light treatment is shown in Fig. 2. The data as shown indicate that T1 depicted higher transpiration rate when compared to counterparts within the time frame of the study. The diurnal mean leaf stomatal conductance of *Brassica chinensis* under different treatments are depicted in Fig 2. All plants T1 conditions gave higher than those in T2, with values at 0.24, 0.20, 0.09 and 0.11 mol H_2O m^{-2} s^{-1} by weekly. The diurnal mean leaf water use efficiency (WUE) of *Brassica chinensis* under various levels of intensity are shown in Fig. 2. The T2 plants recorded higher WUE values than T1 plants thus implying that intensity

influenced the WUE of plants substantially. The result from the comparisons of treatment means for 30 days showed that that there were significant ($p < 0.05$) differences among the treatments.

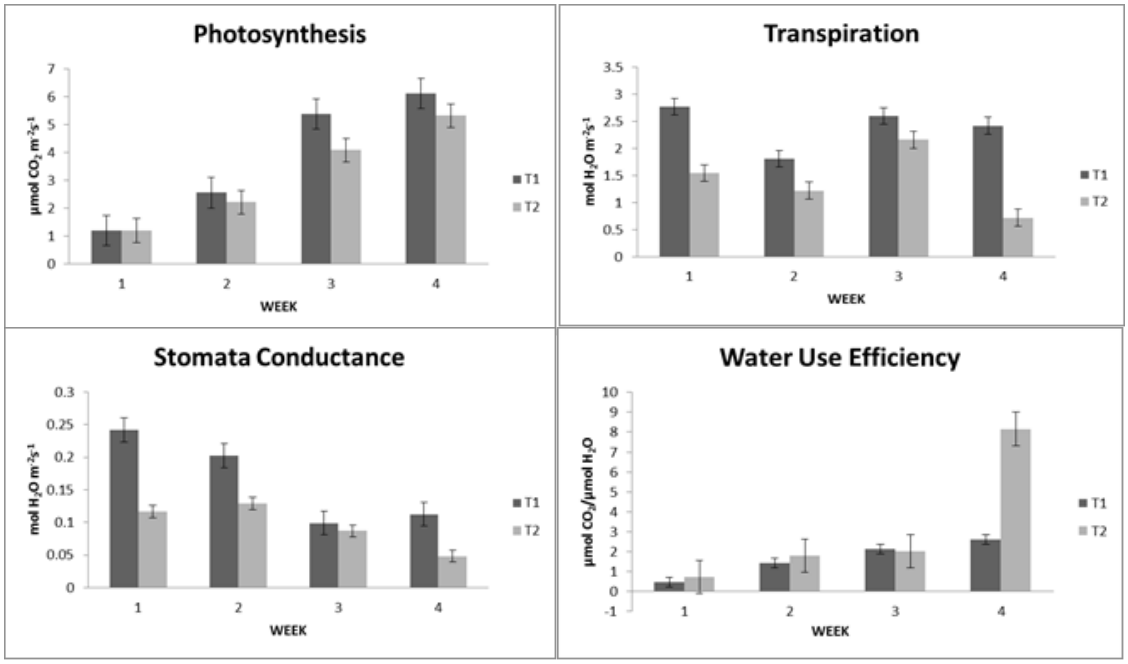

Fig. 2: Effects of RGB light-emitting diodes on *Brassica chinensis* leaf photosynthesis, transpiration, stomata conductance and water use efficiency (WUE) given weekly irradiation at T1 (100 µmol m^{-2} s^{-1}) and T2 (50 µmol m^{-2} s^{-1}).

4. Discussion

The combination of RGB with pulse lighting treatment promotes growth in *Brassica chinensis*. The PFD of intensity was at 100 µmol m^{-2} s^{-1} and also known as a T1and 50 µmol m^{-2} s^{-1} for T2.

The experiment ran for a total of 30 days. The treatment started on the 5th day, after the sowing. The average day temperature and relative humidity values in the research lab were 28.0±◦C and 45% respectively, during the experimental period. During the study, plant physical measurement was recorded once a week until end of the experimental *Brassica chinensis* growth under both treatments was also recorded. After one week transplants, all 100% plants survived for both treatments. After two weeks, 8 plants under T1 were dead while 3 plants were dead under T2. This indicates that plant can survive under 50 µmol m^{-2} s^{-1} since the PFD was low in 28^{o}C temperature condition. Three weeks after transplant, the number of plant dead increased under T1 which is 9 plants and 7 plants were dead under T2. At the final week, no plants were dead for both plants until the harvesting time.

For the T1, the mean values of FW, DW and MC were high due to low intensity compared to T2. This situation must be related to the environment. The temperature of environment at 28^{o}C was not suitable and high for *Brassica Chinensis* growth and development. The relative humidity at 45% was also very dry for plant development and morphology. Physical measurement for LN and SH *Brassica chinensis* increasing week by week but there a high value T2 compared to T1

Due to the environment condition, chl b was analyzed by SPSS and there area shown no significant impact but the value of Chl b was respectively high compared to chl b at T2. Chl b content was quantitatively high but does not influence qualitatively the action spectrum of chlorophyll formation. This could be attributed to the fact that the relative stimulatory effect of blue light is stronger for Chl b than for Chl a.

Plant gas exchange that contribute to leaf photosynthesis rate, transpiration rate and stomata conductance were higher under T1 compared to T2. This result showed that dry humidity and high temperature did not disturb the activity even though they were grown under 100 µmol m^{-2} s^{-1}.

5. Conclusion

RGB in different intensity showed significant impact to *Brassica chinensis* growth and development. However, environmental condition such as temperature and humidity will affect the plant growth but for leaf gas exchange, it was still active as usual for normal growth. For future work, RGB ratio will be changed and used as a light treatment under difference intensity with variable environmental parameters to study *Brassica chinensis* plant growth and photomorhogenesis.

6. Acknowlegment

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