



Growth and phenolic compounds of *Crepidiastrum denticulatum* under various blue light intensities with a fixed phytochrome photostationary state using far-red light

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Abstract

This study aimed to evaluate the effect of the red (R)-to-blue (B) ratio of light on the growth and phenolic compounds of *Crepidiastrum denticulatum* under a phytochrome photostationary state (PSS) for growth in a plant factory with artificial light (PFAL) using light-emitting diodes (LEDs). Three-week-old *C. denticulatum* seedlings were transplanted into a PFAL where the air temperature, relative humidity, CO₂ concentration, and light period were set at 20 °C, 60%, 1000 μmol·mol⁻¹, and 16 h, respectively. Three controls were used with different ratios of R to B light without supplemental FR light: 8:2, 7:3, and 6:4 (based on chip number; 130 μmol·m⁻²·s⁻¹ photosynthetic photon flux density). For the treatments, the same R to B light ratios as in the controls were used but supplemented with FR light, plus a treatment with only R light supplemented with FR, set to a PSS of 0.71. Growth characteristics and total phenolic and individual phenolic contents were measured after a 6-week treatment. When the R light ratio increased, shoot dry weight, leaf length, leaf width, and leaf area increased regardless of supplemental FR light, and the R8B2 with FR light treatment was the most effective, with significantly higher values (1.6–2 times) than the control. FR irradiation did not have any negative effect on total phenolic content, chlorogenic acid, caffeic acid, and chicoric acid per unit dry weight (g); thus, the R8B2 with FR treatment had significantly higher total phenolic and individual phenolic contents per shoot (43% and 52–62%, respectively). Thus, supplemental lighting with FR LEDs was found to be effective to enhance the growth and bioactive compounds of *C. denticulatum* in a PFAL installed with a R and B lighting system, and the effectiveness could be changed by the RB ratio, with 8:2 considered the proper ratio.

Keywords Individual phenolic content · Plant factory · Red/blue ratio · Total phenolic content

1 Introduction

Light, one of crucial environmental factors for plant growth and development, is utilized as an energy source in photosynthesis and perceived as signals by various photoreceptors such as phytochromes, cryptochromes, phototropins, and UVR8 (Fraser et al. 2016). Phytochromes (P) mainly perceive light in the form of red (R) and far-red (FR) and exist as two convertible forms—as P_r in the inactive form, and as P_{fr} in the active form. The phytochrome photostationary

state (PSS), the ratio of P_{fr} to total phytochromes (P_{total}), is changed by the light quality irradiated to plants. R light is effective for changing P_r to P_{fr} so that the PSS value increases, but FR light induces the opposite effect. Variations in the PSS influences plant physiology and morphology (Possart et al. 2014; Craig and Runkle 2016). There is a pronounced difference in the PSS between above (direct solar light) and below the plant canopy (shade). The PSS in a shaded place is lower than that above the plant canopy because most R light is absorbed by plants, and FR light is easily transmitted to below the plant canopy. Previous studies reported that light environments with low PSS induced shade avoidance responses (SAR) such as petiole elongation in *Arabidopsis thaliana* and leaf expansion in tomatoes (*Solanum lycopersicum*) (Djakovic-Petrovic et al. 2007; Chitwood et al. 2015). Decreased PSS conditions due to enriched FR light activate phytochrome interaction factors (PIF) 4 and 5, which are key transcriptional factors in the

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main pathway for photomorphogenesis and control auxin biosynthesis and promote stem elongation as signaling messengers (Hornitschek et al. 2012).

FR irradiation increases the electron transport efficiency in the light-dependent reaction of photosynthesis because FR light is absorbed by light-harvesting complex I (LHC I) in photosystem I (PS I) (Croce and Van Amerongen 2013). Under the visible light range without FR light, R light is absorbed by light-harvesting complex II (LHC II) in the light-dependent reaction and thereby photosystem II (PS II) is activated. Some of LHC II moves to PS I when the light energy in PS II is saturated to control the unbalanced electron transport between PS II and PS I. Although mobile LHC II plays a role in controlling unbalanced electron transport under the visible light range, supplemental FR irradiation with visible light enhances the photosynthetic rate due to more stable and balanced electron transport between PS I and PS II (Rochaix 2011). An enhanced photosynthetic rate of plants subjected to visible light with supplemental FR light is called the Emerson enhancement effect, and eventually, supplemental FR light can improve plant growth.

Plant factories with artificial light (PFAL) enable the control of environmental factors for the growth and development of plants, including light conditions. With light-emitting diodes (LEDs), which have a narrow spectrum, it is possible to make various specific light qualities by the combination of different LEDs. In particular, combined R and B LEDs have been used and studied in a PFAL due to the high efficiency of this combination in photosynthesis (Sabzalian et al. 2014; Piovene et al. 2015). Recently, white (W) and R LEDs are being used as commercial light sources in conjunction with green (G) LEDs (Bian et al. 2016; Park et al. 2016; Son et al. 2018). Few studies using FR LEDs have been reported and FR light has not been commercially used up until now in a PFAL. In our previous study, when FR LEDs were supplemented under R and B combination light sources in a PFAL, we confirmed that a low PSS (0.65–0.70) had positive effects on the growth and bioactive compounds of *Crepidiastrum denticulatum* (Bae et al. 2017). However, there is no information about the effects of the R to B ratio on growth and levels of bioactive compounds in *C. denticulatum* under a low PSS using FR light.

Therefore, this study aimed to evaluate the effects of the R to B light ratio on the growth and phenolic compounds in *C. denticulatum* under a low PSS.

2 Materials and methods

2.1 Plant materials and growth conditions

The mixture of horticultural growing medium (Dongbu Hannong, Seoul, Korea) sifted by a 1.4-mm sieve (500 mL)

and seeds of *C. denticulatum* (15 mL) was blended with distilled water (100 mL) and then incubated in the dark for 48 h in an incubator (air temperature 20 °C; relative humidity 60%). Six 105-cell plug trays were filled with the growing medium mixed with seeds and cultured for 3 weeks in a PFAL (air temperature 20 °C; CO₂ 1,000 μmol·mol⁻¹; white:red LEDs = 9:1, photosynthetic photon flux density (PPFD) 150 ± 5 μmol·m⁻²·s⁻¹; light period 16 h; relative humidity 60%). One week before transplanting, seedlings were thinned out, leaving one seedling per cell. Distilled water (1 L) was supplied to the mixture through subirrigation systems once every 2 or 3 days. After 3 weeks of culture, seedlings were transplanted to pots [6 × 6 × 6 cm (L × W × H)] and filled with a mixture of peat moss and perlite (v/v = 7:3) and two wicks [1.5 × 0.14 cm (W × T), non-woven fabrics, Daegyeongtongsang Co., Seoul, Korea] were inserted into a pot bottom. Six pots were placed on a tray [32 × 22 × 6.5 cm (L × W × H)] and freshly prepared nutrient solution for *C. denticulatum* (Park et al. 2016) was supplied to the pot via two wicks. The wick length from the pot bottom to the surface of the nutrient solution was 2 cm, which was maintained by checking every day. Growth environments in the PFAL, except for light, were the same as in the germination conditions.

2.2 Light treatments

The light treatment was initiated after transplanting. Three controls were used; the ratio of red (R; Itswell, Incheon, Korea; peak 660 nm) to blue (B; Itswell, Incheon, Korea; peak 440 nm) light was 6:4, 7:3, and 8:2 (based on chip number; R6B4, R7B3, and R8B2) in the three controls. Using a spectroradiometer (Jaz System, Ocean Optics Inc., Dunedin, FL, USA), the quantum per unit wavelength (2 nm) were measured and the average total PPFD of each light in the control group was set to 130 ± 5 μmol·m⁻²·s⁻¹. For the treatments, the same R to B light ratios as in the controls were used but supplemented with FR light (Itswell, Incheon, Korea; peak 735 nm), plus a treatment with only R light supplemented with FR light. The PSS was set to 0.71 in all treatments, which was the best condition for the growth of *C. denticulatum* in our previous study (Bae et al. 2017). The PSS value was calculated by the equation described in Sager et al. (1988).

2.3 Growth characteristics

To compare the growth characteristics of *C. denticulatum* cultivated for 6 weeks after transplanting, the dry weight of shoots and roots, shoot-to-root ratio (S/R ratio), number of leaves, leaf area, leaf length, leaf width, and SPAD value were measured. The dry weight of shoots and roots were measured by drying shoot and root samples for 72 h in a

freeze drier (FD8512, Ilshin Lab. Co. Ltd., Dongducheon, Korea) and an oven (70 °C, VS-1202D3, Vision Scientific, Daejeon, Korea), respectively. Dried samples were measured with a digital scale (Si-234, Denver Instrument, Denver, CO, USA). A leaf bigger than 1 cm in length was regarded as a leaf and the number of leaves was counted. Leaf area was measured using a leaf area meter (LI-3100C, Li-Cor, Lincoln, NE, USA). Leaf length and leaf width of the largest leaf were measured and petiole length was included in leaf length. The SPAD value was measured to indirectly quantify the chlorophyll content of fully-unfolded leaves by a portable chlorophyll meter (SPAD-502, Konica Minolta, Tokyo, Japan).

2.4 Photosynthetic rate

A portable photosynthesis system (LI-6400, Li-Cor, Lincoln, NE, USA) was used to measure the photosynthetic rate of the third leaf from the top after 5 weeks of treatment. The CO₂ concentration, air flow rate, and the block temperature were set at 1,000 μmol·mol⁻¹, 400 μmol·s⁻¹, and 20 °C, respectively. A clear chamber bottom (2×3 cm) was attached to the portable photosynthesis system so that the light condition used for measurement of the photosynthetic rate was the same as each light treatment condition. The photosynthetic rate was measured within 2 h of the middle of the light period (7–9 h of lighting).

2.5 Phenolic compounds

Freeze-dried samples of shoots were powdered for 2 min at 15,000 rpm using a mill (Tube Mill control, IKA, Wilmington, NC, USA) to analyze the contents of total phenolics, chlorogenic acid, caffeic acid, and chicoric acid after 6 weeks of treatment.

A modified Folin-Ciocalteu method (Ainsworth and Gillespie 2007) was applied to analyze the total phenolic content. A freeze-dried powdered sample (about 40 mg) mixed with 80% (v/v) acetone (4 mL) was incubated in an ultrasonicator (SK5210HP, Hangzhou Nade Scientific Instrument, Zhejiang, China) for 15 min. The extracted solution was maintained at 4 °C in a refrigerator in the dark for 12 h. The solution was centrifuged at 3,000×g for 2 min using a microcentrifuge (1730R, Gyrozen Inc., Daejeon, Korea) and the supernatant (0.1 mL) was diluted with 80% acetone (0.9 mL) in a 2-mL micro-tube. The remaining protocol was followed as described in Son and Oh (2013). The total phenolic content was represented by milligram of gallic acid equivalent per gram dry weight (mg GAE·g DW⁻¹) or per shoot (mg GAE·shoot⁻¹).

A high-performance liquid chromatography system (YL9100, Young Lin Instrument Co., Ltd, Anyang, Korea) was used to analyze the content of individual phenolic

compounds, such as chlorogenic acid, caffeic acid, and chicoric acid, in *C. denticulatum*. A powder sample of approximately 100 mg was blended with 70% acetonitrile (Honeywell Burdick & Jackson, Muskegon, MI, USA) and 0.5% HCl (Samchun Pure Chemical Co., Ltd., Pyeongtaek, Korea). The solution was hydrolyzed at 80 °C in a water bath (MSB-2011D, Mono-Tech Engineering, Siheung Korea) for 2 h and extracted in the ultrasonicator for 90 min. The solution was centrifuged at 3000×g for 20 min using a microcentrifuge and the supernatant was filtered through a 0.22-μm syringe filter (Noble Bio Co., Ltd, Hwaseong, Korea). The final solution (10 μL) was flowed with solvent A (Acetonitrile, 100%) and B (Acetic acid in water, 0.5%), used for the mobile phase at 0.8 mL·min⁻¹ through an ACE-5-AQ column (4.6×250 mm, Advanced Chromatography Technologies, Reading, UK) at 30 °C. The gradient of solvent A was 0–10 (0–10 min), 10–20 (10–30 min), 20–30 (30–40 min), 30–40 (40–50 min), 40–80 (50–60 min), 80–0 (60–61 min), and 0% (61–70 min). The optical density of individual phenolic compounds was measured at 320 nm and chlorogenic acid, caffeic acid, and chicoric acid (all from Sigma-Aldrich, St. Louis, MO, USA) were used as standards.

2.6 Statistical analysis

The experiment was performed by a completely randomized experimental design with 10 replications. Statistical analysis was performed by SAS (SAS 9.2, SAS Institute, Cary, NC, USA). After an analysis of variance (ANOVA), significant differences among means of treatments was determined by Duncan's multiple range test at the $p < 0.01$ level.

3 Results

3.1 Growth characteristics and photosynthetic rate

In the case of the shoot dry weight, the control group without FR light did not exhibit any significant difference among the different R to B ratios. Among the FR treatments, the R8B2 + FR treatment had the significantly highest shoot dry weight and this was also the highest value among all the treatments, including the control group (Fig. 1a). Comparing between the control group and FR treatments in the same R to B ratio, only the R8B2 + FR treatment showed a significantly (1.6 times) higher shoot dry weight. In the FR treatment group, there was an increasing trend as the R light ratio increased. The R + FR treatment without B light also did not show any significant effect on shoot dry weight. There were no significant differences among the R to B light ratios in the dry weight of roots within the control group or FR treatment group. However, the control group had 2.1–3.3 times higher values than the FR treatment group (Fig. 1b).

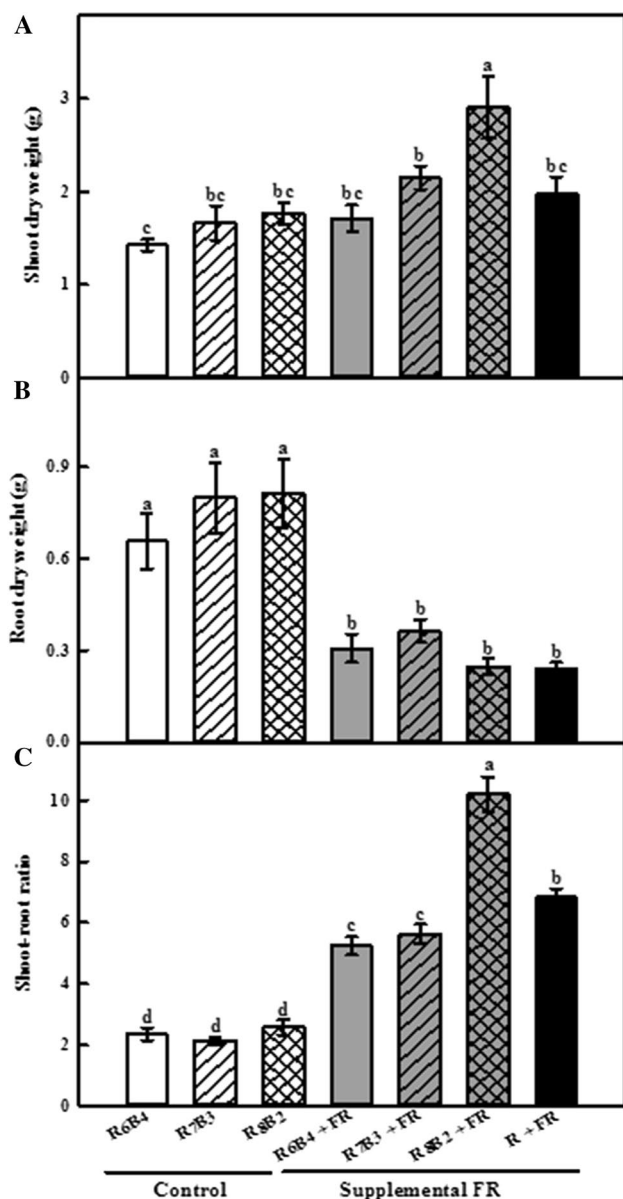


Fig. 1 Dry weight of shoots (a) and roots (b) and the shoot/root ratio (c) of *C. denticulatum* subjected to the control (R:B ratio 8:2, 7:3, and 6:4) and supplemental FR conditions at 6 weeks of treatment. Different letters indicate significant differences at $p < 0.01$ ($n = 10$)

In the control group, the S/R ratio, which expresses shoot development relative to roots, did not differ significantly as the R to B light ratio changed, whereas in the FR treatment group, the S/R ratio was at least 2.2 times higher than in the control group. The value of the S/R ratio in the R8B2 + FR treatment was 10.2, which was the highest (Fig. 1c).

In the control group, the number of leaves in the R6B4 control had the lowest value; however, there were not significant differences with changes in the R to B ratio (Fig. 2a). In the FR treatment group, the R6B4 + FR treatment had the lowest and the R8B2 + FR treatment had the highest value

for number of leaves, but these results were not significantly different from those of the R7B3 + FR and R + FR treatments. Comparing within the same R to B light ratio, the FR treatment group did not have a significant effect on the number of leaves compared to the control group. Leaf area in the control group was not significantly different among the different R to B light ratios, whereas in the FR treatment group, the R8B2 + FR treatment had the highest value and no significant differences were found compared with the other FR treatments (Fig. 2b). The leaf area of all FR treatments increased 1.6–2 times compared to the control group within the same respective R to B light ratio. Leaf length was significantly higher in all FR treatments than in the control group (Fig. 2c). Leaf length of the R6B4 control had a significantly lower value compared to the R7B3 and R8B2 controls, and the B8B2 + FR treatment had the longest leaf length in FR treatment group. When comparing the control group and FR treatment group at the same R to B light ratios, the leaves in the FR treatments were significantly (1.6–1.7 times) more elongated than in the controls. Leaf width did not have any significant differences in the control group, and the R8B2 + FR treatment had the highest value among the FR treatments (Fig. 2d). Regardless of the ratio of R to B light, supplemental FR light lead to a significantly increased (1.3–1.4 times) leaf width compared to the control. By comparing the biggest leaves in each light source, the results of leaf length and leaf width mentioned above could easily be confirmed (Fig. 3).

The SPAD value of leaves under all FR treatments was significantly decreased compared to the control group (Fig. 4a). In the control group, the R to B light ratio had no effect on the SPAD value, and in the FR treatment group, the R7B3 + FR treatment had the highest SPAD values. The photosynthetic rate in the R8B2 + FR treatment was the highest among all the treatments including the control group (Fig. 4b). The photosynthetic rate in the R8B2 + FR treatment was 23% higher than in the R8B2 control. In the control group, the photosynthetic rate did not significantly differ with changes in the R to B light ratio; however, in the presence of FR light, the photosynthetic rate significantly improved as the R light ratio increased. The R + FR treatment did not show significant differences compared with the other two FR light treatments except for the R8B2 + FR treatment (Table 1).

3.2 Phenolic compounds

There was no significant difference in the total phenolic content per unit dry weight within the control group or the FR treatment group with changes in the R to B light ratio. When comparing the control group and the FR treatment group at each R to B light ratio, the total phenolic content per unit dry weight in the R8B2 + FR treatment

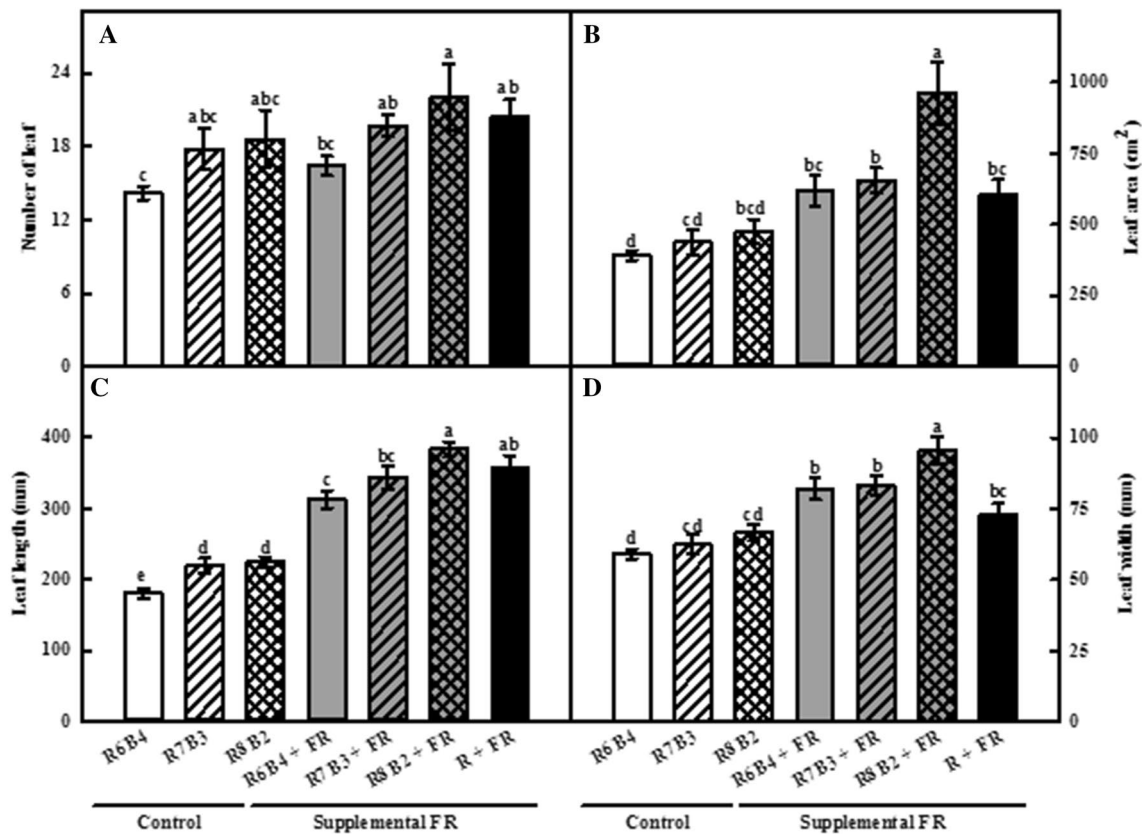


Fig. 2 Number of leaves (a), leaf area (b), leaf length (c), and leaf width (d) of *C. denticulatum* subjected to control (R:B ratio 8:2, 7:3, and 6:4) and supplemental FR conditions at 6 weeks of treatment. Different letters indicate significant differences at $p < 0.01$ ($n = 10$)

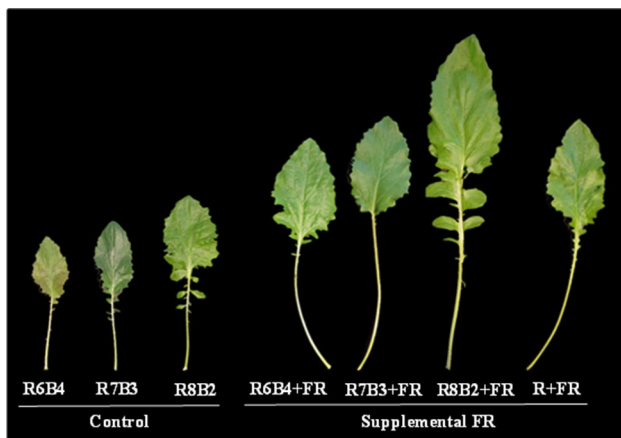


Fig. 3 Leaves of *C. denticulatum* subjected to control (R:B ratio 8:2, 7:3, and 6:4) and supplemental FR conditions at 6 weeks of treatment

significantly decreased (by 15%), but not in the R7B3 + FR and R6B4 + FR treatments (Table 2). In addition, no significant difference was observed in the R + FR treatment compared with the other FR treatments. The total phenolic content per shoot dry weight in the control group had an

increasing tendency as the R light ratio increased, but only the R8B2 + FR treatment had a significantly higher value, which was 43% higher than that of the R8B2 control.

In the control group, the content of individual phenolic compounds per unit dry weight did not differ significantly except for decreased caffeic acid content in the R6B4 control. In the FR treatment group, all individual phenolic compounds decreased significantly under the R6B4 + FR treatment compared with the other FR treatments. When comparing the control group and the FR treatment group at the same R to B light ratio, no significant differences were observed in the content of individual phenolic compounds per unit dry weight except for decreased chicoric acid content under the R6B4 + FR treatment. Regardless of FR light, the content of all individual phenolic compounds per shoot dry weight showed the lowest value in the R6B4 control. The R to B light ratio did not affect the contents in the control group. As the R light ratio increased, the chicoric acid content significantly increased, but the contents of chlorogenic acid and caffeic acid did not change except for in the R6B4 + FR. The content of all individual phenolic compounds per shoot dry weight had the highest value in the

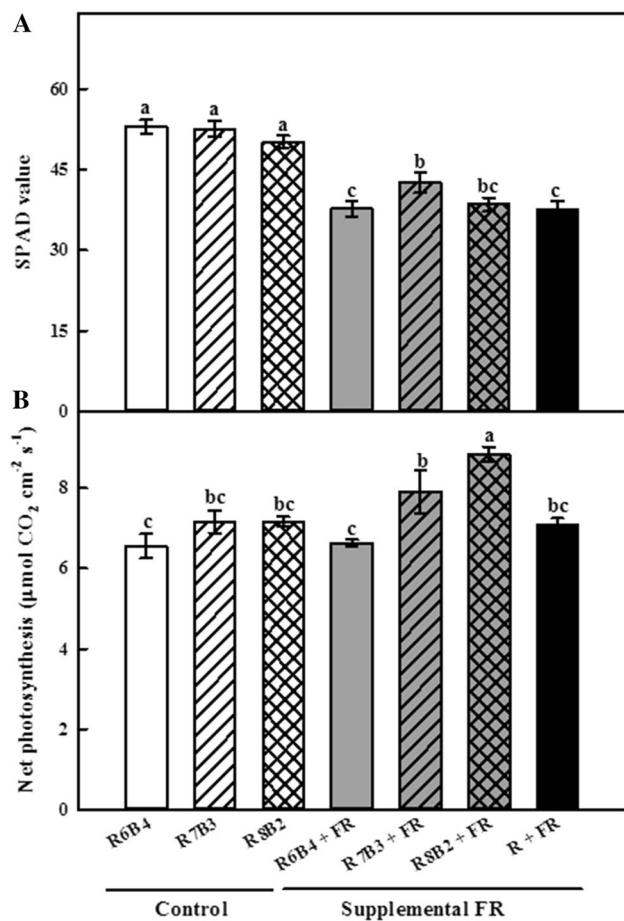


Fig. 4 SPAD value (a) and photosynthetic rate (b) of *C. denticulatum* subjected to control (R:B ratio 8:2, 7:3, and 6:4) and supplemental FR conditions at 6 and 5 weeks of treatment, respectively. Different letters indicate significant differences at $p < 0.01$ ($n = 10$)

R8B2 + FR treatment, which was a 52–62% increase relative to the R8B2 control.

4 Discussion

The R to B light ratio in the control group did not significantly affect the shoot growth of *C. denticulatum*. This result was not consistent with previous results obtained from the studies related to R to B light ratios. Wang et al. (2016) reported that leaf area and shoot dry weight of lettuce (*Lactuca sativa*) grown under combined R and B lighting sources (R:B = 1:1, 4:1, 8:1, and 12:1) for 30 days significantly decreased as the R to B light ratio increased from 1 to 8. Moreover, when cucumber (*Cucumis sativus*) seedlings were cultivated under various R to B light ratios (R:B = 2.5:7.5, 5:5, 7:3, and 9:1) for 17 days, leaf area and dry weight decreased as the B light ratio decreased (Hernández and Kubota 2016). A decreased B light ratio led to increased total leaf area in lettuce and cucumber due to leaf enlargement resulting in significant increases in shoot dry weight; but in the case of *C. denticulatum*, the change of the R to B light ratio without FR light did not show any clear effect on leaf area and shoot dry weight. In contrast, when supplemented with FR light, the effect of the R to B light ratio was more pronounced. In previous studies, supplemental FR light increased leaf area by cell expansion and supplemental FR light induced the SAR as the R to FR light ratio decreased (Lee et al. 2015, 2016). In addition, a low PSS value (~0.7) by supplemental FR light was effective for improving shoot growth of *C. denticulatum* (Bae et al. 2017). In this study, the leaf area in all FR treatments significantly increased, which was consistent with previous results. However, a typical SAR was shown in the decreased B light ratio, although

Table 1 Summary of the spectral qualities tested in this study

Parameter	Control group			Supplemental FR group ^z			
	R6B4	R7B3	R8B2	R6B4+FR	R7B3+FR	R8B2+FR	R+FR
Photon flux density ($\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$)							
Blue (400–500 nm)	61.04 (47.1%) ^y	48.28 (36.5%)	30.68 (23.6%)	61.9 (47.1%)	48.54 (36.4%)	31.92 (24%)	–
Red (600–700 nm)	68.69 (52.9%)	84.07 (63.5%)	99.19 (76.4%)	69.51 (52.9%)	84.84 (63.6%)	100.88 (76%)	131.03 (100%)
Far-red (700–800 nm)	–	–	–	55.3	77.95	86.2	124.16
PPFD (400–700 nm)	129.73 (100%)	132.35 (100%)	129.87 (100%)	131.41 (100%)	133.38 (100%)	132.8 (100%)	131.03 (100%)
Ratios							
Red/far-red	–	–	–	1.26	1.09	1.17	1.05
PSS (P_{fr}/P_{total})	0.87	0.876	0.883	0.71	0.71	0.71	0.71

Spectra were recorded and averaged at nine locations at the height of the plant canopy with a spectroradiometer

^zSame R/B ratios and mono R LED with supplemental FR LEDs

^yThe percentage over photosynthetic photon flux density (PPFD)

Table 2 Contents of total phenolics, chlorogenic acid, caffeic acid, and chicoric acid of *C. denticulatum* subjected to control (R:B ratio 8:2, 7:3, and 6:4) and supplemental FR conditions at 6 weeks of treatment

Treatment	Total phenolic content		Chlorogenic acid content		Caffeic acid content		Chicoric acid content	
	(mg GAE g ⁻¹ DW)	(mg GAE shoot ⁻¹)	(mg g ⁻¹ DW)	(mg shoot ⁻¹)	(mg g ⁻¹ DW)	(mg shoot ⁻¹)	(mg g ⁻¹ DW)	(mg shoot ⁻¹)
Control								
R6B4 ^z	18.0 abc ^y	25.8 b	5.1 bc	7.3 c	0.3 cd	0.4 d	4.3 ab	6.2 c
R7B3	19.6 ab	32.2 b	6.8 b	10.8 bc	0.4 ab	0.6 cd	4.3 ab	7.0 c
R8B2	20.4 a	35.4 b	6.9 b	11.5 bc	0.4 ab	0.6 bcd	4.4 ab	7.7 bc
Supplemental FR								
R6B4 + FR ^x	16.4 c	29.3 b	3.7 c	6.5 c	0.3 d	0.4 d	3.4 c	5.9 c
R7B3 + FR	17.3 bc	36.7 b	7.1 ab	15.9 ab	0.4 a	0.9 ab	4.4 a	9.5 b
R8B2 + FR	17.3 bc	50.8 a	6.1 b	18.7 a	0.3 bc	1.0 a	4.0 b	11.7 a
R + FR	16.9 bc	32.9 b	9.4 a	18.2 a	0.4 ab	0.8 abc	4.1 ab	8.2 bc

^zRed and blue LED number of chips ratio

^yLetters indicate a significant difference at $p < 0.01$

^xFar-red LED supplementation based on the same control group light sources and mono red LEDs

the PSS values of all FR treatments were set as 0.71. This implies the importance of the R to B light ratio in spite of the control of photoconversion of phytochromes. *PIF 4* and *PIF 5*, genes related to the promotion of hypocotyl elongation, were deactivated under a high R to FR light ratio, whereas low R to FR light ratios induced the SAR, such as hypocotyl elongation, due to the activation of *PIF 4* and *PIF 5* by inactivated phytochrome B (Koini et al. 2009). *PIF 4* and *PIF 5* also are activated under low B light ratios. Cryptochromes deactivated *PIF 4* and *5* under high B light ratios, while they promoted the activation of *PIF 4* and *5* under the B light ratio resulting in the SAR (Pedmale et al. 2016). Therefore, it was thought that the SAR of *C. denticulatum* in FR light treatments was more pronounced than in the control group as the B light ratio decreases.

Increased shoot growth of *C. denticulatum*, according to supplemental FR light and a decreased B light ratio, is attributed to signal stimulus of phytochromes that induces morphological changes as well as an improved photosynthetic rate. In previous studies related to the R to B light ratio, enhanced biomass accumulation was caused by increased R light ratios that resulted in relatively higher photosynthetic efficiency than the B light ratio. According to relative quantum efficiency (RQE) curves for light spectrum described in McCree (1972), R light was 25–30% more efficient than B light. However, no significant difference in the photosynthetic rate of *C. denticulatum* was observed when the R light ratio increased in the control group without FR light. In contrast, the photosynthetic rate per unit area (cm²) under the supplemental FR irradiation treatments significantly increased along with leaf enlargement and the R to B light ratio affected shoot dry weight. In the light-dependent reaction of photosynthesis, the reaction center of PS II is

activated by LHC II, which consists of chlorophyll a and b mainly absorbing R and B light. The reaction center of PS I, however, is activated by LHC I, which has relatively more chlorophyll d (mainly absorbing FR light), than LHC II (Hu et al. 1998). Therefore, if the visible light, such as R and B light, were irradiated to plants, the activity of PS I would decrease compared with that of PS II. To solve this imbalanced state, some of the mobile LHC II moves to PS I (Rochaix 2011). Under the visible light conditions supplemented with FR light, the transfer of mobile LHC II to PS I is reduced due to the activation of PS I by FR light absorption in LHC I. Therefore, the energy efficiency of PS II can also be raised. In a state of activated PS I by FR irradiation, the R light ratio that induced the maximum activation of PS II seemed to be 80% in *C. denticulatum*. In the case of the R + FR treatment, which has the highest R light ratio with FR supplemental light, the photosynthetic rate was lower than in the R8B2 + FR treatment because the proper combination of R and B light can increase the efficiency of the total photosynthetic rate of plants compared with mono R irradiation (Goins et al. 1997; Lee et al. 2007).

In the studies of Son and Oh (2013, 2015), as the B light ratio increased from the R9B1 to R6B4 treatments, the dry weight of shoots and leaf area of lettuce were reduced, but total phenolic content and antioxidant capacity per unit fresh weight (g) were improved. Moreover, Johkan et al. (2010) reported that an increased B light ratio improved the contents of bioactive compounds in lettuce. In contrast, the content of total phenolics and individual phenolic compounds, such as chlorogenic acid, caffeic acid, and chicoric acid, per unit dry weight in *C. denticulatum* decreased or did not have significant differences as the B light ratio increased regardless of FR treatments. This difference can be explained by

different light conditions of the original habitat of a plant species.

In summary, supplemental FR light with R and B LED lights improved the growth of *C. denticulatum* based on the increase of leaf area and photosynthetic rate, and subsequently the content of bioactive compounds per plant also showed a positive effect. In addition, the manipulation of the R to B light ratio under the light conditions with proper PSS (0.71) using FR LED lights influenced the growth of *C. denticulatum*, and the highest growth was observed under the R8B2 + FR treatment. These results suggest that FR light is effective for improving the yield and quality of plants produced in PFALs and the application of FR light should be considered with the ratio of existing R and B light sources.

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