

Uncovering LED light effects on plant growth: new angles and perspectives – LED light for improving plant growth, nutrition and energy-use efficiency

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Abstract

Light supplementation can increase crop yield in greenhouses by promoting photosynthesis and plant growth. However, the high energy costs associated with light supplementation are a predominant factor that limits development and profit improvement of controlled environment agriculture. Light-emitting diodes (LEDs) are a promising technology that has tremendous potential to improve irradiance efficiency and to replace traditionally used horticultural lighting. Compared with traditional light sources (e.g., high-pressure sodium lamps and metal halide lamps) used in crop production, LEDs have distinct advantages, such as their small size, long lifetime and high photoelectric conversion efficiency. Most importantly, as a monochromatic light source, the spectrum of LEDs can be adjusted based on plant growth requirements. This project aimed to investigate energy-use efficiency, vegetable nutrition and photosynthesis improvement of light supplementation in a protected horticulture system. In the initial phase, the effects of LED light on plant growth and light-use efficiency for pak choi and photosynthetic performance were investigated. The results showed that the highest fresh and dry weight and leaf area were observed under red and blue LED light, with the blue light percentage at 23%. Compared with fluorescent lamps (FL) with photosynthetic photon flux density (PPFD) at 220 $\mu\text{mol m}^{-2} \text{s}^{-1}$, the light-use efficiency increased by 55, 114 and 115% for mixed red and blue LEDs with PPFD at 100, 150 and 220 $\mu\text{mol m}^{-2} \text{s}^{-1}$, respectively. Monochromatic red- and blue-light LEDs resulted in significant decreases in P_n of tomato plants, but the stomatal conductance (G_s) for monochromatic blue LEDs was higher than that for FL. The effect of light spectrum composition on lettuce nutrition quality was also studied. Continuous light with combined red, green and blue LEDs exhibited a remarkable decrease in nitrate. Moreover, continuous LED light for 24 h significantly increased phenolic compound content and free-radical scavenging capacity in lettuce leaf.

Keywords: LEDs, light quality, photosynthesis, energy-use efficiency, nitrate content, phenolic compounds

INTRODUCTION

Greenhouses have been introduced into commercial crop production for several decades. Currently, small industries and individuals have been involved in urban farming with success in producing fresh food, in a sustainable way, able to deliver to the final consumer in an instant, without transportation costs or storage needs. Also, people that encounter these kinds of businesses and taste the products tend to prefer them, because they are healthier, fresher and last much longer than the imported equivalents. Light is one of the most important factors in plant growth development. Light is not only the energy for driving photosynthesis, but an essential signal to mediate downstream gene expression of substance metabolism for the plant to acclimate to environmental fluctuation (Chen et al.,

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2004). In the commercial greenhouse, light supplementation using artificial light can significantly increase crop yield and nutrition quality, especially in low-light-intensity seasons like winter and late autumn (Lu et al., 2012; Yorio et al., 2001). With the development of urban agriculture, artificial light has become the most important way to control the light conditions. For a long time, people were using fluorescent lamps, filament lamps and high-pressure sodium lamps (HPL), and much research was carried out to test their effects (Tibbitts et al., 1983). However, these kinds of light tend to consume large amounts of electrical energy and release a lot of heat (which will also increase the cooling system cost), and their spectra are not very suitable for plants, which leads to excessive waste of energy (Randall and Lopez, 2014). The most important element in controlling artificial farming costs is supplying light for photosynthesis and growth by light sources with high photoelectric efficiency. Light-emitting diodes (LEDs) have been proposed as alternative light sources in controlled agricultural environments since, compared with traditional horticulture light sources (e.g., HPL), LEDs have drastic advantages, such as superior lifetime, reduced size, cooler emitting temperature, and reduced energy consumption (Massa et al., 2008). An exciting potential of using LED lighting is the development of species-specific light recipes comprising the optimum proportion of specific narrow-band wavelength light that can optimize plant growth, development and other desirable traits (e.g., increase phytochemical content) (Bian et al., 2015, 2016), whilst significantly reducing the energy input compared with traditionally used horticulture light sources. Recently, the effect of LEDs on plant growth and development has aroused increasing interest. However, the results of related studies are sometimes different, and even contradictory (Avercheva et al., 2009; Bian et al., 2016; Hogewoning et al., 2010; Urbonavičiūtė et al., 2007). A hypothesis has been raised that the different application strategies might arouse different responses of plants to LED light treatments. We conducted experiments using different light spectral compositions and application strategies of LEDs to further reveal the disadvantages of LEDs in energy saving and plant nutrition quality improvement.

MATERIALS AND METHODS

Plant material and growth conditions

1. Experiment I.

Pak choi (*Brassica rapa* 'Bonsai', Chinensis group) seeds were sown in commercial substrate and germinated in an environment-controlled growth chambers. After germination, seedlings were randomly treated with red (650-670 nm; R) and blue (455-475 nm; B) LEDs (Green power research module, Philips) with four different R/B ratios for 14 days. The spectral compositions of light treatments are summarized in Table 1. The photosynthetic photon flux density (PPFD) of all light treatments for pak choi was maintained at $130 \mu\text{mol m}^{-2} \text{s}^{-1}$ through adjusting the distance between the light sources and plants. In the next step, pak choi seedlings were grown under mixed red and blue LED light, but with high ($220 \mu\text{mol m}^{-2} \text{s}^{-1}$), medium ($150 \mu\text{mol m}^{-2} \text{s}^{-1}$) and low ($100 \mu\text{mol m}^{-2} \text{s}^{-1}$) light intensities to investigate light-use efficiency. Pak choi grow under fluorescent lamps (FL; Unigro, 12/6N) with a PPFD at $220 \mu\text{mol m}^{-2} \text{s}^{-1}$ were used as a control. Each treatment light had triple replications (four or five plants per replication). In the growth chamber, photoperiod, day/night temperature, relative humidity and CO_2 level were set at 16 h, $25 \pm 1/20 \pm 1^\circ\text{C}$, $65 \pm 5\%$ and 400 ± 20 ppm, respectively.

2. Experiment II.

Tomato (*Solanum lycopersicum*) seeds were sown in commercial substrate and grown under FL (Unigro, 12/6N) with a PPFD at $150 \mu\text{mol m}^{-2} \text{s}^{-1}$. Every other day, tap water was added from the bottom of the pots to keep the substrate wet. When plants had their second true leaves, they were randomly treated with monochromatic red (650-670 nm, R) LEDs, monochromatic blue (455-475nm; B) LEDs, or mixed red and blue LEDs (R/B=1). Plants

grown under FL were used as a control. Every other day, light intensity was monitored with a spectrometer (Skye Instruments Spectra Sense, RS 232). Light intensity for all the light treatments was kept at $200 \mu\text{mol m}^{-2} \text{s}^{-1}$ by adjusting the distance between the light sources and the top of the plants. Other environment factors were the same as in experiment I.

Table 1. Light spectral composition details.

Treatment	Red light (%)	Blue light (%)
0%	100	0
23%	77	23
27%	73	27
30%	70	30
62%	62	38
100%	0	100

3. Experiment III.

Lettuce (*Lactuca sativa* L.) was seeded in plastic seedling trays filled with peat:vermiculite (3:1, v/v) and grown in a controlled growth chamber. Fluorescent lamps (T5, PHILIPS) were used as light sources for seedling cultivation and light intensity was maintained at $100 \mu\text{mol m}^{-2} \text{s}^{-1}$. The other environmental factors were set as in experiment I. When seedlings had their second true leaves, they were transplanted into a hydroponic growth system and grown under white (400-700 nm; W), LED light and combined red (peak at 660 nm; R) and blue (455-475 nm; B) LED light with/without green (515-545 nm; G). The light intensity and photoperiod were $200 \mu\text{mol m}^{-2} \text{s}^{-1}$ and 12 h, respectively and other environmental factors were as described in experiment I. Before harvesting, lettuce plants were conducted continuous light treatment for 24 h. The light treatment details are summarized in Table 2. These LED light sources (VQ-G200; Vanq Technology, Shenzhen, China) were maintained at 20 cm above the canopies of the plants. The PPFD of the LED light sources was monitored daily by a quantum sensor (LI-190SA; LI-COR, Lincoln, NE, USA).

Table 2. Light spectral details for continuous lighting.

Treatment	Before treatment		Continuous light treatment	
	Light source	Spectral ratio (red:blue:green)	Light source	Spectral ratio
W	White LEDs	0.4:1:1	White LEDs	0.4:1:1
RB	Red and blue LEDs	4:1	Red and blue LEDs	4:1
RBG	Red and blue LEDs	4:1	Red and blue LEDs plus green LEDs	4:1:1
rb	Mixed red, blue and green LEDs	1:1:1	Red and blue LEDs	1:1

Plant growth determination

Pak choi seedlings were sampled after treatment with different light spectra for 7 days. Plants were cut from the hypocotyl. The shoots were weighed and then dried in an oven at 72°C to calculate shoot dry weight. Leaf areas were monitored using Li-3001C (LI-COR, Lincoln, NE, USA). To investigate energy-use efficiency, all plants were harvested to determine plant fresh biomass after 3 weeks of growth under different light intensities. The light-use efficiency was calculated as average fresh weight of plant divided by light source power used (kW h^{-1}). Each measurement was repeated three times with three or four plants per replication.

Gas exchange and chlorophyll *a* fluorescent determination

Gas exchange and chlorophyll *a* fluorescence were monitored according to the method



of Yan et al. (2012). The minimum (F_o) and maximum (F_m) Chl fluorescence levels were determined after the leaves had been dark adapted for 30 min. Photosynthesis, stomatal conductance and chlorophyll fluorescence measurements were taken after the leaf reached steady-state photosynthesis at $150 \mu\text{mol m}^{-2} \text{s}^{-1}$ light of the same red/blue proportion as its growth conditions during treatment. The maximal quantum yield (F_v/F_m) and the effective quantum yield (Φ_{PSII}) were calculated as $F_v/F_m = (F_m - F_o)/F_m$ and $\Phi_{\text{PSII}} = (F_m' - F_s)/F_m'$, respectively.

Determination of total phenolic compounds and 2,2-diphenyl-1-picrylhydrazyl (DPPH) free-radical scavenging capacity

Total phenolic compound concentrations were determined spectrophotometrically (Ragaee et al., 2006) with slightly modifications. The free-radical-scavenging capacity of the lettuce leaf under continuous lighting was evaluated by the DPPH free-radical scavenging capacity, as described by Ragaee et al. (2006).

Nitrate content measurements

The method of Cataldo et al. (1975) was used to evaluate nitrate content in lettuce leaves treated with different continuous lighting. Briefly, 2.0 g leaf samples were ground with liquid nitrogen using a mortar and pestle. The sample powder was transferred into a tube with 10 mL, and boiled for 30 min at 100°C in a water bath. The extracted samples were cooled with tap water, filtered, and diluted to 25 mL with distilled water. The extract (0.1 mL) was further diluted with 0.4 mL 5% (w/v) salicylic acid/concentrated sulfuric acid. After reaction for 20 min, 9.5 mL 8% (w/v) NaOH solution was added. The absorbance monitored at 410 nm was used to calculate nitrate content with respect to its standard curve.

Statistical analysis

At the data were subjected to one-way ANOVA using SAS 9.0 software (SAS Institute, Cary, NC, USA). Significant differences between treatments were evaluated by Duncan's multiple range test at $p < 0.05$.

RESULTS

Effects of light spectral composition on pak choi growth

Shoot fresh and dry weights and leaf areas were significantly affected by the blue light percentage in the light source (Figure 1A-C). The fresh and dry weight of shoots and leaf area were the highest under the 23% blue light treatment, followed by 27 and 100% blue light treatments. The lowest values of these parameters were obtained under the monochromatic red light (0% blue) light treatment. Furthermore, there was a significant linear correlation between fresh weight and leaf area of pak choi (Figure 1D).

Light energy-use efficiency under different light intensities

There were significant differences in plant fresh weight and light-use efficiency under different light intensities (Figure 2). Under the same light intensity ($220 \mu\text{mol m}^{-2} \text{s}^{-1}$), the fresh weight of plants under red and blue LEDs was comparable to that of FL (control), but a decrease in light intensity of combined red and blue LEDs resulted in a significant decrease in shoot fresh weight (Figure 2A). There was a significant difference in light-use efficiency between high light intensity and medium light treatment. Compared with FL, light-use efficiency increased by 55, 114 and 115% for mixed red and blue LEDs with PPFD at 100, 150 and $220 \mu\text{mol m}^{-2} \text{s}^{-1}$, respectively.

Photosynthetic performance of tomato under different LED lights

Tomato plants grown under the white LED light displayed significantly higher rates of photosynthesis than those grown under other LED light (Figure 3A). Plants grown under monochromatic red and mixed red and blue light exhibited similar photosynthetic rates, but plants grown under monochromatic blue light displayed significantly impaired rates of

photosynthesis compared with other spectral compositions of light. Stomatal conductance was highest under monochromatic blue light, followed by white, mixed red and blue, and finally monochromatic red light (Figure 3B). Among different LED light treatments, there was no significant difference in F_v/F_m . The highest and lowest Φ_{PSII} were observed under white and mixed red and blue LED light, respectively. However, there was no significant difference between monochromatic red and blue light treatment in tomato leaf.

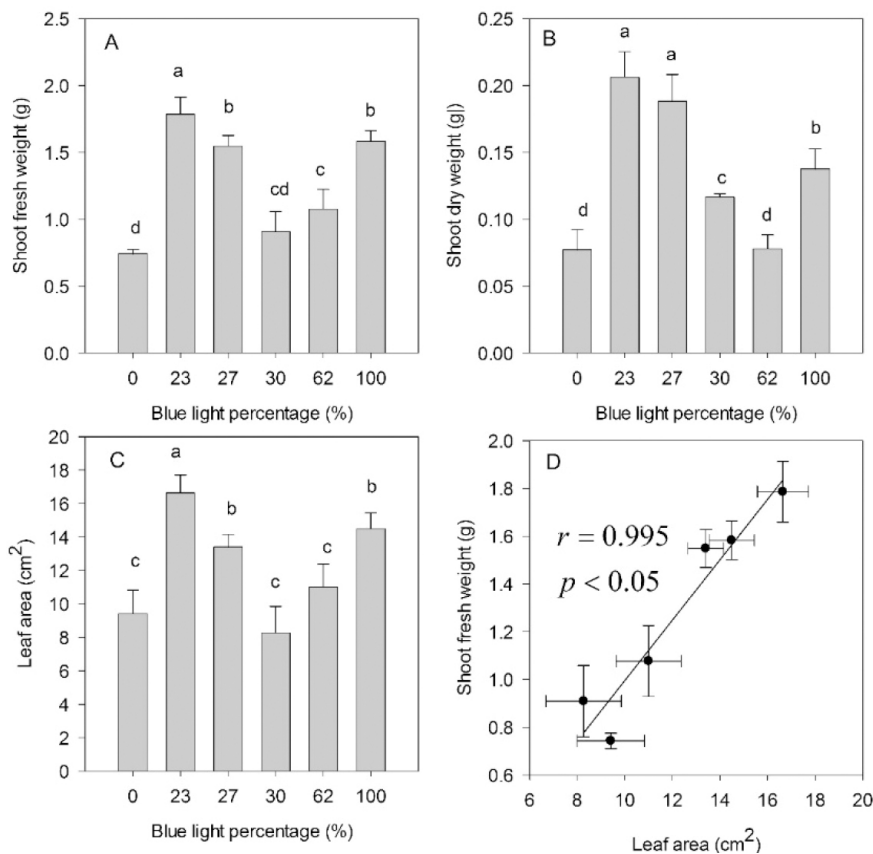


Figure 1. Effects of light spectra on pak choi growth (A-C) and the relation between leaf area and fresh weight (D).

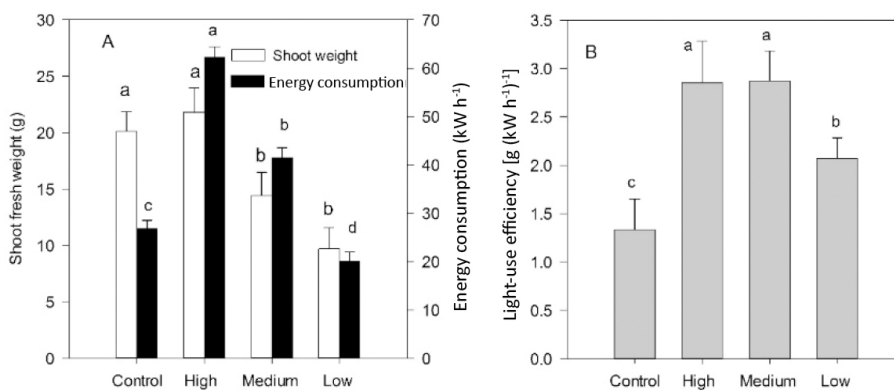


Figure 2. Shoot fresh weight of pak choi and energy-use efficiency under mixed red and blue LED light with high ($220 \mu\text{mol m}^{-2} \text{s}^{-1}$), medium ($150 \mu\text{mol m}^{-2} \text{s}^{-1}$) and low ($100 \mu\text{mol m}^{-2} \text{s}^{-1}$) light intensity.

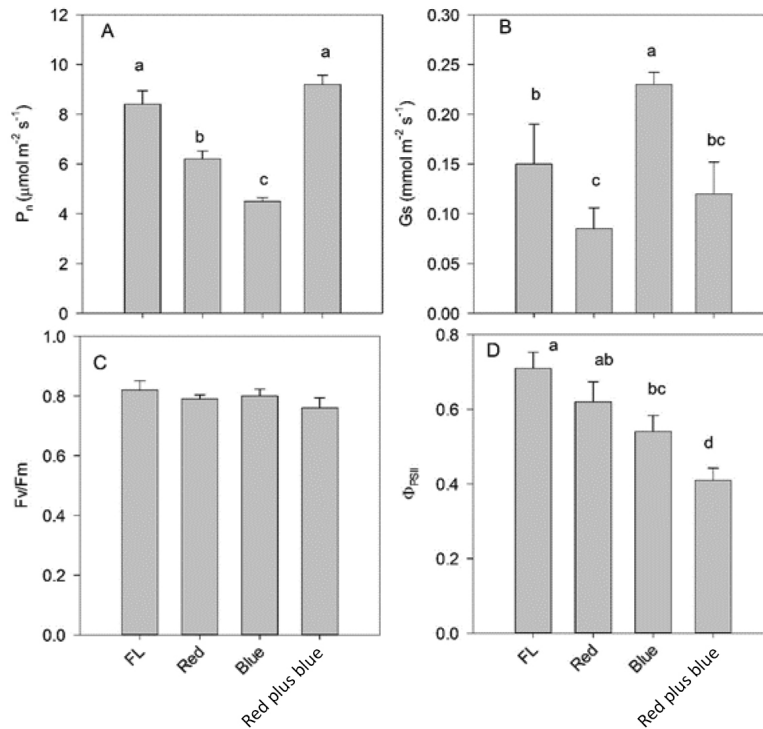


Figure 3. Effects of light spectral composition on photosynthetic performance of tomato seedlings.

Effects of light spectra on nitrate content, total phenolic compounds and scavenging capacity in lettuce under continuous lighting

Nitrate content, total phenolic compounds and free radical scavenge capacity were significantly affected by light spectral composition under continuous light (Table 3). Continuous light (except white LED treatment) for 24 h significantly decreased nitrate accumulation in lettuce leaves. The nitrate content was lowest under RBG treatment, followed by RB, rb and finally by W treatment. Total phenolic compounds and DPPH radical scavenging activity were both markedly increased after 24 h continuous light treatment. However, these parameters did not show significant differences among W, RB and RBG treatments.

Table 3. Nitrate content, total phenolic compounds and scavenging capacity in lettuce after 24 h continuous lighting.

Treatment	Nitrate content (mg kg^{-1})	Total phenolic compounds (mg g^{-1})	DPPH radical scavenging activity ($\mu\text{mol g}^{-1}$)
Control	509.4±22.1a	1.34±0.06c	3.25±0.14c
W	487.3±33.2a	1.69±0.03a	3.90±0.22a
RB	324.7±28.7c	1.70±0.07a	4.26±0.31a
RBG	243.5±21.6d	1.74±0.11a	4.18±0.12a
rb	384.4±20.9b	1.53±0.06b	3.51±0.09b

DISCUSSION

Light is not only the driving-force for photosynthesis but also as important signal to regulate gene expression in plants (Chen et al., 2004). Plant growth and development depend on light intensity, photoperiod, light spectral composition and light direction. In the present study, comparable F_v/F_m of tomato plants were obtained under different light spectra (Figure 3), indicating that light spectral composition showed little effect on

photosynthetic capacity (Osmond, 1994). In plants, there are four types of photoreceptors: phytochromes, cryptochromes, phototropins and UV-B receptors (UVR-8). These photoreceptors share distinct pathway to coordinately regulate transcriptional changes in response to light. Cross-talk among photoreceptors via transcription factors (e.g., HY5 and PIFs) leads to a more complex response of plants to light spectra (Su et al., 2017). Compared with monochromatic red and monochromatic blue LEDs, the significantly lower Φ_{PSII} but markedly higher P_n under mixed red and blue LEDs might lie in coordinated regulation between phytochromes and cryptochromes under combined red and blue light conditions (Wade et al., 2001). Similar results were also reported by Karlsson (1986), who found that there was red and blue light enhancement under combined red and blue light treatment.

A suitable light spectral composition could compensate for the effect of light intensity on photosynthesis and alleviate limitation of low light intensity to photosynthesis (Fan et al., 2013). A similar result was also observed in our study, when different crops were grown under different light intensities and different compositions of light (Figure 2). Besides crop yield, the nutrition and flavour of vegetables are also important for human health, which arouses great concern around the world (Bian et al., 2015). Moderate abiotic and/or biotic stresses are conducive to phytochemical biosynthesis, especially secondary metabolites (Al Hassan et al., 2015). Plants develop complex antioxidant systems to acclimatize to fluctuations in the environment. The fast responses of antioxidant enzyme activities and secondary metabolite accumulation are an important pathway for plants to scavenge reactive oxygen species produced by biotic and/or abiotic stress (Blokhina et al., 2003). During crop or vegetable production, artificial/moderate stress using LEDs could significantly improve plant nutrition quality (Ma et al., 2014). Before harvest, continuous lighting with different light spectra significantly increased lettuce secondary metabolites and concomitantly reduced nitrate content, which poses a threat to human health (Table 3). This is consistent with the study of Steindal et al. (2016), which showed that pre-harvest light spectral composition significantly affected broccoli flavour and nutritional compounds.

Spectral quality drastically effects growth and development. It will probably take several iterations of testing a wide-range of light recipes in order to identify the optimum light regime for growth, development and other desirable traits like flavour and nutrition of vegetables. LED light will make a great contribution to revealing the mechanism of light wavelength regulation of plant growth and substance metabolism. The many different phenotypes already observed between treatments offer many different avenues of exploration to identify the key downstream regulators that are being regulated by specific light signals. LEDs as monochromatic light sources can make flexible combinations based on plant growth requirements. This distinct advantage of LEDs not only provides great help in revealing the mechanism of the plant response to light spectra but also offers a new way to reduce the energy cost of commercial production of crops in the greenhouse.

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