In-door germination and seedling growth of green and red lettuce under LED-light spectrum and subsequent effect on baby leaf lettuce

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Highlights

- *Blue light enhanced germination and increased the number of germinated seeds of green lettuce.*
- *High blue component lights improved the morphology, dry matter percentage, and chlorophyll a/b ratio of lettuce seedlings.*
- *Blue and full-spectrum lights applied to lettuce seedlings affect fresh weight after transplanting.*
- *The anthocyanin content of seedlings was stimulated by blue light at 55 µmol m^{-2} s^{-1}, but even more so by PAR of natural light at 451 µmol m^{-2} s^{-1}.*

Abstract

The spectrum and intensity of light play a significant role in the primary and secondary metabolism of plants. Low intensity can make the photosynthetic process less efficient, while inadequate spectrum can impair plant growth and quality. This study investigates the effect of different LED light spectra at low intensity on germination and growth of lettuce (*Lactuca sativa* L.) seedlings under a temperature-controlled chamber and the subsequent impact on mature plants grown in a greenhouse under natural light. The purpose was to reach a commercial plant seedling using a low amount of energy to achieve the yield potential in a shorter period. The experiment was carried out in three trials. In trial 1, the effect of different LED light wavelengths [100% blue (B); 100% red (R); mixed light 1 (52% blue, 27% green and 21% red) (BGR1), and mixed light 2 (29% blue, 53% green and 17% red and 1% far red) (BGR2)] at low intensity (55 µmol m^{-2} s^{-1} and 12 h light photoperiod) and darkness (control) on germination of two lettuce cultivars ['Levistro' (green) and 'Carmolí' (red)] was evaluated in a controlled temperature chamber (20±1.2°C). In trial 2, the effect of the same light conditions of the first experiment on agronomic characteristics and pigment contents of lettuce seedlings compared to the natural light (control: 451±66 µmol m^{-2} s^{-1}) were evaluated. In trial 3, the seedlings developed under different LED light wavelengths were transplanted to evaluate the subsequent effect on the growth of baby lettuce cultivated hydroponically in the greenhouse under natural light. The results of this study show that red wavelength reduced germination percentage, while lights with a higher blue component (B and BGR1) accelerated germination and increased the number of germinated seeds in 'Levistro'. Red also delayed germination and decreased the number of germinated seeds in 'Carmolí' compared to darkness. Seedlings of 'Levistro' had a higher fresh weight (FW) than 'Carmolí'. In addition, FW increased under BGR2 and R, which coincided with the highest number of leaves and leaf length. Nevertheless, fresh weight was higher under BGR2 and B after transplanting, coinciding with the highest number of leaves. A higher blue component of the light (B and BGR1) increased the dry matter percentage (DMP) of seedlings, but there was no significant difference after transplanting. Chlorophyll (CHL) a and b content increased under BGR2; however, the highest CHL a/b ratio was observed under BGR1 in 'Levistro' and B in 'Carmolí', but it was higher after transplanting when seedlings were grown under B. The anthocyanin (ANT) content of 'Carmolí' seedlings was promoted by a higher blue component of the light (B and BGR1) but significantly increased under natural light (control) at the highest intensity. This work shows that varying the spectrum at low intensity can positively modify the growth and biochemical characteristics of lettuce seedlings, although the effect depends on the cultivar. This modification improves the performance of plants during greenhouse growth after transplanting, especially seedlings grown under B and BGR2.

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Key words: *Lactuca sativa* L.; wavelength; greenhouse; transplanting.

Received for publication: 30 September 2021.
Revision received: 25 May 2022.
Accepted for publication: 25 May 2022.

Italian Journal of Agronomy 2022; 17:1982
doi:10.4081/ija.2022.1982

Introduction

Lettuce (*Lactuca sativa* L.) is one of the most popular leafy vegetables and an important commercial species grown worldwide (Pérez-López *et al*., 2013; Kim *et al*., 2016). Its good taste, low price, and high nutritional content make lettuce highly valued (Pérez-López *et al*., 2013). Furthermore, its seedling stage is an essential operation in crop production (Dusadeerungsikul *et al*., 2020) since a high-quality, healthy, and vigorous seedling allows growers to achieve the maximum potential yield in a shorter period (Gregorio *et al*., 2010; Balliu *et al*., 2017; Song *et al*., 2019). However, limited studies have been conducted on the effects of the light spectrum at low intensity on the germination and development of lettuce seedlings, alongside the subsequent impact on lettuce growth in the greenhouse that mainly affects posterior agronomic characteristics and yield of mature plants.

Light is a form of energy and can vary in quality (colour or wavelength distribution), quantity (intensity, or the amount of energy), and duration (photoperiod) (López *et al*., 2017). The light intensity is an important variable to consider since a higher intensity is correlated with higher electricity consumption (Cui *et al*., 2021). Therefore, a low light intensity can be most advantageous from this point of view. Optimizing intensity, spectrum, and photoperiod could accelerate plant growth (Cui *et al*., 2021) and maximize harvestable yield (Kelly *et al*., 2020). According to Kozai and Niu (2020a), a relatively low light intensity of 100 to 300 µmol m^{-2} s^{-1} is required in indoor systems. However, this type of electric lighting system incurs high capital and operating costs (Kelly *et al*., 2020). Kozai and Niu (2020a) mentioned that electricity consumption for lighting to increase the dry mass of plants reached about 20 to 30% of total costs (Kozai and Niu, 2020b). For the production of annual herbaceous plants, the recommended intensity is 95 to 140 µmol m^{-2} s^{-1} (Currey *et al*., 2012), while for cucumber seedlings, it is 110 to 125 µmol m^{-2} s^{-1} (Cui *et al*., 2021). Various studies have been conducted on lettuce with intensities above 100 µmol m^{-2} s^{-1} (Hoenecke *et al*., 1992; Johkan *et al*., 2010; Chen *et al*., 2014). Nevertheless, studies on lettuce with lower intensities than 95 µmol m^{-2} s^{-1} are limited, although some reported higher nitrate content in leaf vegetables associated with a lower nitrate reductase enzyme activity at an intensity between 52 and 117 µmol m^{-2} s^{-1} (Nájera and Urrestarazu, 2019).

The light spectrum can exert an action on different stages of plant growth. In lettuce seeds, Cantliffe *et al*. (2000) mentioned that germination of some genotypes is controlled by light; specifically, red light plays a promoting role (Contreras *et al*., 2009; Neff, 2012). In seedling production, vegetables prefer blue to red light, but for vegetative growth after transplanting, vegetables prefer red light to blue light (Kozai, 2020). On the other hand, Hernández and Kubota (2014) mentioned that blue-red LED supplemental light caused modifications in cucumber seedlings' morphological parameters and photosynthetic pigment contents. Specifically, blue light decreased hypocotyl length and increased CHL content per leaf area, net photosynthetic rate, and stomatal conductance (Hernández and Kubota, 2016). In lettuce, seedling growth was promoted under fluorescent lights plus red or blue LED lights while inhibited under monochromatic blue LED light (Chen *et al*., 2014). Similarly, blue light prevents the extension and elongation of lettuce seedlings when they are irradiated with monochromatic red light (Hoenecke *et al*., 1992).

The light spectrum can modify several characteristics in mature lettuce plants (full-grown). For example, fresh and dry weights can be improved by combined red and blue LED lights (Lin *et al*., 2013; Bian *et al*., 2016; Lee *et al*., 2016; Naznin *et al*., 2019), particularly when the infrared or blue spectrum was increased in the combination of red and blue LED light (Lee *et al*., 2016; Naznin *et al*. 2019). Also, the red light can increase fresh and dry weights in green and red lettuce plants (Son and Oh, 2013). However, the light absorption could be different between red and green cultivar lettuces exposed to various spectra, provoking variation in the photosynthesis efficiency (Lee *et al*., 2017). Likewise, natural antioxidant pigments such as chlorophylls (CHLs), carotenoids (CAR), and anthocyanins (ANTs) can be modified by the light spectrum. A combination of red and blue LEDs improved CHL content in lettuce (Lee *et al*., 2016; Choong *et al*., 2018) and red light alone (Son and Oh, 2013). However, Lin *et al*. (2013) and Proshkin *et al*. (2020) did not find significant differences in pigment contents (CHL a, b, and CAR) regardless of the light spectrum. On the other hand, Proshkin *et al*. (2020) indicated that under low light conditions in a greenhouse and a minor blue light fraction, red lettuce leaves tend to remain green as ANT accumulation is lower. Other studies showed that a combination of red and blue or red, blue, and green LED lights induced ANTs contents (Stutte *et al*., 2009). In contrast, others mentioned that blue light alone was sufficient to induce ANTs synthesis (Petrella *et al*., 2016). Literature results showed the dissimilar susceptibility of lettuce types to the spectral composition of light (Lee *et al*., 2017; Proshkin *et al*., 2020).

The present study evaluates the differences in germination and seedling characteristics under different red and blue LED lights in a constant temperature chamber and the subsequent effect on baby leaf lettuce growth in a greenhouse under natural light. The primary purpose is to lead to a complete understanding of the growth and phytochemical plant responses to define the appropriate spectrum of light to increase and advance germination, reach a better quality of the seedlings, and higher yield in baby leaf lettuce.

Materials and methods

In this work, a separate experiment was conducted in three trials to determine LED light effects of different wavelengths. First, simulated seed germination was performed, followed by a second seedling trial in which cultivars of green and red lettuces were cultivated at a controlled temperature under different LED light wavelengths. Finally, the subsequent effect on plant growth was tested in the greenhouse in early autumn. Both cultivars, 'Levistro' (green) and 'Carmolí' (red), belong to the type of lettuce named Lollo (*Lactuca sativa* var. *acephala*) and are characterized by loose leaves, curled edges, and wavy lamina. In addition, green and red cultivars were studied to evaluate the content of pigments (CHLs, CAR, and ANTs) in plant response to light quality. Trials 1 and 2 were conducted at a controlled temperature under a growth chamber with LED light treatments. Trial 3 was conducted in a greenhouse at the Faculty of Agricultural Sciences at the University of Chile (33° 34' S, 70° 38' W, Santiago, Chile).

Trial 1. Lettuce seed germination

Experimental treatment conditions

Seeds of green and red lettuce were germinated on four layers of water-saturated papers in Petri dishes. Each Petri dish had 50 seeds of each cultivar. The Petri dishes were placed in a controlled temperature growth chamber (20±1.2°C), 34±8% relative humidity, and under different LED light treatments. The light treatments

included four different wavelengths, and darkness was used as control. For light treatments, LED lamps of different wavelengths composition were used. The spectrums were 100% blue light (B); 100% red light (R); mixed light 1 (52% blue, 27% green and 21% red) (BGR1) and mixed light 2 (29% blue, 53% green and 17% red and 1% far red) (BGR2). Lamps were automatically switched-on for 12 h (8:00 a.m. to 8:00 p.m.). The light intensity of each lamp was adjusted using a dimmer at 55 µmoles of photons m^{-2} s^{-1}. The LEDs' spectral energy distribution scans were recorded at 380 to 780 nm (Figure 1). Using a lighting passport (Asense Tek, Taiwan), the PPFD was regulated under the lamps at the level of the plants. LED light treatments were spatially separated from each other by 0.5 m of space. After four days under light treatments and only when seeds reached a radicle length of ≥3 mm, seedlings were considered to have germinated (ISTA, 1999).

Lettuce seed germination measurements

Germination percentage

Germination percentages (GP) were calculated according to the following equation:

$$GP = (N/Nt)*100 \quad (1)$$

N and Nt were the germinated seeds per day till day 4 and the total number of seeds sown at the beginning, respectively.

Coefficient of the velocity of germination

Coefficient of the velocity of germination (CVG) was used as the number of days in which maximum germination was reached. CVG was calculated according to the equation adapted from Rodríguez *et al*. (2008).

$$CVG = \Sigma (Ni \cdot Di)/\Sigma Ni \quad (2)$$

where Ni and Di were the number of seeds germinated on the day i (i = 1, 2, 3 y 4) and time since sowing (days), respectively.

The velocity of germination

Velocity of germination (VG) indicates the number of seeds that germinated daily. VG was calculated according to the González-Zertuche and Orozco-Segovia (1996).

$$VG = \Sigma (Ni)/\Sigma t \quad (3)$$

where Ni and t were the number of seeds germinated on the day i and germination time from sowing to germination of the last germinated seed, respectively.

Statistical analysis for germination measurements

The trial was set up in a completely randomized design with a factorial structure of 5×2 with four repetitions. Each repetition was an independent Petri dish, and each dish had 50 lettuce seeds. The first factor was the spectrum of LED light, which had five levels: 100% blue (B; peak: 466 nm), 100% red (R; peak: 635 nm), mix light 1 (BGR1: 52% blue, 27% green and 21% red; peak: 467 nm), and mix light 2 (BGR2: 29% blue, 53% green, 17% red and 1% far-red; peak: 452 nm) plus the control (darkness). The second factor corresponded to the lettuce cultivar: 'Levistro' (green) and 'Carmolí' (red). Results are reported as the mean±standard error (SE) values. The data were evaluated by two-way analysis of variance (ANOVA) (F test P≤0.05). The normality of the residuals was checked by the Shapiro Wilk test (P≤0.05). The homogeneity of variance was checked by Bartlett's test and the residuals' independence by descriptive analysis. The differences among the means were compared by LSD Fisher's test (P≤0.05). Statistical analyses were performed with INFOSTAT version 2008.

Trial 2. Lettuce seedling growth

Experimental treatment conditions

Lettuce seeds were sown in seedling trays of 98 cells filled with a mixture of peat and perlite growing medium (1/1=v/v). Each tray was transferred into a controlled temperature growth chamber (20±1.2°C), 44±3.7% relative humidity, and under different LED light treatments. The same light treatments (55 µmoles of photons m^{-2} s^{-1} ×12 h) mentioned above were used. In addition, control was natural light condition applied in a plastic greenhouse with 451±66 µmoles of photons m^{-2} s^{-1} and a photoperiod of 12.2 h at the beginning of the autumn season. The spectral characteristic of each light treatment applied to seedlings is shown in Figure 1. Seedlings were irrigated daily with tap water until the root length reached 5 to 6 cm [30 days after sowing (DAS)].

Biomass measurements

Biomass production was measured on day 30 as fresh weight (FW) and dry matter percentage (DMP). First, the leaves were weighed using an analytical balance (RADWAG, AS/100/C/2, Radom, Poland) and recorded as FW. Then, the leaves were dried at 70°C in an air circulating oven (LabTech, model LDOS50F, Korea) until dry weight (DW) was constant. Finally, the leaf number and leaf length were measured at 30 DAS of all leaves of each seedling from petiole insertion to apex using a ruler.

Pigments content measurements

The method described by Lichtenthaler and Wellburn (1983) was used to measure the CHLs (CHL a and CHL b) and CAR contents. First, 100 mg of leaf tissue was subjected to extraction in 2 mL of 96% (v/v) ethanol and centrifugation at 7300 g for 5 min at 4°C (Hermle Brand centrifuge model Z326K, Wehingen, Germany). Then, the absorbance was measured using a microplate reader (Asys UVM 340, Biochrom, Cambridge, UK).

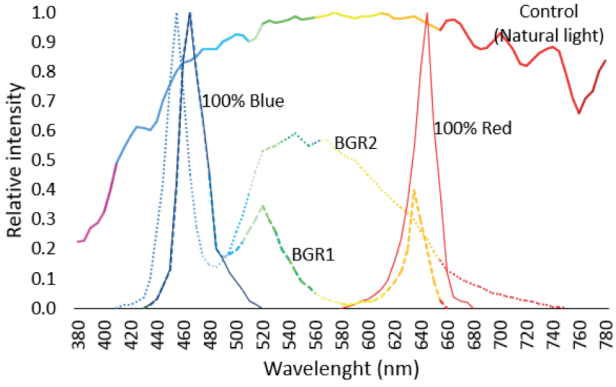

Figure 1. Light spectrum used in the experiment: 100% blue (B; peak: 466 nm); 100% red (R; peak: 635 nm); mix light 1 (BGR1: 52% blue, 27% green and 21% red; peak: 467 nm); mix light 2 (BGR2: 29% blue, 53% green, 17% red and 1% far red; peak: 452 nm) and control (natural light: 29% blue, 16% green, 29% red and 26% far red; peak: 574 nm) for trial 3.

The determination of the total anthocyanin content (TAC) was performed only for 'Carmolí' (red) seedlings by spectrophotometry using the differential pH method according to the protocol described previously by Giusti and Wrolstad (2001) and Nabli *et al*. (2013) with some modifications. On 100 mg of leaf tissue, an extraction in 2 mL of methanol acidified with 1% HCl for 24 h at 5°C was made. Subsequently, the extract was centrifuged at 7300 g for 10 min at 4°C (Hermle Brand centrifuge model Z326K, Wehingen, Germany). Each sample obtained after extraction was divided into two aliquots diluted with the corresponding buffer solutions at pH 1 and 4.5. The absorbance readings were measured at 510 and 700 nm using a microplate reader (Asys UVM 340, Biochrom, Cambridge, UK). Total anthocyanin content in each sample was calculated using the following expression:

$$TAC\ (mg\ L^{-1}) = (A \times MW \times DF \times 1000)/(\varepsilon \times l) \quad (1)$$

where A was calculated as $(A510nm - A700nm)_{pH1.0}$ - $(A510nm - A700nm)_{pH4.5}$; MW corresponded to the molecular weight of cyanidin-3-*O*-glucoside (449.2 g mol^{-1}); DF: dilution factor; l: optical way and ε: molar extinction coefficient of cyanidin-3-*O*-glucoside (20900 L mol^{-1} cm^{-1}). TAC was expressed in µg of ANT per g of leaf powder of DW.

Statistical analysis for biomass and pigment content measurements in seedlings

This part of the experiment was set up in a completely randomized design with a factorial structure of 5×2 with four repetitions. Each repetition considered four independent 98 cells trays containing the same number of lettuce plants. The first factor was the spectrum of LED light, which had five levels: 100% blue (B; peak: 466 nm), 100% red (R; peak: 635 nm), mixed light 1 (BGR1: 52% blue, 27% green and 21% red; peak: 467 nm), and mixed light 2 (BGR2: 29% blue, 53% green, 17% red and 1% far-red; peak: 452 nm) plus the control (natural light). The second factor corresponded to the lettuce cultivar: 'Levistro' (green) and 'Carmolí' (red). Results were reported as the mean±standard error (SE) values of four biological replicates (n=4). Data were evaluated by two-way analysis of variance (ANOVA) (F test P≤0.05). The normality of the residuals was checked by the Shapiro Wilk test (P≤0.05). The homogeneity of variance was checked by Bartlett's test and residuals independence by descriptive analysis. Finally, the differences between the means were compared by LSD Fisher's test (P≤0.05). Statistical analyses were performed with INFOSTAT version 2008.

Table 1. Composition of the nutrient solution used in the hydroponic lettuce crop cycle.

Fertilizer	Concentration (g L^{-1})
Calcium nitrate	116.0
Magnesium nitrate	221.0
Monopotassium phosphate	264.0
Potassium nitrate	223.0
Ammonium nitrate	140.0
Boric acid	0.4
Manganese sulphate	1.2
Copper sulphate	0.1
Ammonium molybdate	0.1
Zinc sulphate	0.1
Iron chelate (6% Fe)	7.0

Trial 3. Lettuce plant growth

Experimental treatment conditions

All seedlings from different LED light treatments grown in the chamber and control (natural light) from the greenhouse were transplanted to an NFT (nutrient film technique) hydroponic system under the same plastic greenhouse. The entire growing period was carried out in a plastic chapel greenhouse 8 m wide, 33 m long, and 5.8 m at zenith height. A polyethylene film on the top and sides 200 µm thick with more than 90% of global light transmission was used to cover the greenhouse. The NFT was formed by 8 0.15×0.07 m and 7 m in length pipes. The plants were transplanted when the seedlings developed 2nd to 3rd true leaves with a root length of 5 to 6 cm that enabled contact with the nutrient solution (30 DAS). The nutrient solution is described in Table 1 (Lara *et al*., 2021). It was kept in constant recirculation, reaching a dissolved oxygen concentration between 8.2 to 9.2 mg L^{-1} measured by an oxygen meter (Oxyguard Handy Polaris, Denmark). The pH was kept between 5.6 and 5.8 and measured with a potentiometer (Hi99301, Hanna Instruments, USA). Adjustments to the pH parameter were made with a prepared acid solution (1.2% phosphoric acid +3.8% nitric acid +95% water). Electrical conductivity was maintained between 2.0 and 2.3 mS measured with a conductivity meter (Hi99301, Hanna Instruments, USA).

Each seedling on a 3×3 cm sponge (polyfoam) square was placed on the NFT system at a density of 46 plants per m^2. 9 plants from each light treatment were replicated three times at the same time on 1 NFT system. The harvest was done after 14 days from transplant with stainless steel scissors when plant leaves reached a maximum length of 10 cm. 4 plants per experimental unit were harvested to perform the measurements. The ambient conditions during the culture were 20.5±3.3°C and 351±95.6 µmoles m^{-2} s^{-1} (Figure 2).

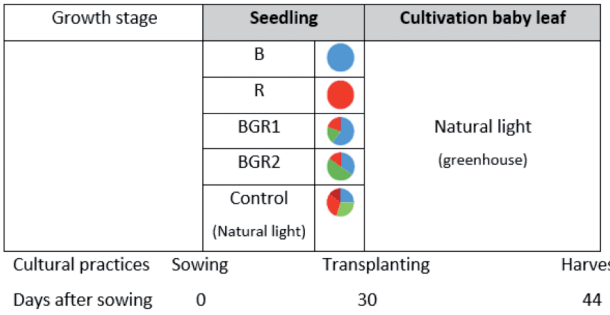

Figure 2. Diagram of the light treatments used during lettuce seedling growth and subsequent cultivation. The light treatments were: B (100% blue), R (100% red), BGR1 (52% blue, 27% green and 21% red), BGR2 (29% blue, 53% green, 17% red and 1% far red) and control (natural light: 29% blue, 16% green, 29% red and 26% far red). All LED light and control treatments were applied at 55 and 451 µmol m^{-2} s^{-1}, respectively, during seedling growth and natural light (351±95.6 µmoles m^{-2} s^{-1}) when grown in the greenhouse.

Biomass and pigment contents measurement

Biomass, leaf number, and pigment content were measured 44 days after sowing using the same methodology described above.

Statistical analysis for biomass and pigment content measurements in plant

This part of the experiment was set up in a completely randomized design with a factorial structure of 5×2 with four repetitions. Each repetition considered nine plants. The first factor was the spectrum of LED light, which had five levels: 100% blue (B; peak: 466 nm), 100% red (R; peak: 635 nm), mixed light 1 (BGR1: 52% blue, 27% green and 21% red; peak: 467 nm), and mixed light 2 (BGR2: 29% blue, 53% green, 17% red and 1% far-red; peak: 452 nm) plus the control (natural light). The second factor corresponded to the lettuce cultivar: 'Levistro' (green) and 'Carmolí' (red). Results were reported as the mean ± standard error (SE) values of four biological replicates (n=4). Data were evaluated by two-way analysis of variance (ANOVA) (F test P≤0.05). The normality of the residuals was checked by the Shapiro Wilk test (P≤0.05). The homogeneity of variance was checked by Bartlett's test and the residuals' independence by descriptive analysis. The differences between the means were compared by LSD Fisher's test (P≤0.05). In addition, a correlation analysis was performed to determine the relationship between seedling biomass and plant biomass. Statistical analyses were performed with INFOSTAT version 2008.

Results

Trial 1. The germination of lettuce seeds is affected by the wavelength of the LED light

Germination percentage

In comparison with darkness, the GP was significantly affected only by Red light treatment (Table 2). Specifically, the GP under Red light was 97.5%, while under dark was 99.5%, which implied a decrease of 2%.

Coefficient of the velocity of germination

CVG was significantly affected by the interaction of light × cultivar, *i.e.*, the effect of light depended on cultivar. Light treatments with a higher proportion of blue light (B and BGR1) decreased CVG in 'Levistro' (green) compared to control in darkness, which means that the seeds germinated 12 hours earlier (Table 2). At the same time, R increased CVG in 'Carmolí' (red), which means the seeds germinated 12 hours later. In addition, compared to 'Carmolí' (red), 'Levistro' (green) decreased the CVG significantly under light treatments, except for the darkness control (Table 2).

Velocity of germination

A significant effect of interaction between light and cultivar was recorded for VG (Table 2). Compared to darkness control, 'Levistro' (green) seeds under BGR1 and B raised the VG significantly by 56 and 42%, respectively, whereas in 'Carmolí' (red) R decreased, the VG considerably by 19.1% (Table 2). On the other hand, 'Levistro' (green) increased the VG significantly under light treatments compared to 'Carmolí' (red), except for the control (Table 2).

Trial 2. Lettuce growth is affected by the wavelength of the LED light

Biomass and morphology of lettuce seedlings

The biomass (*i.e.*, fresh weight and dry matter percentage) and morphology (leaf length and leaf number) of green and red lettuces seedlings are described in Tables 3. The FW of seedlings was significantly affected by both LED light treatments and cultivars without significant interaction between the two factors. Compared to natural light (control), the FW of seedlings of both cultivars

Table 2. Values of germination (%) (GP), coefficient of the velocity of germination (CVG), and velocity of germination (VG) of lettuce cv. 'Levistro' (green) and 'Carmolí' (red) seeds under different light treatments: B (100% blue), R (100% red), BGR1 (52% blue, 27% green and 21% red), BGR2 (29% blue, 53% green, 17% red and 1% far red) and control (darkness). All LED light treatments were applied at 55 µmol m^{-2} s^{-1}.

Factor	Level	GP (%)	CVG (days)	VG (N° seeds $days^{-1}$)
Light treatments (L)	B	100.0±0.0^{a}	1.8±0.1^{c}	29.7±2.8^{a}
	R	97.5±1.0^{b}	2.3±0.2^{a}	25.5±3.7^{b}
	BGR1	99.8±0.4^{a}	1.9±0.2^{bc}	30.5±4.0^{a}
	BGR2	99.8±0.4^{a}	2.0±0.2^{bc}	25.9±2.2^{b}
	Control	99.5±0.4^{a}	2.1±0.1^{ab}	24.4±0.9^{b}
Cultivar (C)	Levistro	99.3±0.6	1.8±0.1^{b}	31.3±2.8^{a}
	Carmolí	99.3±0.8	2.2± 0.2^{a}	23.1±1.4^{b}
L × C	B Levistro	100.0±0.0	1.6±0.07^{de}	34.4±0.92^{ab}
	B Carmolí	100.0±0.0	2.0±0.01^{bc}	24.9±0.03^{de}
	R Levistro	98.0±0.8	2.0±0.15^{bc}	31.2±1.42^{bc}
	R Carmolí	97.0±1.3	2.6±0.17^{a}	19.9±0.8^{f}
	BGR1 Levistro	99.5±0.5	1.5±0.06^{e}	37.8±0.68^{a}
	BGR1 Carmolí	100.0±0.0	2.2±0.04^{b}	23.3±0.15^{ef}
	BGR2 Levistro	100.0±0.0	1.8±0.04^{cd}	29.0±0.44^{cd}
	BGR2 Carmolí	99.5±0.5	2.3±0.23^{b}	22.9±1.01^{ef}
	Control Levistro	99.0±0.5	2.1±0.02^{bc}	24.2±1.29^{ef}
	Control Carmolí	100.0±0.0	2.1±0.01^{bc}	24.6±1.41^{de}
F-test	L	*	*	*
	C	NS	*	*
	L × C	NS	*	*

$^{a-f}$Mean separation within columns by LSD Fisher's multiple range test (P≤0.05); mean (n=4) ±standard error. NS, not significant.

increased significantly under BGR2 and R by about 30.8% and 19.2%, respectively, in 'Levistro' (green) and by 15% and 25%, respectively in 'Carmolí' (red) (Table 3). Among both cultivars, the FW of 'Levistro' (green) seedlings was significantly higher than 'Carmolí' (red) seedlings.

The effect of light on the seedling's DW depended on the interaction light × cultivar (Table 3). In 'Levistro' (green) seedlings, BGR1 and Blue promoted a higher DW than natural light (control) by 24 and 18.8%, respectively. In 'Carmolí' (red), only BGR2 prompted a lower DW of seedlings than natural light (control) by 37.6%.

The DMP of seedlings was affected by the interaction of light and cultivar (Table 3). In particular, the DMP of 'Levistro' (green) seedlings under BGR1 showed a significantly higher value increase by 14.3% than natural light (control). Similar DPM values between 10.2 and 10.5% under B, BGR1, and control were reached in the 'Carmolí' (red) seedlings. At the same time, in both cultivars, DMP were significantly lower in R and BGR2 by 50.5 and 48.6%, respectively, in 'Levistro' (green) and by 29.4% and 37.3%, respectively, in 'Carmolí' (red) compared to control under natural light.

A significant effect of the interaction of light and cultivar was recorded for leaf length of seedlings, *i.e.*, the impact of light depended on the cultivar (Table 3). Compared to natural light (control), leaf length was significantly longer under R by about 279% in 'Levistro' (green) and 174% in 'Carmolí' (red). For B and BGR2, leaf length enhancement was also observed compared to control in both cultivars, while BGR1 reached the lowest values.

Pigment content of the lettuce seedlings

The pigment content of seedlings is reported in Table 5. CHL a, CHL b, and CAR were affected by the interaction of light and cultivar, where the effect of light depended on cultivar. Compared to control, BGR2, BGR1, and B enhanced the CHL a content in both cultivars. On the other hand, in 'Levistro' (green), the CHL content under R was similar to the control while 'Carmolí' (red) under R decreased it. Concerning CHL b content and compared to control, in 'Levistro' (green), BGR2 followed by B increased the content of this pigment; meanwhile, in 'Carmolí' (red), the same effect was observed under BGR2 followed by BGR1. On the other hand, for 'Levistro' (green), the lowest values were observed in control, followed by BGR1, while 'Carmolí' (red) grown in R showed the same effect. In addition, 'Carmolí' (red) had a significantly higher CHL b content than 'Levistro' (green). Regarding CAR content, both cultivars showed an increase under BGR2 and BGR1 compared to the control. In contrast, the lowest content was observed in R light. Between cultivars, 'Carmolí' (red) had a higher carotenoid content than 'Levistro' (green).

Also, the CHL a/b ratio was affected by the interaction of the factors light and cultivar (Table 5). Seedlings of 'Levistro' (green) under BGR1 increased CHL a/b ratio compared to all LED light treatments, while R and BGR2 decreased it. Seedlings of 'Carmolí' (red) under B enhanced CHL a/b ratio compared to all LED light treatments.

Anthocyanin content was measured only in 'Carmolí' (red) and was affected significantly by LED light treatments (Figure 3). Total anthocyanin content was substantially higher in control, while the values were the lowest in R. Moreover, a high blue component in light (>52%) at low PAR promoted the ANT accumulation of 'Carmolí' (red) seedlings more than BGR2 and R (Figures 3 and 4).

Trial 3. Lettuce growth after transplanting is affected by the wavelength of the LED light applied during seedling rearing

Biomass and morphology of lettuce plants

The biomass and leaf number of green and red lettuces plants are described in Table 4. LED light treatments applied during

Table 3. Fresh weight, dry weight, dry matter percentage, leaf length, and leaf number of seedlings of lettuce cv. 'Levistro' (green) and 'Carmolí' (red) grown under different light treatments: B (100% blue), R (100% red), BGR1 (52% blue, 27% green and 21% red), BGR2 (29% blue, 53% green, 17% red and 1% far red) and control (natural light: 29% blue, 16% green, 29% red and 26% far red). All LED light treatments and control were applied at 55 and 451 µmol m^{-2} s^{-1}, respectively.

Factor	Level	Fresh weight (mg seedling^{-1})	Dry weight (mg seedling^{-1})	Dry matter (%)	Leaf length (cm)	Leaf number seedling^{-1}
Light treatments (L)	B	260.9±25.3^{ab}	30.2±4.1^{a}	10.9±0.6^{a}	5.2±0.3^{c}	2.0±0.1^{b}
	R	288.0±44.6^{a}	18.2±3.4^{b}	6.2±0.8^{b}	11.6±0.8^{a}	2.6±0.2^{a}
	BGR1	243.6±29.7^{b}	30.2±5.2^{a}	11.2±0.9^{a}	2.9±0.3^{e}	2.2±0.1^{b}
	BGR2	289.7±43.5^{a}	15.3±1.9^{b}	5.9±0.8^{b}	7.6±0.4^{b}	2.6±0.2^{a}
	Control	230.2±22.3^{b}	26.7±3.5^{a}	10.4±0.6^{a}	3.6±0.3^{d}	2.0±0.1^{b}
Cultivar (C)	Levistro	297.7±35.2^{a}	27.8±5.7^{a}	8.9±1.6	6.4±1.8^{a}	2.2±0.2
	Carmolí	227.2±26.3^{b}	20.5±2.8^{b}	8.9±1.1	6.0±1.5^{b}	2.3±0.2
L × C	B Levistro	290.7±20.5	36.7±2.8^{a}	11.3±0.3^{ab}	5.3±0.2^{d}	2.0±0.0
	B Carmolí	231.1±20.9	23.7±1.9^{c}	10.5±0.4^{b}	5.0±0.1^{d}	1.9±0.2
	R Levistro	307.1±44.8	16.4±3.6^{de}	5.2±0.3^{d}	12.5±0.6^{a}	2.5±0.3
	R Carmolí	268.8±44.8	20.0±3.2^{cd}	7.2±0.3^{c}	10.7±0.6^{b}	2.6±0.3
	BGR1 Levistro	284.3±26.9	38.3±4.3^{a}	12.0±0.1^{a}	3.1±0.2^{f}	2.2±0.2
	BGR1 Carmolí	202.9±14.6	22.1±1.6^{c}	10.3±0.1^{b}	2.6±0.1^{f}	2.2±0.2
	BGR2 Levistro	344.8±46.8	16.6±2.2^{de}	5.4±0.4^{d}	7.6±0.4^{c}	2.5±0.2
	BGR2 Carmolí	234.6±11.0	14.1±1.3^{e}	6.4±0.8^{cd}	7.6±0.2^{c}	2.7±0.3
	Control Levistro	261.7±19.4	30.9±2.6^{b}	10.5±0.3^{b}	3.3±0.1^{ef}	2.0±0.2
	Control Carmolí	198.6±10.9	22.6±3.1^{c}	10.2±0.3^{b}	3.9±0.2^{e}	2.0±0.3
F-test	L	*	*	*	*	*
	C	*	*	NS	*	NS
	L × C	NS	*	*	*	NS

$^{a-f}$Mean separation within columns by LSD Fisher's multiple range test (P≤0.05); mean (n=4) ±standard error. NS, not significant.

seedling growth had a subsequent effect on the biomass of plants harvested 14 days after transplanting. Compared to natural light (control), lettuce plants treated with B and BGR2 were significantly heavier by 16.5% and 18.9%, respectively (Table 4).

The dry weight of plants at harvest after 14 days from transplanting was significantly affected by both LED light treatment and cultivar without significant interaction between the two factors (Table 4). Specifically, only R significantly diminishes the DW of the plant compared to control by 14.3%. Furthermore, 'Levistro' (green) showed a higher DW than 'Carmolí' (red). Nevertheless, no significant differences were found for

Table 4. Fresh weight, dry weight, dry matter percentage, and leaf number of plants of lettuce cv. 'Levistro' (green) and 'Carmolí' (red) grown in a greenhouse for 14 days after application of different light treatments during seedlings: B (100 % blue), R (100% red), BGR1 (52% blue, 27% green and 21% red), BGR2 (29% blue, 53% green, 17% red and 1% far red) and control (natural light: 29% blue, 16% green, 29% red and 26% far red). All LED light treatments and control were applied at 55 and 451 µmol m^{-2} s^{-1}, respectively.

Factor	Level	Fresh weight (g $plant^{-1}$)	Dry weight (g $plant^{-1}$)	Dry matter (%)	Leaf number $plant^{-1}$
Light treatments (L)	B	24.0±3.4^{a}	1.6±0.2^{a}	6.6±0.2	6.90±0.5^{a}
	R	18.8±2.2^{b}	1.2±0.3^{b}	6.4±0.2	5.7±0.2^{c}
	BGR1	20.6±3.0^{b}	1.4±0.2^{ab}	6.8±0.3	6.6±0.6^{ab}
	BGR2	24.5±3.8^{a}	1.6±0.1^{a}	6.5±0.3	7.1±0.9^{a}
	Control	20.6±2.6^{b}	1.4±0.2^{a}	6.9±0.5	6.3±0.7^{b}
Cultivar (C)	Levistro	26.1±2.9^{a}	1.7±0.2^{a}	6.5±0.4	7.4±0.5^{a}
	Carmolí	17.3±1.6^{b}	1.2±0.1^{b}	6.8±0.2	5.6±0.4^{b}
L × C	B Levistro	28.8±3.1	1.9±0.2	6.4±0.3	7.7±0.4^{b}
	B Carmolí	19.2±1.7	1.3±0.1	6.7±0.2	6.2±0.4^{c}
	R Levistro	22.3±1.5	1.4±0.1	6.1±0.2	6.1±0.3^{c}
	R Carmolí	15.3±1.1	1.0±0.1	6.7±0.2	5.2±0.2^{d}
	BGR1 Levistro	24.5±2.4	1.6±0.2	6.6±0.3	7.4±0.3^{b}
	BGR1 Carmolí	16.7±2.2	1.2±0.1	7.0±0.3	5.7±0.4^{cd}
	BGR2 Levistro	30.6±2.8	2.0±0.3	6.4±0.4	8.6±0.3^{a}
	BGR2 Carmolí	18.4±1.3	1.2±0.1	6.6±0.2	5.6±0.5^{cd}
	Control Levistro	24.3±2.5	1.7±0.2	7.0±0.7	7.2±0.5^{b}
	Control Carmolí	16.9±0.8	1.2±0.1	6.8±0.3	5.3±0.5^{d}
F-test	L	*	*	NS	*
	C	*	*	NS	*
	L × C	NS	NS	NS	*

$^{a-d}$Mean separation within columns by LSD Fisher's multiple range test (P≤0.05); mean (n=4) ±standard error. NS, not significant.

Table 5. Chlorophyll a, chlorophyll b, carotenoids, and chlorophyll a/b ratio contents of seedlings of lettuce cv. 'Levistro' (green) and 'Carmolí' (red) grown under different light treatments: B (100 % blue), R (100% red), BGR1 (52% blue, 27% green and 21% red), BGR2 (29% blue, 53% green, 17% red and 1%) and control (natural light: 29% blue, 16% green, 29% red and 26% far red). All LED light treatments and control were applied at 55 and 451 µmol m^{-2} s^{-1}, respectively.

Factor	Level	Chlorophyll a (µg g FW^{-1})	Chlorophyll b (µg g FW^{-1})	Carotenoids (µg g FW^{-1})	Chlorophyll a/b ratio
Light treatments (L)	B	198.6±27.5^{c}	42.0±6.0^{b}	53.7±7.8^{b}	4.8±0.1^{b}
	R	122.04±19.5^{e}	32.5±5.6^{c}	34.9±5.2^{c}	3.8±0.1^{d}
	BGR1	220.5±37.4^{b}	45.2±10.8^{b}	73.9±9.0^{a}	5.3±0.4^{a}
	BGR2	271.1±28.9^{a}	73.6±10.3^{a}	70.5±7.7^{a}	3.8±0.1^{d}
	Control	143.3±24.5^{d}	36.1±8.4^{c}	49.8±7.6^{b}	4.2±0.3^{c}
Cultivar (C)	Levistro	149.9±26.4^{b}	33.2±7.4^{b}	46.8±9.3^{b}	4.7±0.4^{a}
	Carmolí	232.5± 38.9^{a}	58.5±10.6^{a}	66.3±9.0^{a}	4.0±0.3^{b}
L × C	B Levistro	157.4±13.0^{b}	33.0±3.3^{b}	40.6±3.0^{c}	4.8±0.2^{b}
	B Carmolí	240.0±22.2^{c}	50.9±4.6^{c}	66.9±4.9^{b}	4.7±0.1^{b}
	R Levistro	106.8±13.4^{c}	27.6±3.6^{bc}	29.6±3.4^{d}	3.9±0.2^{c}
	R Carmolí	138.0±4.0^{e}	37.3±3.4^{d}	40.1±3.1^{c}	3.7±0.4^{c}
	BGR1 Levistro	152.9±13.2^{b}	25.4±2.6^{c}	67.9±11.5^{a}	6.1±0.2^{a}
	BGR1 Carmolí	288.1±17.0^{b}	65.1±4.8^{b}	79.9±4.1^{a}	4.5±0.1^{b}
	BGR2 Levistro	230.3±14.4^{a}	58.3±4.5^{a}	59.2±3.5^{b}	4.0±0.1c
	BGR2 Carmolí	312.0±25.1^{a}	88.8±8.6^{a}	81.9±6.4^{a}	3.5±0.1^{d}
	Control Levistro	102.2±10.1^{c}	21.6±2.9^{c}	36.8±3.6^{d}	4.8±0.2^{b}
	Control Carmolí	184.4±15.4^{d}	50.7±5.2^{c}	62.8±4.3^{b}	3.7±0.1^{cd}
F-test	L	*	*	*	*
	C	*	*	*	*
	L × C	*	*	*	*

$^{a-e}$Mean separation within columns by LSD Fisher's multiple range test (P≤0.05); mean (n=4) ±standard error.

DW at plant harvest after 14 days from transplanting (Table 4).

In lettuce plants grown in a greenhouse for 14 days, the number of leaves was significantly affected by the interaction of light and cultivar. Specifically, plants originating from seedlings grown under BGR2 and B increased the number of leaves in 'Levistro' (green) and 'Carmolí' (red), respectively. Furthermore, between both cultivars, 'Levistro' (green) had a significantly higher leaf number than 'Carmolí' (red) (Table 4).

Pigment content of the lettuce plants

The pigment content of post-transplant lettuce is reported in Table 6. CHL a, CHL b, and CHL a/b ratio were affected by the interaction of light on the cultivar. For 'Levistro' (green), CHL a content was significantly higher under R than all other LED light treatments. In 'Carmolí' (red), no significant differences were observed in CHL. Furthermore, 'Carmolí' (red) seedlings had higher CHL content than 'Levistro' (green) seedlings.

Regarding CHL b content, B decreased the concentration of this pigment compared to control in both cultivars. In the same way, R promoted the same effect in 'Carmolí' (red), reducing CHL content. Comparing both cultivars, 'Carmolí' (red) showed a higher CHL b content than 'Levistro' (green). Regarding CHL a/b ratio, R also decreased significantly this ratio compared to control, while B showed the highest values with no significant differences from the control. On the other hand, CAR content was affected only by LED light treatments (Table 6), where R followed BGR2 promoted a higher CAR content than the other LED light treatments.

Discussion

Germination of lettuce seeds is affected by the wavelength of LED lights

Light is a crucial factor for lettuce seed germination (Cantliffe *et al*., 2000; Neff, 2012; Paniagua *et al*., 2016). The light spectrum mainly affects the germination percentage of the lettuce seeds (Contreras *et al*., 2009). According to the results, high blue-containing LED lights (BGR1 and B) resulted in a faster germination process and more germinated seeds per day in 'Levistro' (green). At the same time, R delayed germination and decreased the number of seeds germinated per day in 'Carmolí' (red) (Table 2), indicating that the responses are cultivar dependent. Evenari *et al*. (1957) found that the blue region of the spectrum exhibits effects of promotion and inhibition of germination in lettuce seeds. Small *et al*. (1979) mentioned that blue light could induce germination in dormant lettuce seeds. Similarly, Shinomura *et al*. (1996) identified some responses to blue light through phytochrome A (phyA) in *Arabidopsis*, irreversibly triggering seed germination photoinduction upon irradiation under irradiation with extremely low blue light intensity. At the same time, Poppe *et al*. (1998) suggested the role of phyA in promoting seed germination by blue light, which reduces germination rates in the wild type and phyA mutants. The same authors found that the quantitative induction of phyB mutants through blue light was similar to treatment with white light in the wild-type phyA and phyB mutants. This response would demonstrate the dominant role of phyA in promoting seed germination by blue light. On the other hand, red light promoted seed germination by increasing phytochrome-mediated gibberellin levels (Yamaguchi and Kamiya, 2002; Sawada *et al*., 2008), although red light also activates phytochrome B (phyB) (Cho *et al*., 2012). This study indicates that the seeds of both green and red lettuce cultivars under R showed 2% lower results than control in darkness, with a germination percentage of 97.5% (Table 2). This germination percentage was substantially higher than that observed by Sawada *et al.* (2008), who observed 80% germination

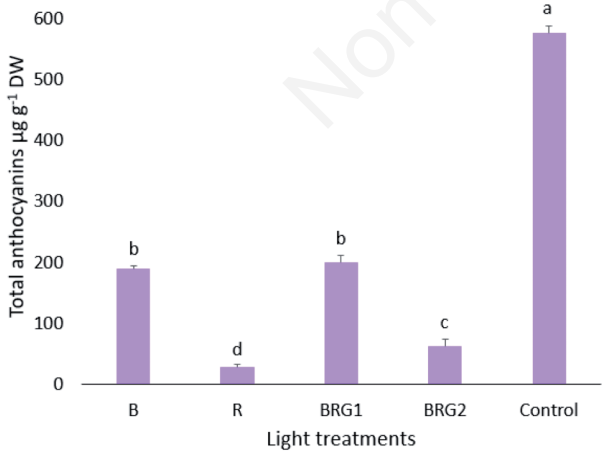

Figure 3. Total anthocyanin content of lettuce cv. 'Carmolí' (red) seedlings grown under different light treatments: B (100% blue), R (100% red), BGR1 (52% blue, 27% green and 21% red), BGR2 (29% blue, 53% green, 17% red and 1%) and control (natural light: 29% blue, 16% green, 29% red and 26% far red). All LED light treatments and control were applied at 55 and 451 µmol m^{-2} s^{-1}, respectively.

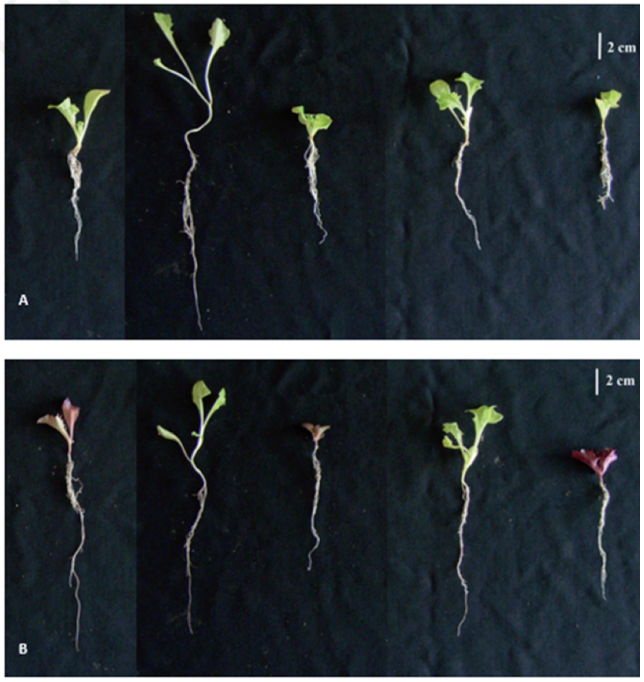

Figure 4. Morphology of seedlings of lettuce cv. 'Levistro' (green) (A) and 'Carmolí' (red) (B) grown under different light treatments. From left to right, plants treated with B (100% blue light); R (100% red light); BGR1 (52% blue, 27% green and 21% red); BGR2 (29% blue, 53% green, 17% red and 1%) and control (natural light; 29% blue, 16% green, 29% red and 26% far red). All LED light treatments and control were applied at 55 and 451 µmol m^{-2} s^{-1}, respectively.

under red light in 'Grand Rapids' lettuce. The difference in results may be since the effect of light depends on the species and cultivar (Naznin *et al*., 2019). On the other hand, in 'Levistro' (green) and 'Carmolí' (red), germinability was stimulated mainly by high blue-containing LED lights (B and BGR1), which denotes a significant action of blue light.

Biomass and morphology of seedlings and lettuce plants are affected by LED light wavelength

According to this study, LED light treatments and cultivar affected the FW of seedlings, while the interaction of light and cultivar affected DMP and leaf length. R and BGR2 promoted a higher FW in lettuce seedlings given to excessive leaf elongation (Table 3; Figure 4), especially under R. Similarly, Tosti *et al*. (2018) observed that red light increased lettuce growth, whereas Battistoni *et al*. (2021) observed a positive impact on FW of spinach. Ngilah *et al*. (2018) found etiolated and long narrow leaves of 'Red Fire' lettuce grown under monochromatic red light after three weeks. Red light favored leaf cell expansion (Tosti *et al*., 2018), promoted shoot length in wheatgrass (Benincasa *et al*., 2020), and hypocotyl growth in cucumber seedlings (Hernández and Kubota, 2016). A low red/far-red ratio effectively inhibits hypocotyl growth via phyA; therefore, a higher proportion of red light would stimulate hypocotyl elongation (Casal *et al*., 2014). This effect of the red wavelength would explain the excessive elongation of the seedlings found in this present study under R (Figure 4). In contrast, hypocotyl elongation was significantly inhibited by the increased intensity of blue light (Kwack *et al*., 2015). Likewise, Hernández and Kubota (2016) indicated that cryptochromes inhibit hypocotyl elongation, consistent with the short stems observed in both cultivars grown in B and BGR1. According to these results, hypocotyl elongation inhibition under blue light was crucial in developing seedlings by maintaining the quality and characteristic rosette shape of green and red lettuces. Notably, LED light treatments applied during seedling growth had a subsequent effect on the plant biomass harvested 14 days after transplanting. Lettuce plants grown in the greenhouse whose seedlings were treated previously with B and BGR2 were significantly heavier than the other plants treated with the other LED light treatments at the harvest. The higher FW of the plants in the greenhouse would be explained by the higher FW of the seedlings after the light treatments. Due to the FW correlations between plants and seedlings reached moderate coefficients (for B: r=0.47 (P=0.05) and R^2=0.2131; for BGR2: r=0.66 (P=0.0027) and R^2=0.4326) other factors could also elucidate the values for B and BGR2. For example, the higher number of leaves and the high CHL a/b ratio would account for the weight gain. A high CHL a/b ratio directly influences the photosynthetic capacity of plants (Li *et al*., 2018). While the stomatal aperture is stimulated by blue light (Hernández and Kubota, 2016), the stomatal conductance is increased by exposure to red light, and weak blue light superimposed on red light-induced further stomatal opening (Hosotani *et al*., 2021). Therefore, there is a synergistic action between the blue and red fraction of light concerning stomatal behaviour. In contrast, the plants whose seedlings were grown under R showed the lowest FW at harvest even though the seedlings grown in the chamber under R showed the highest FW versus natural light (control). Seedlings grown under R showed a lower DW, which would indicate that the higher FW was due to higher water content. Consequently, a lower DMP was also observed under R (Table 3). BGR2 also caused a significant decrease in dry matter accumulation in the seedlings of both cultivars, which was due to a lower DW. On the other hand, Okamoto *et al*. (1997) demonstrated that dry matter production is inhibited in lettuce seedlings with an excessively high red/blue ratio. In the present investigation, because BGR2 and monochromatic red light had a red/blue ratio >0.9, a significant decrease in dry matter accumulation in lettuce seedlings could be caused. In contrast, high blue-containing LED

Table 6. Chlorophyll a, chlorophyll b, carotenoids, and chlorophyll a/b ratio contents of plants of lettuce cv. 'Levistro' (green) and 'Carmolí' (red) after of LED light treatments applied during seedlings: B (100% blue), R (100% red), BGR1 (52% blue, 27% green and 21% red), BGR2 (29% blue, 53% green, 17% red and 1%) and control (natural light: 29% blue, 16% green, 29% red and 26% far red). All LED light treatments and control were applied at 55 and 451 µmol m^{-2} s^{-1}, respectively.

Factor	Level	Chlorophyll a (µg g FW^{-1})	Chlorophyll b (µg g FW^{-1})	Carotenoids (µg g FW^{-1})	Chlorophyll a/b ratio
Light treatments (L)	B	329.9±67.5^{b}	109.8±33.3	77.6±13.4^{c}	3.5±0.4
	R	366.4±51.3^{a}	117.0±25.7	88.9±11.3^{a}	3.3±0.3
	BGR1	338.1±62.4^{b}	117.0±33.9	77.1±11.0^{c}	3.2±0.3
	BGR2	354.2±65.7^{ab}	117.2±31.1	84.1±13.3^{ab}	3.3±0.3
	Control	352.5±66.0^{ab}	126.4±38.8	80.8±12.2^{bc}	3.2±0.4
Cultivar (C)	Levistro	236.5±29.4^{b}	62.7±9.0^{b}	60.5±7.5	3.8±0.2^{a}
	Carmolí	459.9.5± 26.1^{a}	172.3±23.2^{a}	102.9±4.6	2.8±0.2^{b}
L × C	B Levistro	209.2±28.0^{c}	53.5±9.7^{d}	53.9±7.3	4.1±0.4^{a}
	B Carmolí	450.6±29.7^{a}	166.2±21.3^{b}	101.2±4.5	2.8±0.2^{c}
	R Levistro	280.0±30.6^{b}	76.7±10.3^{c}	70.0±7.7	3.7±0.2^{b}
	R Carmolí	452.8±23.1^{a}	157.3±19.8^{b}	107.9±3.4	3.0±0.2^{c}
	BGR1 Levistro	228.0±23.0^{c}	61.8±6.6^{cd}	58.5±6.1	3.7±0.2^{b}
	BGR1 Carmolí	448.2±32.8^{a}	172.2±26.7^{ab}	95.8±5.2	2.7±0.2^{cd}
	BGR2 Levistro	232.9±26.3^{c}	62.5±6.3^{cd}	60.2±7.2	3.7±0.1^{b}
	BGR2 Carmolí	475.6±19.9^{a}	171.9±19.1^{ab}	108.1±2.2	2.9±0.2^{c}
	Control Levistro	232.6±29.6^{c}	59.2±7.7^{cd}	59.9±7.7	3.9±0.1^{ab}
	Control Carmolí	472.5±22.0^{a}	193.6±25.6^{a}	101.7±4.1	2.5±0.2^{d}
F-test	L	*	NS	*	NS
	C	*	*	NS	*
	L × C	*	*	NS	*

$^{a-d}$Mean separation within columns by LSD Fisher's multiple range test (P≤0.05); mean (n=4) ±standard error. NS, not significant.

lights (B and BGR1) positively affected DMP, exceeding 10%. In 'Carmolí' (red), B and BGR1 promoted a similar DMP concerning the control, while in 'Levistro' (green), BGR1 significantly increased DMP. This result is because the lights with a higher blue component promoted a higher DW in the seedlings (Table 3). Similarly, blue-containing LED lights increased the DW of the shoots and roots of the lettuce seedlings compared with those seedlings grown under a white fluorescent lamp (Johkan *et al*., 2010). Likewise, Hogewoning *et al*. (2010) mentioned that dry leaf mass per unit of leaf area in cucumber increased with a higher percentage of blue, up to 50%. Furthermore, the addition of only 7% of blue light prevents dysfunctional photosynthesis, which is related to growth. On the contrary, Chang and Chang (2014) observed that lights of higher red component applied during the growth of lettuce seedlings and then in mature plants promoted a higher DW, inducing a DMP over 12%. Meanwhile, Johkan *et al*. (2012) noted DMP values of over 14% in lettuce plants under white fluorescent lamps versus green light-emitting diodes.

Pigment content of lettuce seedlings and plants is affected by the wavelength of LED lights

The spectrum of PAR plays an essential role in CHL biosynthesis (Frank, 1946). Monochromatic blue light or combined with other wavelengths increases CHL accumulation (Clavijo-Herrera *et al*., 2018; Hernández and Kubota, 2016; Wang *et al*., 2016, Chung *et al*., 2020). In addition, blue light can reverse the low accumulation of CHL under red light (Sood *et al*., 2005; Son *et al*., 2017). According to the present study, the effect of light on CHL content and its ratio in seedlings and plants after transplant were cultivar dependent. Seedlings of both cultivars grown under BGR2 significantly promoted CHL a and b, followed by B in 'Levistro' (green) and BGR1 in 'Carmolí' (red). At the same time, monochromatic red light and natural light (control) showed the lowest values indicating that spectral balance was relevant for CHL formation. A different pattern was observed on the plants treated previously by other light treatments harvested from the greenhouse. In 'Levistro' (green), plants treated with R during seedling rearing showed a higher CHL a and b content than all LED light treatments and control. While 'Carmolí' (red) showed no significant differences in CHL a content, plants whose seedlings were grown under B and R showed a significantly lower CHL b content than the control. Despite these differences, seedlings under B and their subsequent harvest stage showed a higher CHL a/b ratio. This result is supported by Johkan *et al*. (2010) in lettuce seedlings, Wang *et al*. (2016) in mature lettuce, Yousef *et al*. (2021) in tomato, and Hogewoning *et al*. (2010) in cucumber. The higher CHL a/b ratio was mainly due to a low CHL b content. This pigment mainly absorbs blue-purple light (Li *et al*., 2018) and is the main component of the light-harvesting complex (Cammarisano *et al*., 2021). This result indicates a smaller light-harvesting complex per reaction centre, suggesting that seedlings subjected to higher blue component lights are more efficient in light harvesting. On the other hand, Zhang *et al*. (2016) observed a decrease in the CHL a/b ratio due to increased CHL b content in response to low intensity (100 µmol m^{-2} s^{-1}). However, in this work, the low intensity does not affect CHL content and CHL a/b ratio in the seedlings, but rather the spectrum since seedlings kept in natural light whose intensity is eight times higher than LED light treatments did not show high values in CHL content and CHL a/b ratio.

It is relevant to mention that light plays a crucial role in pigment content. The literature showed that CHL content increased in lettuce plants by increasing the blue component relative to red (Son *et al*., 2017; Wang *et al*., 2016). Blue light aids the synthesis and accumulation of CHL biosynthetic enzymes (Sood *et al*., 2005). On the other hand, various studies indicate that red light decreases CHL content in lettuce (Chen *et al*., 2014; Borowski *et al*., 2015; Naznin *et al*., 2019), basil, spinach, kale, pepper (Naznin *et al*., 2019), einkorn wheatgrass (Bartucca *et al*., 2020), and non-heading Chinese cabbage (Fan *et al*., 2013). The current study also observed that the CHL concentration was lower under monochromatic R light (Table 5). Fan *et al*. (2013) found CHL biosynthesis precursors, protoporphyrin IX, Mg proporphyrin IX, and protochlorophyllide in non-heading Chinese cabbage were the lowest under R light. Seedlings under natural light also showed low CHL content, which may be associated with a higher light intensity than LED treatments. Zhang *et al*. (2016) suggest that high light intensity (500 µmol m^{-2} s^{-1}) delays CHL synthesis during the greening process by a metabolic signal, the mitochondrial alternative oxidase-derived plastidial NADPH/NADP+ ratio change. Moreover, at high light intensities, the degradation rate of CHL in plant leaves was higher than the synthesis rate, leading to a decrease in CHL concentration due to chloroplast formation inhibition (Ilić and Fallik, 2017). Fu *et al*. (2012) observed a reduction in CHL content above 400 µmoles m^{-2} s^{-1}, suggesting that higher intensities negatively affect CHL formation.

Carotenoids protect the CHLs from excess light or the wavelengths unsuitable for photosynthesis, thus acting as a selective filter (Ilić and Fallik, 2017). Carotenoids can also absorb excess energy from light that may otherwise lead to singlet oxygen formation from excited CHL molecules (Pizarro and Stange, 2009; Ilić and Fallik, 2017). On the other hand, light is an essential factor in CAR biosynthesis (Pizarro and Stange, 2009), and the spectrum may influence the accumulation of CAR in vegetables (Frede *et al*., 2018). The current study shows that the effect of light on CAR content in seedlings was cultivar dependent. In contrast, the CAR content in plants after transplant was independently affected by light and cultivar. In general, seedlings of both cultivars had high CAR pigments when grown under BGR2 and BGR1 compared to the control. However, after transplanting, the plants whose juvenile stage (seedling) were treated with these same lights did not significantly differ from the control. Only plants whose seedlings were grown under R showed a higher CAR content than control. Johkan *et al*. (2010) found that lettuce seedlings treated with R plus B (RB) light under indoor conditions had higher CAR content compared to fluorescent light (FL) and R light; however, plants subsequently grown in the greenhouse showed no significant differences. In contrast, Chen *et al*. (2016) observed that RB and FL promoted a similar CAR content in seedlings of two rice varieties. In lettuce plants, Amoozgar *et al*. (2017) indicated that CAR concentrations increased in the plants grown under blue and red light compared to those in the greenhouse.

On the other hand, this research showed that the 'Carmolí' (red) had significantly higher CAR content than 'Levistro' (green) in seedlings and the plants after transplanting. Chen *et al*. (2016) also observed differences in CAR content between two rice varieties under various light spectra. So light spectrum and cultivar are relevant factors affecting CAR content. The blue light acts on phytoene synthase and phytoene desaturase (Bohne and Linden, 2002; Frede *et al*., 2018), enzymes that contribute to the core structure of biosynthesis of CAR (Frede *et al*., 2018). The results observed in this research and the literature suggest that the synergistic action of blue and red light is likely to occur during carotenoid formation, activating blue-light photoreceptors and red light photoreceptors, which may have led to the increase of these pigments in lettuce seedlings under BGR1 and BGR2 in indoor conditions. However,

the effect of the wavelength may be masked under greenhouse conditions by both full-spectrum and intensity. Light intensity also plays a role in carotenoid biosynthesis. Under increased light intensity, the grade of photooxidation is greater than the synthesis grade, and CAR are disintegrated (Simkin *et al*., 2003), explaining the low accumulation of CAR in lettuce seedlings under control observed in this research. By contrast, Brazaitytė *et al*. (2015) pointed out that concentrations of various CAR in red pak choi and tatsoi were higher under the illumination of 330 to 440 µmol m^{-2} s^{-1} and in mustard at 110-220 µmol m^{-2} s^{-1}. Hence, the effect of intensity is probably species-dependent.

Effects of wavelength on ANT content have been reported for lettuce (Stutte *et al*., 2009; Baek *et al*., 2013). In the present research, high blue-containing LED lights, such as B and BGR1, promoted the accumulation of ANTs in 'Carmolí'(red) lettuce seedlings. Blue lights would play an essential role in ANT biosynthesis (Stutte *et al*., 2009; Baek *et al*., 2013) by activating genes of enzymes associated with their formation (Meng *et al*., 2004) mediated by cryptochromes (Petrella *et al*., 2016). In contrast, green light can generate the opposite response to that induced by blue light by inhibiting the accumulation of ANTs (Zhang and Folta, 2012; Carvalho and Folta, 2016). Agreement to Zhang and Folta (2012) complete reverse requires the 2:1 ratio of green/blue. These data would explain the low accumulation of ANTs observed in 'Carmolí' (red) seedlings under BGR2, whose spectrum has almost twice the green component concerning the blue part.

On the other hand, the role of intensity cannot be ruled out. As is noted in the present study, the high intensity of natural light (8 times greater than the rest of the LED treatments) increased ANT concentration in seedlings of 'Carmolí' (red) (Figure 3). Anthocyanin biosynthesis is dependent on the light intensity, but results vary according to the species (Kang *et al*., 2013). For example, Petrella *et al*. (2016) mentioned that 1000 µmoles m^{-2} s^{-1} is sufficient to increase ANT content in rough bluegrass. In contrast, Kang *et al*. (2013) observed higher ANT content in lettuce at 290 µmoles m^{-2} s^{-1}. According to Trojak and Skowron (2017), the onset of ANT synthesis and accumulation is due to excessive radiation for the photosynthetic machinery's capacity and sudden exposure to high light exerting a protective action against the raised intensity of light. Our results showed that the ANT content responds to blue light and intensity. Still, the combined effort of these factors observed in natural light also affected the production of this pigment.

Conclusions

Spectrum modification at low intensity significantly impacts the different growth stages of lettuce, although the differences depend on the cultivar. High blue component lights positively affect the germination process by accelerating it and increasing germinated seeds per day. Also, high blue component lights improved the DMP, CHL a/b ratio, and ANT concentration in lettuce seedlings. In addition, broad-spectrum light such as BGR2 positively affects the FW of the seedlings and, together with blue light, increases FW by about 18% in the mature lettuce grown under natural light conditions. On the other hand, LED technology would be feasible to use in vertical farms for seedling production because of its advantages of long service life and low power consumption. In addition, the yield per unit area in controlled growth systems can be up to twice as high as in traditional agriculture.

References

Amoozgar A, Mohammadi A, Sabzalian M, 2017. Impact of light-emitting diode irradiation on photosynthesis, phytochemical composition and mineral element content of lettuce cv. Grizzly. Photosynthetica 55:85-95.

Baek G, Kim M, Kim C, Choi C, Jin B, Son J, Kim H, 2013. The effect of LED light combination on the anthocyanin expression of lettuce. IFAC Proceedings Volumes 46:120-3.

Balliu A, Maršiü N, Gruda N, 2017. Seedling production. In: W. Baudoin, A. Nersisyan, A. Shamilov, A. Hodder, D. Gutiérrez, S. de Pascale, S. Nicola, N. Gruda, L. Urban, J. Tanny (Eds.), Good agricultural practices for greenhouse vegetable production in the South East European countries - Principles for sustainable intensification of smallholder. FAO Plant Production and Protection Paper, Rome, Italy, Volume 230, pp. 189-206.

Bartucca M, Guiducci M, Falcinelli B, Del Buono D, Benincasa P, 2020. Blue:Red LED light proportion affects vegetative parameters, pigment content, and oxidative status of einkorn (Triticum monococcum L. ssp. monococcum) wheatgrass. J. Agric. Food Chem. 68:8757-63.

Battistoni B, Amorós M, Tapia M, Escalona V, 2021. Effect of blue, green or red LED light on the functional quality of spinach (Spinacia oleracea L.). Rev. FCA UNCuyo. 53:98-108.

Benincasa P, Tosti G, Farneselli M, Maranghi S, Bravi E, Marconi O, Falcinelli B, Guiducci M, 2020. Phenolic content and antioxidant activity of einkorn and emmer sprouts and wheatgrass obtained under different radiation wavelengths. Ann. Agric. Sci. 65:68-76.

Bian Z, Cheng R, Yang Q, Wang J, 2016. Continuous light from red, blue, and green light-emitting diodes reduces nitrate content and enhances phytochemical concentrations and antioxidant capacity in lettuce. J. Am. Soc. Hortic. Sci. 141:186-95.

Bohne F, Linden H, 2002. Regulation of carotenoid biosynthesis genes in response to light in Chlamydomonas reinhardtii. Biochim Biophys Acta 1579:26-34.

Borowski E, Michałek S, Rubinowska K, Hawrylak-Nowak B, Grudziński W, 2015. The effects of light quality on photosynthetic parameters and yield of lettuce plants. Acta Sci. Pol. Hortorum Cultus 14:177-88.

Brazaitytė A, Sakalauskienė S, Samuolienė G, Jankauskienė J, Viršilė A, Novičkovas A, Sirtautas R, Miliauskienė J, Vaštakaitė V, Dabašinskas L, Duchovskis P, 2015. The effects of LED illumination spectra and intensity on carotenoid content in Brassicaceae microgreens. Food Chem. 173:600-6.

Cammarisano L, Donnison I, Robson P, 2021. The effect of red & blue rich LEDs vs fluorescent light on Lollo rosso lettuce morphology and physiology. Front. Plant Sci. 12:603411.

Cantliffe D, Sung Y, Nascimento W, 2000. Lettuce seed germination. Hortic Rev. 24:229-275.

Carvalho S, Folta K, 2016. Green light control of anthocyanin production in microgreens. Acta Hortic. 1134:13-8.

Casal J, Candia A, Sellaro R, 2014. Light perception and signalling by phytochrome A. J. Exp. Bot. 65:2835-45.

Chang C, Chang K, 2014. The growth response of leaf lettuce at different stages to multiple wavelength-band light-emitting diode lighting. Sci. Hortic. 179:78-84.

Chen X, Guo W, Xue X, Wang L, Qiao X, 2014. Growth and quality responses of 'Green Oak Leaf' lettuce as affected by monochromic or mixed radiation provided by fluorescent lamp (FL) and light-emitting diode (LED). Sci. Hortic. 172:168-75.

Cho J, Ryu J, Jeong Y, Park J, Song J, Amasino R, Noh B, Noh Y,

2012. Control of seed germination by light-induced histone arginine demethylation activity. Dev. Cell 22:736-48.
Choong T, He J, Qin L, Lee S, 2018. Quality of supplementary LED lighting effects on growth and photosynthesis of two different Lactuca recombinant inbred lines (RILs) grown in a tropical greenhouse. Photosynthetica 56:1278-86.
Chung I, Paudel N, Kim S, Yu C, Ghimire B, 2020. The influence of light wavelength on growth and antioxidant capacity in Pachyrhizus erosus (L.) Urban. J. Plant Growth Regul. 39:296-312.
Clavijo-Herrera J, van Santen E, Gómez C, 2018. Growth, water-use efficiency, stomatal conductance, and nitrogen uptake of two lettuce cultivars grown under different percentages of blue and red light. Horticulturae 4:16.
Contreras S, Bennet M, Metzger J, Tay D, Nerson H, 2009. Red to far-red ratio during seed development affects lettuce seed germinability and longevity. HortSci. 44:130-4.
Cui J, Song S, Yu J, Liu H, 2021. Effect of daily light integral on cucumber plug seedlings in artificial light plant factory. Horticulturae 7:139.
Currey C, Hutchinson V, López R, 2012. Growth, morphology, and quality of rooted cuttings of several herbaceous annual bedding plants are influenced by photosynthetic daily light integral during root development. HortSci. 47:25-30.
Dusadeerungsikul P, Liakos V, Morari F, Nof S, Bechar A. 2020. Smart action. In: A. Castrignano, G. Buttafuoco, R. Khosla, A. Mouazen, D. Moshou, O. Naud (Eds.), Agricultural internet of things and decision support for precision smart farming, Academic Press Ltd., Massachusetts, United States, pp. 225-77.
Evenari M, Neumann G, Stein G, 1957. Action of blue light on the germination of seeds. Nature 180:609-10.
Fan X, Zang J, Xu Z, Guo S, Jiao X, Liu X, Gao Y, 2013. Effects of different light quality on growth, chlorophyll concentration and chlorophyll biosynthesis precursors of non-heading Chinese cabbage (Brassica campestris L.). Acta Physiol. Plant. 35:2721-6.
Frank S, 1946. The effectiveness of the spectrum in chlorophyll formation. J. Gen. Physiol. 29:157-79.
Frede K, Schreiner M, Zrenner R, Graefe J, Baldermann S, 2018. Carotenoid biosynthesis of pak choi (Brassica rapa ssp. chinensis) sprouts grown under different light emitting diodes during the diurnal course. Photochem. Photobiol. Sci. 17:1289-300.
Fu W, Li P, Wu Y, 2012. Effects of different light intensities on chlorophyll fluorescence characteristics and yield in lettuce. Sci. Hortic. 135:45-51.
Giusti M, Wrolstad R, 2001. Characterization and measurement of anthocyanins by UV-visible spectroscopy. Curr. Protoc. Food Anal. Chem. F1.2.1-F1.2.13.
González-Zertuche L, Orozco-Segovia A, 1996. Data analysis methods in seed germination, an example: Manfreda Brachystachya. Bol. Soc. Bot. México 58:15-30.
Gregorio N, Herbohn J, Harrison S, 2010. Guide to quality seedling production in smallholder nurseries. Visayas State University: Leyte, Philippines, pp. 1-43.
Hernández R, Kubota C, 2014. Growth and morphological response of cucumber seedlings to supplemental red and blue photon flux ratios under varied solar daily light integrals. Sci. Hortic. 173:92-9.
Hernández R, Kubota C, 2016. Physiological responses of cucumber seedlings under different blue and red photon flux ratios using LEDs. Environ. Exp. Bot. 121:6-74.
Hoenecke M, Bula R, Tibbitts T, 1992. Importance of 'blue' photon levels for lettuce seedlings grown under red-light-emitting diodes. HortSci. 27:427-30.
Hogewoning S, Trouwborst G, Maljaars H, Poorter H, van Ieperen W, Harbinson J, 2010. Blue light dose-responses of leaf photosynthesis, morphology, and chemical composition of Cucumis sativus grown under different combinations of red and blue light. J. Exp. Bot. 61:3107-17.
Hosotani S, Yamauchi S, Kobayashi H, Fuji S, Koya S, Shimazaki K, Takemiya A, 2021. A BLUS1 kinase signal and a decrease in intercellular CO_2 concentration are necessary for stomatal opening in response to blue light. Plant Cell. 33:1813-27.
Ilić Z, Fallik E, 2017. Light quality manipulation improves vegetable quality at harvest and postharvest: a review. Environ. Exp. Bot. 139:79-90.
International seed testing association (ISTA), 1999. International rules for seed testing. Seed Sci. Technol. 23:1-269.
Johkan M, Shoji K, Goto F, Hashida S, Yoshihara T, 2010. Blue light-emitting diode light irradiation of seedlings improves seedling quality and growth after transplanting in red leaf lettuce. HortSci. 45:1809-14.
Johkan M, Shoji K, Goto F, Hashida S, Yoshihara T, 2012. Effect of green light wavelength and intensity on photomorphogenesis and photosynthesis in Lactuca sativa. Environ. Exp. Bot. 75:128-33.
Kang J, KrishnaKumar S, Sua S, Jeong B, Hwang S, 2013. Light intensity and photoperiod influence the growth and development of hydroponically grown leaf lettuce in a closed-type plant factory system. Hortic. Environ. Biotechnol. 54:501-9.
Kelly N, Choe D, Meng Q, Runkle E, 2020. Promotion of lettuce growth under an increasing daily light integral depends on the combination of the photosynthetic photon flux density and photoperiod. Sci. Hortic. 272:109565.
Kim M, Moon Y, Tou J, Mou B, Waterlan N, 2016. Nutritional value, bioactive compounds and health benefits of lettuce (Lactuca sativa L.). J. Food Compos. Anal. 49:19-34.
Kozai T, 2020. PFAL business and R&D in Asia and North America: status and perspectives. In: T. Kozai, G. Niu, M. Takagaki (Eds.), Plant factory: an indoor vertical farming system for efficient quality food production (2nd ed.). Academic Press Ltd., Massachusetts, United States, pp. 35-76.
Kozai T, Niu G, 2020a. Plant factory as a resource-efficient closed plant production system. In: T. Kozai, G. Niu, M. Takagaki (Eds.), Plant factory: an indoor vertical farming system for efficient quality food production (2nd ed.). Academic Press Ltd., Massachusetts, United States, pp. 35-76.
Kozai T, Niu G, 2020b. Role of the plant factory with artificial lighting (PFAL) in urban areas. In: T. Kozai, G. Niu, M. Takagaki (Eds.), Plant factory: an indoor vertical farming system for efficient quality food production (2nd ed.). Academic Press Ltd., Massachusetts, United States, pp. 35-76.
Kwack Y, Kim Y, Hwang H, Chun C, 2015. Growth and quality of sprouts of six vegetables cultivated under different light intensity and quality. Hortic. Environ. Biotechnol. 56:437-43.
Lara O, Amorós A, Tapia M, Escalona V, 2021. Effect of a photoselective filter on the yield and postharvest quality of 'Viroflay' baby spinach (Spinacia oleracea L.) leaves cultivated in a hydroponic system. Sci. Hortic. 277:109804.
Lee M, Son K, Oh M, 2016. Increase in biomass and bioactive compounds in lettuce under various ratios of red to far-red LED light supplemented with blue LED light. Hortic. Environ. Biotechnol. 57:139-47.
Lee J, Kang W, Park K, Son J, 2017. Spectral dependence of electrical energy-based photosynthetic efficiency at single leaf and canopy levels in green- and red-leaf lettuces. Hortic. Environ.

Biotechnol. 58:111-8.
Li Y, He N, Hou J, Xu L, Liu C, Zhang J, Wang Q, Zhang X, Wu X, 2018. Factors influencing leaf chlorophyll content in natural forests at the biome scale. Front. Ecol. Evol. 6:64.
Lichtenthaler H, Wellburn A, 1983. Determinations of total carotenoids and chlorophylls a and b of leaf extracts in different solvents. Biochem. Soc. Trans. 11:591-2.
Lin K, Huang M, Huang W, Hsu M, Yang Z, Yang C, 2013. The effects of red, blue, and white light-emitting diodes on the growth, development, and edible quality of hydroponically grown lettuce (Lactuca sativa L. var. capitata). Sci. Hortic. 150:86-91.
López R, Fisher P, Runkle E, 2017. Introduction to specialty crop lighting. In: R. López, E. S. Runkle (Eds.), Light management in controlled environments. Meister Media Worldwide, Willoughby, OH, pp. 12-20.
Meng X, Xing T, Wang X. 2004. The role of light in the regulation of anthocyanin accumulation in Gerbera hybrid. Plant Growth Regul. 44:243-50.
Nabli R, Achour S, Jourdes M, Teissedre P, Hela A, Ezzili B, 2013. Anthocyanins composition and extraction from grenache noir (Vitis vinifera L.) vine leaf using an experimental design II- by ethanol or sulfur dioxide in acidified water. J. Int. Des Sci. Vigne Vin 47:301-10.
Nájera C, Urrestarazu M, 2019. Effect of the intensity and spectral quality of LED light on yield and nitrate accumulation in vegetables. HortSci. 54:1745-50.
Naznin M, Lefsrud M, Gravel V, Azad M, 2019. Blue light added with red LEDs enhance growth characteristics, pigments content, and antioxidant capacity in lettuce, spinach, kale, basil, and sweet pepper in a controlled environment. Plants 8:93.
Neff M, 2012. Light-mediated seed germination: connecting phytochrome B to gibberellic acid. Dev. Cell 22:687-8.
Ngilah E, Tsan F, Yap B, 2018. Photoperiod and light spectrum effects on growth, pigment and ascorbic acid content of Lactuca sativa cv. Fire Red under controlled growth environment. Int. Food Res. J. 25:1300-8.
Okamoto K, Yanagi T, Kondo S, 1997. Growth and morphogenesis of lettuce seedlings raised under different combinations of red and blue light. Acta Hortic. 435:149-58.
Paniagua G, Velázquez S, Cruz A, Hernández C, Domínguez F, Rico F, 2016. Pulsed LED light in germination and growth of lettuce seeds. Bothalia J. 46:13.
Patel E, Chandawat D, Patel Y, 2017. Effect of light on seed germination of Vigna radiata. EJPMR 4:444-8.
Pérez-López U, Miranda-Apodaca J, Muñoz-Rueda A, Mena-Petite A, 2013. Lettuce production and antioxidant capacity are differentially modified by salt stress and light intensity under ambient and elevated CO_2. Plant Physiol. 170:1517-25.
Petrella D, Metzger J, Blakeslee J, Nangle E, Gardner D, 2016. Anthocyanin production using rough bluegrass treated with high-intensity light. HortSci. 51:111120.
Pizarro L, Stange C, 2009. Light-dependent regulation of carotenoid biosynthesis in plants. Cienc. Investig. Agrar. 36:143-62.
Poppe C, Sweere, U, Drumm-Herrel H, Schäfer E, 1998. The blue light receptor cryptochrome 1 can act independently of phytochrome A and B in Arabidopsis thaliana. Plant J. 16:465-71.
Proshkin Y, Semenova N, Smirnov A, Chilingaryan N, 2020. The LED phyto lighting for improving the environmental friendliness of growing and productivity of lettuce varieties with red and green leaves. IOP Conf. Ser. Earth Environ. Sci. 578.
Rodríguez I, Adam G, Durán J, 2008. Ensayos de germinación y análisis de viabilidad y vigor en semillas. Agric. Rev. Agropecuaria 912:836-42.
Sawada Y, Aoki M, Nakaminami K, Mitsuhashi W, Tatematsu K, Kushiro T, Koshiba T, Kamiya Y, Inoue Y, Nambara E, Toyomasu T, 2008. Phytochrome- and gibberellin-mediated regulation of abscisic acid metabolism during germination of photoblastic lettuce seeds. Plant Physiol. 146:1386-96.
Shinomura T, Nagatani A, Hanzawa H, Kubota M, Watanabe M, Furuya M, 1996. Action spectra for phytochrome A- and B-specific photoinduction of seed germination in Arabidopsis thaliana. Proc. Natl. Acad. Sci. U. S. A. 93:8129-33.
Simkin A, Zhu C, Kuntz M, Sandmann G, 2003. Light-dark regulation of carotenoid biosynthesis in pepper (Capsicum annuum) leaves. J. Plant Physiol. 160:439-43.
Small J, Spruit C, Blaauw-Jansen G, Blaauw O, 1979. Action spectra for light-induced germination in dormant lettuce seeds. Planta 144:133-6.
Son K, Lee J, Oh Y, Kim D, Oh M, 2017. Growth and bioactive compound synthesis in cultivated lettuce subject to light-quality changes. HortSci. 52:584-91.
Son K, Oh M, 2013. Leaf shape, growth, and antioxidant phenolic compounds of two lettuce cultivars grown under various combinations of blue and red Light-emitting Diodes. HortSci. 48:988-95.
Song J, Cao K, Hao Y, Song S, Su W, Liu H, 2019. Hypocotyl elongation is regulated by supplemental blue and red light in cucumber seedling. Gene 707:117-25.
Sood S, Gupta V, Tripathy B, 2005. Photoregulation of the greening process of wheat seedlings grown in red light. Plant Mol. Biol. 59:269-87.
Stutte G, Edney S, Skerrit T, 2009. Photoregulation of bioprotectant content of red leaf lettuce with light-emitting diodes. HortSci. 44:79-82.
Tosti G, Benincasa P, Cortona R, Falcinelli B, Farneselli M, Guiducci M, Onofri A, Pannacci E, Tei F, Giulietti, M, 2017. Growing lettuce under multispectral light-emitting diodes lamps with adjustable light intensity. Ital. J. Agron. 13:883.
Trojak M, Skowron E, 2017. Role of anthocyanins in high-light stress response. World Sci. News 81(2):150-68.
Wang J, Lu W, Tong Y, Yang Q, 2016. Leaf morphology, photosynthetic performance, chlorophyll fluorescence, stomatal development of lettuce (Lactuca sativa L.) exposed to different ratios of red light to blue light. Front. Plant Sci. 7:250.
Yamaguchi S, Kamiya Y, 2002. Gibberellins and light-stimulated seed germination. J. Plant Growth Regul. 20:369-76.
Yousef A, Ali M, Rizwan H, Ahmed M, Ali W, Kalaji H, Elsheery N, Wróbel J, Xu J, Chen F, 2021. Effects of light spectrum on morpho-physiological traits of grafted tomato seedlings. PLoS One 16:e0250210.
Zhang D, Yuan S, Xu F, Zhu F, Yuan M, Ye H, Guo H, Lv X, Yin Y, Lin H, 2016. Light intensity affects chlorophyll synthesis during greening process by metabolite signal from mitochondrial alternative oxidase in Arabidopsis. Plant Cell Environ. 39:12-25.
Zhang T, Folta K, 2012. Green light signaling and adaptive response. Plant Signal. Behav. 7:1-4.