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Mohammad Sabzalian, Parisa Heydarizadeh, Morteza Zahedi, Amin Boroomand, Mehran Agharokh, Mohammad Sahba, Benoît Schoefs

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RESEARCH ARTICLE

High performance of vegetables, flowers, and medicinal plants in a red-blue LED incubator for indoor plant production

Mohammad R. Sabzalian · Parisa Heydarizadeh · Morteza Zahedi · Amin Boroomand · Mehran Agharokh · Mohammad R. Sahba · Benoît Schoefs

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Abstract In urban agriculture, plant growth is limited by the availability of light. Light emitting diodes (LED) could provide specific quality and quantity of light overcoming existing limitations for normal plant growth. However, there have been very few investigations on the applications of LED in incubators and plant growth chambers. The devices fabricated in this study, were lighted with 100 % red, 100 % blue, 70 % red plus 30 % blue, or 100 % white LED. We cultivated *Mentha piperita*, *Mentha spicata* and *Mentha longifolia*, lentil, basil, and four ornamentals to test the effect of various LED lights on plants productivity compared with field and greenhouse conditions. Our results show that 70/30 % red-blue LED light increased *Mentha* essential oil yield up to four times along with increases in plant photosynthesis and fresh weight compared with field condition. The red-blue LED incubator also led to a better growth of lentil and basil and to higher flower buds and less days to flowering for pot flowers versus greenhouse conditions. Our findings demonstrate that LED could improve economic characteristics of plant species by probably stimulating plant metabolism.

M. R. Sabzalian (✉) · P. Heydarizadeh · M. Zahedi · M. Agharokh
Department of Agronomy and Plant Breeding, College of Agriculture, Isfahan University of Technology, Isfahan 84156-83111, Iran
e-mail: sabzalian@cc.iut.ac.ir

A. Boroomand
Biocomputing Group, Department of Biology, University of Bologna, Bologna 40126, Italy

M. R. Sahba
Arvin Tajhiz Espadana Company, Isfahan Science and Technology Town, Isfahan 84156-83111, Iran

P. Heydarizadeh · B. Schoefs
MicroMar, Mer Molécules Santé, IUML – FR 3473 CNRS, University of Le Mans, EA 2160, Faculté des Sciences et Techniques, Le Mans, France

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1 Introduction

Food supply shortage due to increasing population, limited cultivated lands, serious droughts, floods, and storms as well as pest and disease outbreaks and climate changes, are forcing people to indoor and urban plant production (Yeh and Chung 2009). With demanding world of low energy input and high plant quality output, the desired planting systems should be clean; safe and eco-friendly; and simultaneously, fast, economic, and profitable. Urban culture systems and vertical farming constitute responses to these challenges to make progress in efficiently production of crop plants and vegetables.

In the past, plant culture in controlled-environments had frequent constraints particularly commercially available light sources, which could not provide a stable level of radiant energy with high photosynthetic photon flux and a spectrum close to that of sunlight. These limitations for plants growth were evident, especially for those cultured inside phytotrons and small growth chambers (incubators) (Delepoulle et al. 2008). However, the recent application of light emitting diodes (LED) in different studies suggest that they are high intensity sources of visible radiation for growing horticultural and agronomic plants under closed conditions, dominantly illuminated by blue, red, red-blue, or white LED lights (Brown et al. 1995; Yanagi and Okamoto 1997; Duong et al. 2002; Kurilcik et al. 2008). The recent decrease of both blue and red LED price together with the increase in their brightness has made LED light as an important alternative irradiation possibility, allowing better growth and production of plants and microorganisms (Table 1). For recent review, see Darko et al. (2014).

The invention of light emitting diodes (LED) could be considered as the next great innovation in lighting. The basic

Table 1 Examples of positive effects of LED lighting on plants and microorganisms productions

Type of LED illumination	Effects	Plant/organism	Reference
Red—10 % Blue fluorescent light	Higher shoot dry weight, higher seed yield	Wheat	Goins et al. 1997
Red-blue	Higher shoot and root fresh weight	Micropropagated strawberry plants	Nhut et al. 2000
Red-blue	Larger and higher bulblet fresh and dry weight	*Lilium*	Lian et al. 2002
Red-blue	Improved flower induction, higher number of flower buds and open flowers	*Cyclamen persicum*	Heo et al. 2003
Blue	Higher carotenoid production	*Thraustochytrium* sp. CHN-1	Yamaoka et al. 2004
Red-blue	Higher leaf area and photosynthetic rate	Radish and lettuce	Tamulaitis et al. 2005
Blue	Astaxanthin production	*Haematococcus pluvialis*	Katsuda et al. 2006
Red	Better growth	*Spirulina platensis*	Wang et al. 2007
Red	Higher antioxidant activity	pea	Wu et al. 2007
Red	Higher rooting percentage	grape	Poudel et al. 2008
Red-blue	Economic production	Lettuce	Martineau et al. 2012
Red	Increase in volatile molecules	Petunia, strawberry	Colquhoun et al. 2013

LED: Light Emitting Diode

LED consists of a semiconductor diode, i.e., a chip of semiconductor material doped with impurities to create a junction emitting light wavelength, depending on the band gap energy of the materials that forms the junction (Yam and Hassan 2005). They have been evolved from low-intensity signal indicators into powerful light sources (Yeh and Chung 2009). They are suitable for many applications from street lighting to lighting greenhouses and illuminating urban agricultural system, which is now a growing high-tech industry. With high efficiency, long life expectancy, small physical dimensions, low operating temperatures, and ease of control, LED lights are, therefore, expected to be developed further and become a light source with considerable potential for high-power lighting, as used where plant production could be continued all year round.

For plant culture, in addition to their monochromatic bandwidth, LED lights present several advantages including (1) maintaining constant light output over years, (2) consuming low electricity, and (3) producing low heat radiation while emitting high light intensities (Yeh and Chung 2009). The last property allows providing higher photosynthetic photon flux levels at least 500 μmol m^{-2} s^{-1} and higher ratio of light intensity to heat radiation compared with conventional lighting systems. This introduces LED as a promising lighting source for sustainable production in growth chambers and greenhouses.

To commercialize LED-equipped systems and to make them available to the market, they must be accompanied with sophisticated accessories to assist automatically controlling and adjusting light and probably other environmental parameters. One of these growing systems was constructed and reported for the first time by Folta et al. (2005) for plant research, which could control environmental growth factors; however, plant growth performance was not reported. In the present study, to investigate the suitability of such growth chambers, they were equipped with red-blue LED arrays (Fig. 1) in order to (1) evaluate the potential of the chambers for high quality plant production and (2) determine the effects of LED light on the growth of some medicinal and ornamental plants as compared with those grown in field or greenhouse conditions.

2 Material and methods

2.1 Growth chamber construction

The growth chamber was 1.0 width × 0.6 depth × 1.5 m height surrounded by thick blocks of polyurethane foam (3 cm) for insulation (Fig. 1). A linear temperature gradient of 26–44 °C could be produced by heat flow from the warm copper block to the cool copper block through the walls, floor, and lid of the chamber (each 1 cm thick). Temperature was measured with a digital thermometer accurate to 0.1 °C (ATE040, Arvin Tajhiz Espadana Co., Iran).

2.2 Light control system

Four sets of the control unit (CU) were independently designed to support 120 LED lights in four growth cabinets. LED arrays (OSRAM, Germany), emitting white (380–760 nm), red (650–665 nm), blue (460–475 nm), and red-blue (70 %:30 %) light were affixed to a ceramic and steel support to facilitate efficient heat transfer to the mounting substrate. All the LED lights were 1.0 W (0.25 A of input current) and were driven by a circuit consisted of a standard 2 A power supply delivering 110 VDC to a common bus feeding LED lights (Kaming, Taiwan) in series. Voltage to the arrays that is the illumination intensity was tuned via a self-made potentiometer up to 500 μmol m^{-2} s^{-1} on each separate

Fig. 1 Constructed light emitting diode (LED) incubator with mint plants grown under irradiation platform. This growth chamber is in size of 1.0 width × 0.6 depth × 1.5 m height, equipped with microcontroller for setting the plant growth parameters of light, temperature and humidity to support plant superior growth

incubator at the plant leaf surface. The light intensity was also measured via a light meter (LI-250A, LI-COR Inc., USA) with a 2 π quantum sensor (LI-190, LI-COR Inc., USA) during the plants' growth. A 0.72 K Ω (50 W) power resistor was placed in the circuit as a current limiter. Input and output capacitors were also provided to improve transient response. This configuration was repeated for each growth chamber. The CU was outfitted with two 100 mm 12 V fans, one facing into and one facing out of the CU. Each individual LED sheet was also outfitted with a heat sink to ensure adequate cooling.

A microcontroller containing a logic control for setting the growth parameters was written in the assembly language of ASM51 (Arvin Tajhiz Espadana Co., Iran) and applied on each growth cabinet to adjust temperature, LED brightness, and light/dark duration (16/8 h). The cabinets were thereafter used to raise some vegetables and potted flowers, which are economically important, not previously reported to be studied under LED illumination and cultivable in indoor environments.

2.3 Mint growth evaluation

Five randomly selected rhizomes with the same size of three species of mint, i.e., *Mentha spicata* (spear mint), *Mentha piperita* (pepper mint), and *Mentha longifolia* (horse mint) were cultured in plastic pots (10 × 10 cm) filled with a loam soil amended with cow manure. Mints were collected from the natural habitats of Iran (Heydarizadeh et al. 2013) and planted in the research field (Isfahan University of Technology, Isfahan, Iran, 32° 40′N, 51° 40′E). Rhizomes were planted in pots 1-cm deep. Pots from each mint species were placed in four LED incubators and in the field with three replications. Growth temperature was set at 25±2 °C similar to the outside average daily temperature. Pots were irrigated once a day with tap water (hardness 13, pH 7.5) and nourished with nutrient solution (1 g/L) containing the main nutritive elements (K, Ca, Mg, N, P, and S) once a week. Grown plants were photographed 60 days after planting, and net CO_2 assimilation was measured by a portable photosynthesis meter (LCi ADC Instruments, UK). The aboveground part of the plants was harvested, and fresh and dry weights were determined. Dried leaves were ground using an electric grinder. The fine powder was mixed with 500-mL distilled water and submitted to distillation for 6 h using a Clevenger-type 5 apparatus (British Pharmacopoeia 1980). The oil fraction was collected and weighted, and the percentage of essential oil was calculated based on dry weight unit. Plants grown in the field at the same time were treated similarly.

2.4 Green and potted flower cultivation

Basil (*Ocimum basilicum* L.) and lentil (*Lens culinaris* Medic) were seeds planted and seedlings of primula (*Primula vulgaris* Huds.), marigold (*Calendula officinalis* L.), treasure flower (*Gazania splendens* Moore), and stock plant (*Matthiola incana* (L.) R. Br.) were transplanted into the pots (10 × 10 cm) filled with horticultural soil. Pots were placed inside a red-blue LED incubator (light: 500 μmol m^{-2} s^{-1}; temperature: 25±2 °C; humidity: 60±5 %) and in a greenhouse (as a control) in three replications. Plants were grown to full vegetative growth (for basil and lentil) or full flowering stage (for

potted flowers), photographed and compared with plants grown under the greenhouse condition (natural light: 235–1,800 μmol m^{-2} s^{-1}; temperature: 25±2 °C; humidity: 60±5 %), in terms of days to full growth/flowering, the number of flowers, and plant height.

2.5 Statistical analysis

Plant pots at three replications were arranged in growth cabinets considered as different environments. Data were analyzed, using the Statistical Analysis System (SAS Institute Inc. 1999) program package, according to completely randomized design, and the combined analysis was performed to compare the environments. After an analysis of variance (ANOVA), significant differences among means were determined by least significant difference (LSD) test ($p<0.05$). Principle components analysis (PCA) was also performed using SPSS (SPSS Inc. Chicago IL.V. 17).

3 Results and discussion

3.1 LED light effects on plant growth

To determine the effectiveness of light emitting diodes (LED) irradiation in plant production, economically important plants such as mint, basil, lentil, marigold, primula, treasure flower, and stock plant were grown conventionally (in the greenhouse) or in cabinets equipped with red-blue (70:30 %) LED. Generally, the plants grown under LED light were as healthy as or healthier than those grown in the greenhouse (Fig. 2). In this study, except for mint, only plant parameters mainly determining the price and marketability of the plants including days to flowering and full growth, dwarfness, and profuse flowering (Roh and Lawson 1996; Singh 2006) were recorded. As reported in Table 2, plants grown under red-blue LED irradiation were significantly smaller in size. Okamoto et al. (1997) also found that stem length in lettuce was decreased significantly with an increase in blue light. Under LED irradiation, basil and lentil reached to full growth, and buds of potted flowers were opened significantly earlier than those raised in the greenhouse. The plants grown for flowering developed significantly more floral buds per plants (~twofold) and produced plenty of flowers (Table 2).

According to our knowledge about LED lighting effects on plants, it is difficult to detail the reasons for such effects but it could be suggested that the red irradiation in the absence of far-red light is continuously stimulating phytochromes, photoreceptors controlling node elongation (Schaer et al. 1983), floral transition (Boss et al. 2004), and flowering (Runkle and Heins 2001). On the other hand, blue light inhibits cell growth, and blue light photoreceptors might regulate and change gene expression through which stem elongation is prohibited (Lin 2000; Banerjee and Batschauer 2005).

In *Mentha* species, plant fresh weight was significantly higher in the field in *M. piperita*, while in *M. longifolia*, red-blue LED had significantly higher values than the other environments including field. For *M. spicata*, there was no significant difference in this regard between red-blue LED incubator and field. However, plant dry weight was significantly greater in the field in all species (Table 3). In the absence of red light, i.e., in the incubator with pure blue LED, the fresh growth was significantly lower when compared with pure red LED, except for *M. longifolia*, which did not show significant difference between the two lighting conditions (Table 3). In contrast, plant dry weight was not significantly different between the two pure colors incubators; however, both had lower values than that taken from red-blue LED cabinet. It has been reported that the spectral composition of red LED matches with the red absorbance area of chlorophylls *a* and *b* present in chloroplasts of higher plants (Schoefs 2002; Wang et al. 2007), nevertheless, it has been also reported that blue light has complementary effect. Although, red light may have higher contribution to the plant photosynthesis, our results indicate that neither pure red nor blue LED is enough to satisfy full growth of mint. Brown et al. (1995) compared pepper (*Capsicum annuum* L.) plants grown under red LED with similar plants grown under red LED plus blue light emitted from fluorescent lamps. Pepper biomass was reduced when plants were grown under red LED light without blue wavelengths, compared with those grown under supplemental blue lamps. Therefore, it seems that plant species could not terminate their normal growth under pure red LED light (Yorio et al. 2001).

The plants grown under white LED light displayed a significantly higher fresh weight than blue LED light (Table 3), but except for *M. longifolia*, there was no significant difference between white and red LED lights in this respect. White light is a combination of low intensities of red and blue lights and other low efficient light wavelengths, diluting the effect of red-blue light on the net photosynthesis. This may decrease the growth rate of plants illuminated by white light compared with red-blue LED lights.

Among LED lights, the maximum height belonged to the plants grown under white and red, whereas those grown under blue and red-blue were significantly shorter. However, plants grown in the field were taller than those raised under LED lights (Table 3). Light quality, especially blue and red wavelengths, controls the opening and closure of stomata (Shimazaki et al. 2007). This may change the amount of water in plant tissues, which in turn can affect plant size and height. As indicated in Table 3, the water content in plants grown in the LED cabinets fluctuated from 80.04 to 88.20 %, while the water content of plants raised in the field never exceeded 67.61 %. These data suggest that the shorter height of plants grown under LED compared with field was not related to

Fig. 2 Comparison of growth of peppermint, basil, and marigold under red-blue light emitting diode (LED) in incubator (**a**–**c**) and inside greenhouse (**d**–**f**). The plants grown under red-blue LED were as healthy as or even better than those grown in greenhouse in terms of productivity or the number of flowers

water shortage. Instead, the constant solicitation of blue photoreceptors is likely the source of reduction of plant size. The results showed that red-blue LED light improved water content and fresh weight of mint plants as good as or even better

Table 2 Mean (±SE) comparison of potted plants grown under LED incubator and greenhouse in terms of days to flowering/full growth, the number of flowers and height, indicating the superiority of red-blue LED incubator

Plant	Species	Planting source	Environment	Days to flowering/full growth	No. flowers/pot	Height (cm)
Basil	*Ocimum basilicum*	Seed	Red-blue LED incubator	25±2.1[b]*	–	23±5.6[b]
			Greenhouse	50±7.6[a]	–	47±8.9[a]
Lentil	*Lens culinaris*	Seed	Red-blue LED incubator	21±1.5[b]	–	19±2.5[b]
			Greenhouse	30±5.3[a]	–	28±4.3[a]
Primula	*Primula vulgaris*	Seedling	Red-blue LED incubator	18±2.2[b]	20±4.4[a]	12±2.1[b]
			Greenhouse	40±6.3[a]	10±3.6[b]	20±5.5[a]
Marigold	*Calendula officinalis*	Seedling	Red-blue LED incubator	20±1.5[b]	30±5.2[a]	16±1.8[b]
			Greenhouse	50±6.7[a]	15±6.3[b]	28±4.8[a]
Treasure flower	*Gazania splendens*	Seedling	Red-blue LED incubator	37±2.6[b]	15±4.5[a]	18±2.3[b]
			Greenhouse	60±6.9[a]	7±2.1[b]	32±7.2[a]
Stock	*Matthiola incana*	Seedling	Red-blue LED incubator	28±1.1[b]	45±4.5[a]	21±2.3[b]
			Greenhouse	58±8.5[a]	22±4.8[b]	36±6.4[a]

LED light emitting diode, *SE* standard error of means

*Means followed by different letters in each column in each plant are significantly different according to LSD test ($p<0.05$)

Table 3 Mean comparisons of fresh and dry weight, height, water content, essential oil, and photosynthetic rate of mint plants, sampled 60 days after planting, grown in different LED cabinets and field condition

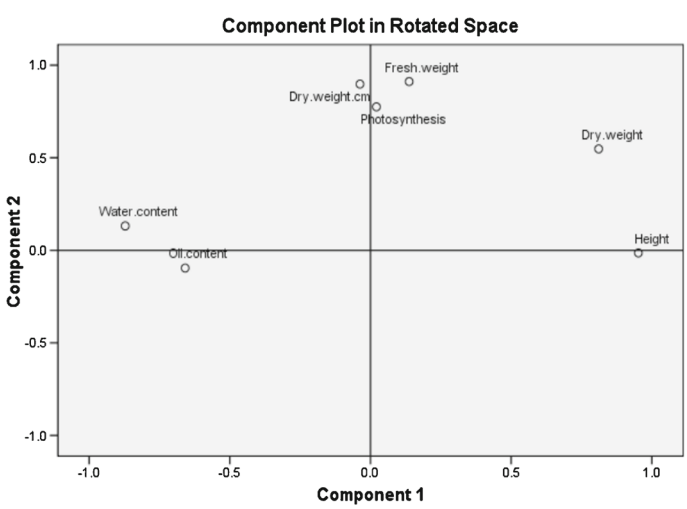
Incubator/ environment	Mentha species	Photosynthesis (μMol CO_2 m^{-2} s^{-1})	Height (cm)	Fresh weight (g/plant)	Dry weight (g/plant)	Dry weight (g) cm^{-1}	Water content (% of fresh weight)	Essential oil content (% of dry weight)
Red LED	*Mentha piperita*	14.28	20.8	16.73	2.24	0.11	86.61	7.00
	Mentha spicata	8.74	24.1	18.72	2.21	0.09	88.20	4.34
	Mentha longifolia	5.62	28.1	13.37	1.89	0.07	85.84	4.37
Blue LED	*Mentha piperita*	8.83	12.1	6.39	1.27	0.10	80.04	3.11
	Mentha spicata	4.96	12.7	6.24	1.24	0.10	80.10	5.03
	Mentha longifolia	3.21	16.2	10.27	1.95	0.12	81.02	3.19
Red-blue LED	*Mentha piperita*	20.70	13.2	27.36	4.45	0.34	83.71	5.12
	Mentha spicata	16.17	14.5	25.89	4.17	0.29	83.89	2.60
	Mentha longifolia	6.48	19.4	36.99	6.10	0.31	83.50	4.86
White LED	*Mentha piperita*	15.27	23.5	16.03	2.67	0.11	83.33	2.34
	Mentha spicata	10.81	26.7	17.85	3.30	0.12	81.48	2.58
	Mentha longifolia	4.52	31.9	17.70	2.70	0.08	84.75	3.53
Field	*Mentha piperita*	18.89	52.1	32.90	11.23	0.22	65.87	1.40
	Mentha spicata	14.00	52.1	23.06	7.47	0.14	67.61	0.66
	Mentha longifolia	4.61	59.7	11.60	8.08	0.14	30.34	3.33
LSD* (0.05)	–	2.08	5.65	3.31	1.06	0.03	5.31	0.59

LSD least significant difference, *LED* light emitting diode

*Means having difference lower than LSD are not significantly different ($P<0.05$).

than field condition. It is worth mentioning that high fresh weight and water content are the two important characteristics of mint for fresh uses.

Since the total biomass production of plants could be influenced by plant size as a function of light quality, the dry weight per each height unit was used as a proxy of yield index after 2 months. The lowest values of 0.08–0.12 g dry weight cm^{-1} were found under red and white LED irradiation and the highest values of 0.29–0.34 g dry weight cm^{-1} were observed under red-blue LED. The values obtained for the plants grown in the field were intermediate (Table 3). The spectral composition of blue (460–475 nm) and red (650–665 nm) LED fits



	Rotated Components	
Characteristic	1	2
Photosynthesis	.021	.775
Height	.952	-.014
Fresh Weight	.137	.912
Dry Weight	.811	.548
Dry Weight cm^{-1}	-.038	.898
Water Content	-.872	.132
Oil Content	-.659	-.096
% of variance	45.29	31.05

Fig. 3 The collinearity among mint characteristics using principle components analysis. The plot of principle components shows high trend between the photosynthetic activity with fresh weight and dry weight per plant size indicating that higher photosynthesis under LED is correlated with increase in fresh and specific dry weight

well to the light absorption spectrum of carotenoids and chlorophyll pigments (Schoefs 2002). Therefore, it was determined whether the increase in dry weight per plant size unit is due to an increase in the photosynthetic activity of the plants. To test this, the CO_2 fixation was first measured and then principle components analysis (PCA) was performed in order to understand the relation between photosynthesis rate and the other variables (Fig. 3). Despite the fact that the values of photosynthesis greatly varied with species and light quality, the highest values were always found in *M. piperita* and the lowest in *M. longifolia* (Table 3). The plot of depicting variables based on the two first principle components shows no particular trend between the photosynthetic activity and dry weight, suggesting that the strategy in utilization of fixed CO_2 is different, depending on the light source. However, there was high collinearity between dry weight per plant size and fresh weight with photosynthesis indicating that higher photosynthesis under LED is correlated with increase in fresh and specific dry weight.

3.2 LED light effects on mint essential oil

It has been well-established that light quality constitutes signals that can trigger metabolic modifications (Liu et al. 2004). To test this with LED light, three mint species grown in growth cabinets each equipped with red, blue, red-blue, or white LED were analyzed for their essential oil content. Our results demonstrated that mint plants of all three species grown under red or red-blue LED light accumulated dramatically higher essential oil content compared with those grown in the field. The maximum increase in oil content was fourfold higher in *M. piperita* grown under red LED light compared with the field. Under blue or white LED light, significant increases in essential oil content were also observed compared with the field except for *M. longifolia* (Table 3).

There is limited information on stimulation of the essential oil accumulation in plants with medicinal properties under LED lights. It seems that red LED may affect the metabolic pathways, leading to an increase in essential oil content. The positive effect of LED light on metabolic pathways has not been well-documented; however, there are possible hypotheses about the role of LED light on increasing biosynthesis of some metabolites. Liu et al. (2004) hypothesized that red LED light may repress the expression of negative regulator genes of pigmentation like LeCOP1-LIKE, resulting in plants with dark green leaves and elevated carotenoid levels. It has been also postulated that LED light could affect secretion or stability of or sensitivity to phytohormones, consistent with the improvement in morphogenesis and productivity of the plant in response to LED lighting (Tamulaitis et al. 2005). However, how these changes take place and affect essential oil accumulation is not yet known and warrant further investigations because of their high positive impact on economic extraction and value of essential oil from plants grown under LED lighting.

4 Conclusion

The development of human population mostly relies on plant species for nutrition, health, and other human activities. Due to environmental constraints and limited cultivated lands, it is critical to develop indoor systems, allowing significantly higher or at least similar production of yield than outdoor environments. To fulfill this demand, a LED incubator was constructed and evaluated in this study. It offers LED lighting regimes supporting complete plant growth and development. The device provided conditions for a faster growth of mint, lentil and basil, and some ornamental plants in our experiments. LED lights were used because they do not include the drawbacks of traditional nondurable lamp systems. The results of this work demonstrated that the studied vegetables and potted flowers took benefits from LED lighting such as dwarfness and increased essential oil production. Among the LED light qualities, most of the beneficial effects were best obtained when red-blue illumination was applied. This conclusion agreed with those attained on the growth and morphogenesis of lettuce and radish in the previous research. LED lighting may provide a novel tool and a new challenge for agricultural research and production alongside its influence on plant morphology and composition. LED lights could be easily integrated into incubators having control systems in which complex lighting programs are facilitated, including selected spectral composition over a growth period or the whole plant developmental stage for improving quality and economic yield of plant species.

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