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Original article

Effects of blue and red LED lights on soilless cultivated strawberry growth performances and fruit quality

S. Nadalini[1], P. Zucchi[2] and C. Andreotti[1]

[1] Free University of Bozen-Bolzano, Faculty of Science and Technology, 39100 Bolzano-Bozen, Italy
[2] Fondazione Edmund Mach, Technology Transfer Centre, 38010 San Michele all'Adige, Italy

Summary

The use of Light Emitting Diodes (LEDs) for protected cultures is quickly growing during the last years. Recent researches have demonstrated that it is possible to use LEDs alone and in combination with other lighting systems to manipulate plant behavior, productivity and crop quality. Nevertheless, our knowledge on the use of single color LEDs to control plant growth and yield is still scarce. In our experiment, the effects of three different lighting systems (LED blue, LED red and fluorescence neon tubes as control) on soilless cultivated strawberry growth and fruit quality were evaluated. Results showed that LED blue light (400–500 nm) induced a higher biomass accumulation, especially at root and crown level. Moreover, LED blue treated plants showed a 25% enhanced fruit set that caused a relevant higher final yield (65 g plant^{-1}) as compared to control and red LED treated plants (45 and 35 g plant^{-1} respectively). Fruit main quality traits were not modified by treatments, the only differences being in fruit color (blue and red LED treated strawberries showed a less saturated color) and anthocyanin concentration (lower level of pelargonidin-3-glucoside in both blue and red LED treated fruits as compared to the control ones). Based on these results we suggest that the use of blue light can be feasible to enhance yield, while maintaining fruit quality, in protected strawberry cultivation systems.

Keywords
anthocyanins, growth chamber, single color light, phenolic compounds, strawberry quality

Significance of this study

What is already known on this subject?

- The use of LEDs delivering different light color alone or in combination with other light sources has been tested on different horticultural crops. Results are nevertheless contrasting as for plant performances and final fruit quality.

What are the new findings?

- Blue LED light was found able to enhance fruit set and final yield of strawberry plants cultivated in growth chamber. The quality of LED-treated strawberries was within the standard of the cultivar for protected cultivation.

What is the expected impact on horticulture?

- The use of LEDs, especially the blue one, can be considered as an interesting tool to enhance productivity of strawberry cultivated under protected conditions. The way of LEDs use (alone or in combination with other light sources) and timing (phenophases) have to be further investigated prior to a commercial implementation of this lighting system.

Introduction

Light is one of the most relevant environmental factors influencing plant behavior. Plants, throughout their photosensory receptors, can react to changes in light quality (color, wavelength), quantity (fluence rate), direction and duration (Fankhauser and Chory, 1997; Folta and Childers, 2008). From a practical standpoint, the manipulation of light characteristics can be used to modulate relevant aspects of horticultural production such as crop yield, quality, nutrient composition and tolerance to abiotic and biotic stresses (Pocock, 2015).

LEDs (Light-Emitting Diodes) represent an innovative tool for the fine-tuning of the light conditions in which plants are cultivated and over the last decade have represented a dynamic field of research for plant scientists and horticulturists. Besides the technical advantages generated by this technology (higher efficiency in converting energy into photosynthetically active radiation, lower cooling requirements, robustness and cost effectiveness as compared to traditional HPS lamps) (Mitchell et al., 2012), LEDs can be considered as a powerful tool to study basic aspects related to the development of specific traits in horticultural plants (Folta and Carvalho, 2015). LEDs have been tested in many experiments in combination with other artificial light sources or as supplement of natural illumination (Li and Kubota, 2009; Johkan et al., 2010; Samuolienė et al., 2012; Choi et al., 2015). Other trials have focused also on the use of LEDs as sole illumination source in growth chamber under controlled conditions (Folta and Childers, 2008; Yoshida et al., 2012; Lin et al., 2013; Piovene et al., 2015; Choi et al., 2015). Within this context, LEDs give the possibility to custom the spectral distribution of light therefore allowing the study of the role of specific wavebands for the growth, development and metabolic accumulation processes in plants (Folta and Carvalho, 2015).

Plants are sensitive to radiation wavelengths in the range from the UV (280–400 nm) to the far-red (700–800 nm) (Folta and Carvalho, 2015). Light is indeed needed by plants not only for photosynthesis, but also for other relevant aspects of their life cycles (i.e., vegetative growth, sexual reproduction) (Dueck et al., 2016). The different plant responses to

light begin with the activation of pigments and photoreceptors that are sensitive to specific wavelengths of light (Folta and Carvalho, 2015). Blue (420–450 nm) and red (600–700 nm) wavelengths are the most effective regions of the light spectra for the determination of the overall plant growth and yield. These radiations are primarily absorbed by the chlorophyll *a* and *b* pigments, which initiate the photosynthetic process (Ouzounis et al., 2015). Moreover, both blue and red light are also able to activate other photoreceptors. Blue light activates cryptochromes, phototropins and FKF1 (Flavin-binding Kelch), which are involved in a large number of physiological processes (biosynthesis of hormones and secondary metabolites, stomatal activity, cell wall elasticity and others) and developmental phases of plants (bud outgrowth, internode elongation, flowering, protection against biotic and abiotic factors). These aspects were extensively reviewed by Huché-Thélier et al. (2016). Similarly, red and far-red (700–800 nm) wavelengths are able to activate other photoreceptors (phytochromes) involved in the regulation of different processes through the plant life cycle (including seed germination, shade avoidance, flowering time, vegetative development, root growth and nutrient uptake) as reviewed by Demotes-Mainard et al. (2016).

The effects of different light compositions on strawberry (*Fragaria* × *ananassa*) growth, yield and fruit quality were the object of some studies conducted recently with the use of LEDs. Leaves and fruits biomass production was found increased in strawberry treated with different combinations of red and blue lights as compared to standard fluorescent lamps (Piovene et al., 2015). Choi et al. (2015) showed that blue light was the most effective in increasing the length of strawberry leaflets in growth chamber conditions, whereas lights with high red to blue ratios (10:1 and 19:1) were found particularly effective in enhancing the number of runners and inflorescences per plant by Naznin et al. (2016). Blue light illumination increased the color index, the sugar content and the antioxidant potential of strawberry fruits during storage (Xu et al., 2014). Moreover, blue light enhanced the level of anthocyanins during strawberry ripening (Kadomura-Ishikawa et al., 2013) as also demonstrated on other berries (Kondo et al., 2014, on grapevine). Differently, Piovene et al. (2015) found a lower amount of flavonoids in red and blue LED-treated fruits as compared with control ones under white fluorescent light. The inconsistency of these results found in literature may have been also partly due to differences in the light intensity that often represents a problem when attempting the comparison of results of experiments conducted under different light parameters (Lin et al., 2013).

In Europe, the strawberry production for fresh market in protected conditions is increasing (Neri et al., 2012). Strawberry soilless cultivation system, performed under greenhouses or plastic tunnels to protect fruits from adverse weather conditions, is considered as necessary to achieve the goal of a year-round fruit availability as requested by the market. The use of artificial light in protected growing conditions is also used to affect relevant aspects of crop physiology, such as for instance the transition phase between vegetative and reproductive growth in strawberry by mean of different duration of the photoperiod (Bosc and Demené, 2009). Moreover, not only the photoperiod but also different light wavelengths (monochromatic lights) have been found able to control the occurrence of flower differentiation in strawberry (Yanagi et al., 2016). Against this background, it appears that our knowledge on the potential benefit of the use of single color lights during the whole process of strawberry growth, development and fruit yield is still limited. We therefore aimed with this research to test the suitability of single color LED lighting systems for protected cultivation of strawberry and to enhance our understanding of the changes induced by blue and red lights on strawberry plant growth dynamic and fruit quality as compared with the one obtained with the traditional fluorescent lamps.

Materials and methods

Plant material and growing conditions

Strawberry frigo plants (*Fragaria* × *ananassa* 'Elsanta'), type waiting bed, 15–18 mm size, were purchased from the Sant'Orsola Cooperative (Pergine Valsugana, TN, Italy). Plants were washed with tap water and roots were cut to approximately 15 cm length. Strawberry plants were afterward planted in 12 L plastic containers containing peat as substrate (Silver Torf, Nordtorf LTD, Latvia). Each plastic container hosted 5 plants that were fertilized once at the beginning of the trial with a mineral-organic fertilizer (Strawberry fertilizer, Compo, 9–5–13 NPK, 4 Mg, 35 S, 25% of organic matter; calculated dose: 0.7 g plant^{-1}) and irrigated with 2 L tap water at two-days interval for the whole experiment duration period.

The experiment was conducted in the growth chamber located at the Free University of Bozen-Bolzano under controlled conditions (day/night cycle: 14/10 h; temperature: 25°C/day, 22°C/night; humidity: 70%). Strawberry plants were grown under 3 different lighting systems: i) neon tubes Fluora 58/77, 2250 Lumen, 58 Watt (Osram GmbH, Munich, Germany) (control); ii) LED blue (600 Watt) (FL 300, Fionia Lighting, Senmatic A/S, Søndersø, Denmark); iii) LED red (same model and producer described above). Spectral distributions of the three lighting systems were measured with a USB2000+RAD spectroradiometer (Ocean Optics Inc., Dunedin, USA) and are shown in Figure 1.

The lighting systems were placed at different distances over the plant canopies in order to adjust the photosynthetic photon flux density (PPFD) to the same value for all treatments (100 µmol m^{-2} s^{-1}; these measurements were performed with a SpectroSense2+ instrument, Skye Instruments Ltd., Llandrindod Wells, United Kingdom). The growth chamber was divided into 3 compartments, each with a bench surface of 1.44 m^2 (1.2 × 1.2 m), by means of wooden panels in order to avoid light contamination among treatments (supplementary material, Figure S1). Five containers of 5 plants each (25 plants per treatment) were used for the 3 lighting systems (75 plants in total). Lighting treatments began 7 days after transplanting (acclimation period) and lasted for 78 days, when there were no fruits left to be harvested and the final plant samplings were performed.

Characterization of plant growth and productivity

The homogeneity of the plant material was evaluated at the beginning of the trial by assessing the total fresh weight (FW, g plant^{-1}) of one plant per container. Strawberry growth was determined at the end of the experiment (day 78) by measuring the FW and dry weight (DW, g plant^{-1}) of the plants. Moreover, the FW and DW of single plant organs (root system, crown, leaves and fruits) were also assessed after dissection. Dry mass was determined after 6 days at 65°C, when weight loss was not measurable anymore. Leaf area (m^2 plant^{-1}) was assessed at the end of the trial with a portable leaf area meter (model LI-3000C; LiCOR, Lincoln, NE, USA). Petioles and flower stems lengths were measured

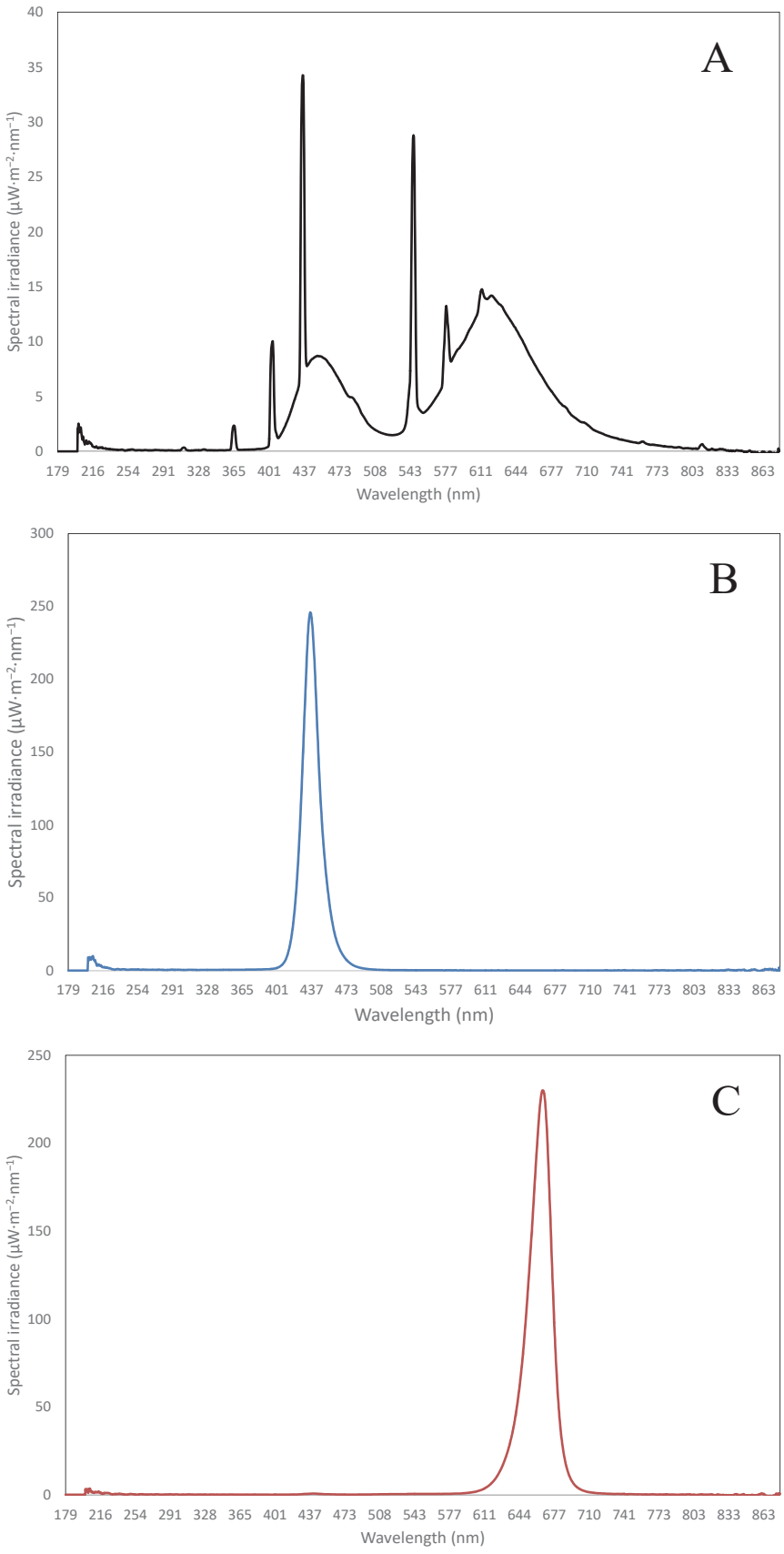

Figure 1. Spectral distribution of the three light treatments used in the experiment: A) neon tubes (control); B) LED blue (peak wavelength at 436 nm); C) LED red (peak wavelength at 666 nm).

Figure S1. The setup of the growth chamber that hosted the experiment.

at the end of the experiment with a flexible ruler; 5 randomly selected petioles and flower stems were measured from each container of the 3 treatments.

The chlorophyll content was indirectly determined by measuring the light transmittance of fully expanded leaves using a portable chlorophyll meter (model SPAD-502; Minolta, Osaka, Japan) and presented as SPAD index values. Measurements were carried out once a week during the whole time-span of the experiment on fully developed leaves. For each plant, 3 SPAD measurements were taken and averaged at each sampling date.

Reproductive parameters were measured at single plant level and average values per plant calculated. The flowers number per plant was determined by counting only the completely open flowers every 2 days during the blooming period (from day 9 to day 19 of the trial). The flowers pollination was performed manually with the help of a paintbrush by visiting each flower at two-day interval for the whole duration of the blooming period. The fruit set ratio was calculated as the percentage of the number of strawberries that grew (diameter over 10 mm) and reached maturation stage to the number of flowers. The average yield per plant (g plant^{-1}) and the average fruit weight (g) were determined by counting and weighting fruits that were picked every 3 days during the harvesting period (from day 37 to day 59 of the trial).

Strawberry quality

Fruit samples for the analysis of quality traits weighed approximately 20 g FW and consisted of 4–5 fully ripe strawberries (the entire fruit surface showed a deep red color). These samples were collected 3 to 5 times during the period of peak productivity (from day 39 to day 50 of the trial) from each container of the different treatments and immediately analyzed for the parameters described below. Flesh firmness (FF) was measured with a penetrometer (PCE-FM200; PCE Instruments Southampton, UK) equipped with a cylindrical probe 3 mm in diameter and expressed in Newton (N). Soluble solid content (SSC), reported as Brix degrees (°Brix), was measured with a digital refractometer (Atago, Tokyo, Japan) on fresh strawberry juice obtained after homogenization of fruits with a kitchen blender and juice filtration with a muslin cloth. Titratable acidity (TA) was determined by adding 20 mL distilled water to 5 mL fresh strawberry juice obtained as described above. This mixture was automatically titrated (Titration Unit Titro-Line easy; Schott Instruments, Mainz, Germany) with a solution of 0.1 mol L^{-1} NaOH to a final pH of 8.1 and the final result was expressed as g L^{-1} of citric acid. Strawberry color as affected by treatments was characterized by colorimetric coordinates (L*, a*, b*) measured with a digital colorimeter (CR-400 Chroma Meter, Konica Minolta Inc., Tokyo, JP). Colorimetric values were measured once on strawberry samples coming from the quantitatively most relevant harvest (day 50 of the trial). Twenty fruits per container (100 fruits per treatment) were individually measured in 3 different points of the strawberry's surface and their colorimetric values averaged.

Analysis of phenolic compounds by HPLC

Phenolic compounds were analyzed following the methodology described in Guerrero-Chavez et al. (2015). Briefly, strawberry samples (collected as described above) were frozen, lyophilized (by freeze-drying for 3 days), extracted with methanol and stored at -80°C until analysis. These were conducted with a Breeze 2 HPLC System (Waters Corporation, Milford, MA, USA) equipped with a photodiode array detector (PDAD). Compounds separation was achieved with a reverse-phase Supelcosil© LC-18 HPLC column (15 cm long, 4 mm internal diameter and octadecylsilane particles of 5 µm diameter). The mobile phase was 0.01 M phosphoric acid (solvent A) and 100% methanol HPLC grade (solvent B) with the following gradient: 0 min 0% B; 0–40 min 50% B; 40–45 min 100% B followed by an equilibrium time of 15 min. The injection volume was 10 µL and the flow rate was 1 mL min^{-1}. For each class of phenolic compounds, the quantitatively most relevant compounds in strawberry fruits (as also reported in Valentinuzzi et al., 2015) were identified and quantified. These were: pelargonidin-3-glucoside (belonging to the class of anthocyanins); catechin (class of flavan-3-ols); quercetin-3-galactoside (class of flavonols) and *p*-coumaric acid (class of hydroxycinnamic acids). Identification was con-

ducted by comparing the UV spectra (within the wavelength range 200–600 nm) and retention times of each selected compounds with the ones of standard molecules acquired from Sigma Aldrich (St. Louis, MO, USA). Quantification was performed with the external calibration curve method at 280 nm wavelength.

Statistical analysis

For the statistical analysis of the results, the experimental set up was considered as a nested design, using the light treatment as the fixed factor and the containers (used as independent blocks) as the random factor, nested in the light treatments. Data from all measured parameters were considered as dependent variables and subjected to hierarchic (nested) ANOVA combined with a post hoc multiple comparison of the means using the Tukey test (for $p \leq 0.05$). Moreover, the Principal component analysis (PCA) was used to display the samples in an unsupervised pattern recognition map (score plot) and to observe relationships between samples and variables (from the comparison of the score and loading plot). Autoscaling of data was performed when variables with different unit of measure were considered, therefore obtaining variables with zero mean and unit standard deviation. Statistical analysis were performed with the software IBM SPSS Statistics version 23.

Results and discussion

Plant growth and yield performances

Light treatments did not affect significantly strawberry weight accumulation (both FW and DW) at the end of the trial (Table 1). Differently, when the DW/FW ratio was calculated, it was found significantly higher in LED blue treated plants, both at root and crown level when compared to control plants and at root level only when compared with LED red plants. A similar effect of LED blue light (at 470 nm) in promoting root dry matter accumulation was also evidenced in red leaf lettuce 17 and 45 days after sowing (Johkan et al., 2010). LED blue treated plants were characterized by significantly reduced total leaf area (0.08 m^2 $plant^{-1}$) as compared to both LED red and control plants. The effect of lighting treatments on strawberry plants morphology was also evident at petioles and flower stems level. LED blue light treatment induced a significant greater petiole (+15%) and flower stem (+30%) elongation as compared to control and LED red lightings. Similar results were also observed by Choi et al. (2015) on strawberry 'Daewang'. This effect could be explained in the context of the positive role played by blue light in the cell elongation processes and promotion of vertical shoot elongation as part of the mechanisms that plants adopt to avoid shade from other neighboring plants (Huché-Thélier et al., 2016).

Chlorophyll in strawberry leaves was monitored throughout the entire growing period and expressed as SPAD index (Figure 2). Chlorophyll in leaves presented a decreasing trend in all the 3 treatments with control leaves showing higher chlorophyll content during the whole duration of the experiment (78 d). LED blue light decreased significantly the chlorophyll content in leaves as compared to control (approximately a 20% reduction), whereas LED red presented intermediate values. Our results are in agreement with the ones of Choi et al. (2015) that showed how blue light illumination was the less effective for the accumulation of chlorophyll in strawberry leaves sampled 15 days before fruit harvest. Plants tend to adapt their content of chlorophyll pigments to the available light spectrum and this process has been found specie-dependent (Huché-Thélier et al., 2016). Cucumber leaves as well as lettuce seedlings treated with blue light showed lower total chlorophyll accumulation when compared with white light (Wang et al., 2015; Johkan et al., 2010). Differently, blue light was found more effective than red light in the induction of chlorophyll synthesis in barley leaves (Bukhov et al., 1992). Further studies are needed (for instance regarding the Chlorophyll a/b ratio) in order to explain possible relationships between light quality, chlorophyll accumulation, photosynthesis and primary productivity during the strawberry growing cycle.

Final fruit yield ranged from 35 to 65 grams per strawberry plant (Table 2). Similar low-level yields were also found by Piovene et al. (2015) and Choi et al. (2015) for different strawberry cultivars ('Elsinore' and 'Daewang', respectively) cultivated under growth chamber conditions. As for the effect of treatments on this parameter, yield was significantly higher in LED blue plants (65.8 g $plant^{-1}$). The enhanced productivity of LED blue plants derived mainly from an approximately 25% higher fruit set percentage

Table 1. Strawberry growth parameters as affected by light treatments. In the same row, different letters indicate significant differences according to Tukey HSD test at $p \leq 0.05$.

	Control	LED blue	LED red	*p*-value
Tot FW (g $plant^{-1}$) $_{time\ 0}$	60.5[x]	62.7	56.6	ns
Tot FW (g $plant^{-1}$)	110.7	140.5	116.9	ns
Tot DW (g $plant^{-1}$)	13.9	18.2	16.1	ns
Roots DW/FW	0.20 b	0.25 a	0.20 b	0.021
Crown DW/FW	0.29 b	0.36 a	0.32 ab	0.008
Leaves DW/FW	0.26	0.30	0.29	ns
Fruits DW/FW	0.08	0.08	0.08	ns
Plant tot DW/FW	0.24 b	0.28 a	0.25 ab	0.017
Leaf:root weight ratio	1.44	1.25	1.61	ns
Leaf area (m^2 $plant^{-1}$)	0.11 a	0.08 b	0.10 a	0.010
Petiole length (cm)	23.4 b	27.6 a	25.2 b	0.041
Flower stem length (cm)	14.7 b	21.8 a	16.5 b	0.029

$_{time\ 0}$: beginning of the trial (all other data refer to time 78 d, end of the trial).
[x] Mean values; $n = 20$ for leaf area, $n = 25$ for petiole and flower stem length; $n = 5$ for the other parameters.

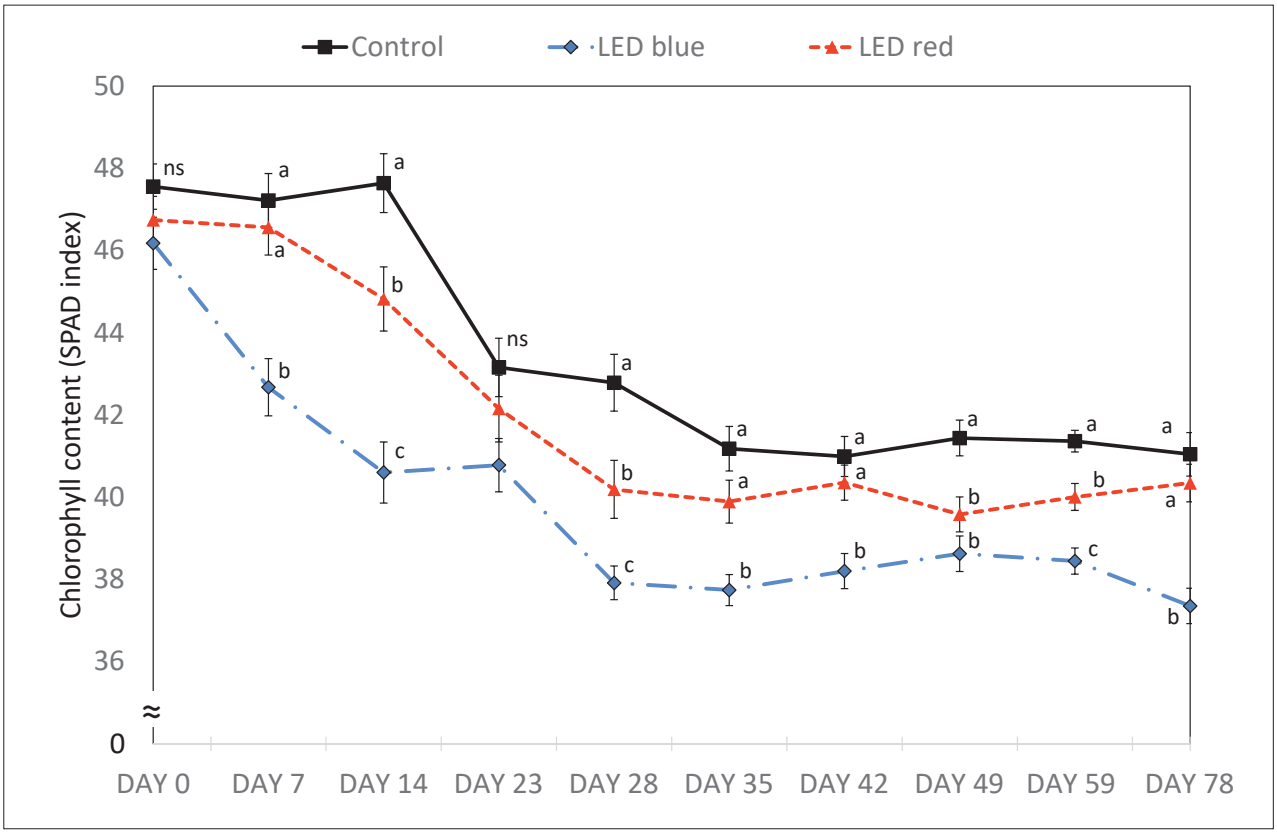

Figure 2. Chlorophyll content (SPAD index) during strawberry plants growth as affected by light treatments. Different letters indicate significant differences according to Tukey HSD test at $p \leq 0.05$, $n = 25$ for each date.

as compared to both control and LED red treatments (Table 2). Despite the higher number of fruits per plants, LED blue strawberries showed similar size as control and even a higher average fruit weight than LED red ones. The positive effect of blue light on strawberry fruit yield was also described in other studies, but always when blue light was combined (or added) to other light sources (Choi et al., 2015; Piovene et al., 2015). As far as we are aware, this is the first finding of a yield-promoting effect of LED blue light as a sole light source in growth chamber conditions. Supplemental LED blue light was found effective in enhancing the yield of strawberry plants grown under plastic greenhouse exposed to natural sunlight (Choi et al., 2015). Moreover, strawberry yield was improved by a combination of red and blue LED light treatment when compared with fluorescent light (control) (Piovene et al., 2015). The effect of blue LED light in promoting yield was also described in other species, such as *Solanum lycopersicum*. Two tomato cultivars ('Myoko' and 'Beiju') showed significantly higher yield when additionally supplemented with blue light during night time as compared with red, white and no additional illumination (Xu et al., 2012). Differently from our results, this yield enhancement was attributed mainly to an increase in fruit size instead of fruit number per plant and was supported by higher leaf photosynthetic activities (i.e., photosynthetic capacity and maximum quantum yield). The blooming time, as well as the fruit maturation dynamic, was only slightly affected by treatments. LED blue treated plants flowered one day earlier and reached 50% of the total yield 4 days in advance as compared to the other treatments (data not shown). These results are partially in agreement with the ones by Yoshida et al. (2012), obtained with ever-bearing strawberry selections.

Strawberry quality

Main quality parameters of soilless cultivated strawberries under artificial light were within limits of literature data for 'Elsanta' cultivar (Josuttis et al., 2010; Vóca et al., 2007; Guerrero-Chavez et al., 2015). As shown in Table 3, the main

Table 2. Strawberry productive parameters as affected by light treatments. In the same row, different letters indicate significant differences according to Tukey HSD test at $p \leq 0.05$.

	Control	LED blue	LED red	*p*-value
Number of flowers (n plant^{-1})	17.9[x]	16.4	18.4	ns
Number of fruits (n plant^{-1})	9.2 b	12.4 a	8.4 b	0.027
Fruit set (%)	50.9 b	75.9 a	49.7 b	0.022
Average fruit weight (g)	4.7 ab	5.4 a	4.3 b	0.042
Yield (g plant^{-1})	45.7 b	65.8 a	35.8 b	0.006

[x] Mean values ($n = 25$).

Table 3. Strawberry fruit quality traits at harvest as affected by light treatments. In the same row, different letters indicate significant differences according to Tukey HSD test at $p \leq 0.05$.

	Control	LED blue	LED red	*p*-value
Total soluble solids (°Brix)	6.2[x]	7.2	6.6	ns
Flesh firmness (N)	2.2	2.3	2.4	ns
Titratable acidity (g L^{-1})	7.3	6.8	7.4	ns
L*	41.9	40.9	42.4	ns
a*	40.7 b	39.5 c	41.2 a	<0.001
b*	30.8 b	28.3 c	31.6 a	0.005

[x] Mean values ($n = 15$) for °Brix, FF and TA; $n = 100$ for colorimetric coordinates (L*, a* and b*).

fruit quality traits (total soluble solids, flesh firmness, titratable acidity) were similar after the 3 applied lighting treatments. Also in Choi et al. (2015) the majority of the considered fruit quality traits (fructose, glucose, oxalic acid, citric acid, malic acid) were not affected by light treatments under growth chamber conditions, sucrose being the only sugar compound that was found lower in blue light treated strawberries.

Strawberries belonging to different treatments could not be distinguished for their color by eyes, nevertheless colorimetric data showed some differences between treatments. Fruits belonging to the three different treatments had the same lightness (L*), but different a* and b* coordinates. More precisely, LED blue strawberries resulted characterized by a significantly lower a* (less red) and b* (less yellow) color coordinates as compared to both the control and red light treated ones, therefore describing fruits characterized by a less saturated color. Moreover, LED red strawberries presented significantly higher a* and b* values when compared to control fruits. We did not find direct evidence of pre-harvest treatments with LEDs of strawberry color coordinates in the literature. Anthocyanins (primary responsible pigments for the red coloration of fruits) content was found positively affected by treatments with blue radiations (at 405 and 465 nm) of immature strawberries at white stage (Kadomura-Ishikawa et al., 2013). Kim et al. (2011) showed that post-harvest treatments with LEDs at different wavelengths (470, 525 and 630 nm) resulted in a higher anthocyanins synthesis (up to +21%) and reddish coloration 4 days after harvest. Similar evaluation conducted on another fruit specie (tomato) showed a positive effect of blue or white additional light treatment on red color development as compared with red or no additional light (Xu et al., 2012).

Strawberry phenolic composition

Chromatographic evaluation was conducted at 280 nm, a wavelength that allows a good detection of the phenolic compounds belonging to the four main classes of phenolic compounds in strawberry fruits (anthocyanins, flavan-3-ols, flavonols and hydroxycinnamic acids). The HPLC chromatograms showed homogeneous profiles, independently from the lighting treatments. There were however differences in the concentration of some of the considered compounds induced by light treatments (Table 4). The pelargonidin-3-glucoside concentration was significantly reduced by both LED lighting treatments as compared to control (approximately 2.3 mg g^{-1} DW in LED red and blue treated fruits vs. 2.8 mg g^{-1} DW of control ones). The concentration of the other considered phenolic compounds (catechin, *p*-coumaric acid and quercetin-3-glucoside) were not significantly affected by treatments. There are contrasting examples in literature regarding the effect of LEDs lighting treatments on the phenolic accumulation in strawberry fruits. Choi et al. (2015) found that, both in growth chamber and plastic greenhouse, red LED light was the most effective in the final accumulation of phenolics in strawberry fruits, whereas the combination of red and blue LED induced the highest final accumulation of anthocyanins. On the contrary, similarly to our results, Piovene et al. (2015) found no positive effect of different LEDs treatments, but higher flavonoids concentration in control fruits developed under a fluorescent light. Further studies conducted on other horticultural species (i.e., red leaf lettuce, *Lactuca sativa*), agreed on outlining a positive effect of the blue light component in enhancing the final anthocyanins accumulation in leaves (Li and Kubota, 2009; Johkan et al., 2010; Goto, 2012). According to our findings and the information available in the literature, the effect of LEDs on the phenolic compounds metabolism can be therefore defined as highly specie-dependent.

LEDs effects on strawberry performances: characterization with PCA

PCA was performed in order to evidence differences between the three treatments and to outline those variables which can better explain them. Figure 3 shows the scores (treatments, on the left) and the loadings (variables, on the right) of the PCA analysis. Samples belonging to the LED blue treatment are grouped separately from samples of the other two treatments. Clusters separation occurs mainly along the PC 2 (explaining 24.1% of the total variance), whereas

Table 4. Main phenolic compounds concentration in strawberry fruits as affected by light treatment. In the same row, different letters indicate significant differences according to Tukey HSD test at $p \leq 0.05$.

mg g^{-1} DW	Control	LED blue	LED red	*p*-value
Pelargonidin-3-glucoside	2.82[x] a	2.28 b	2.32 b	0.023
Catechin	1.17	1.12	0.97	ns
p-Coumaric acid	0.48	0.45	0.45	ns
Quercetin-3-galactoside	0.36	0.28	0.28	ns

[x] Mean values ($n = 5$).

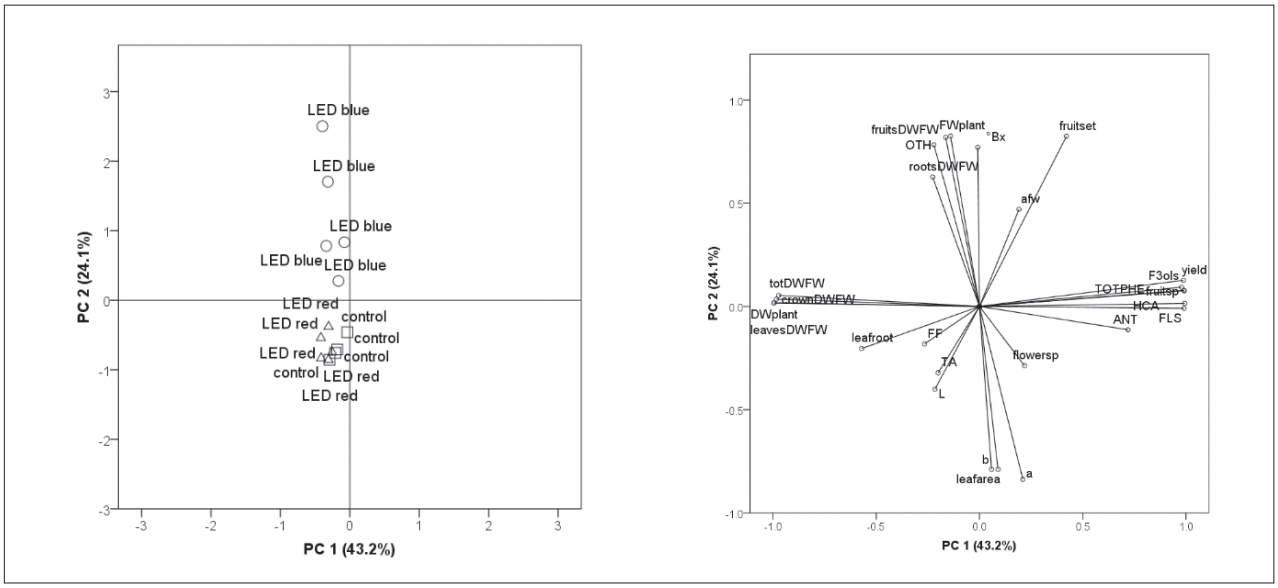

Figure 3. Principal components analysis: score plot (left) and loading plot (right) of the lighting treatments. All variables selected to describe strawberry growth, yield, fruit quality and phenolic composition were considered for the statistical analysis. Legend: yield (total fruit yield per plant); FF (flesh firmness); °Bx (soluble solids content); TA (titratable acidity); L, a, b (colorimetric coordinates); flowersp (number of flowers per plant); fruitsp (number of fruits per plant); fruitset (% fruit set); afw (average fruit weight); FWplant (plant total fresh weight); DWplant (plant total dry weight); totDWFW (DW/FW of the whole plant); rootsDWFW (DW/FW of roots); crownDWFW (DW/FW of crowns); leavesDWFW (DW/FW of leaves); fruitsDWFW (DW/FW of fruits): leafroot (leaf:root weight ratio); leafarea (total leaf area per plant); F3ols (catechin concentration); HCA (p-coumaric acid concentration); ANT (pelargonidin-3-glucoside concentration); FLS (quercetin-3-galactoside concentration); OTH (other phenolic compounds concentration); TOTPHE (total phenolic compounds concentration).

separation along PC 1 (43.2% of the total variance) is less evident. The loading vectors along the PC 2 explain that LED blue samples differ especially because of their high values of plant FW, fruit set, soluble solids content, fruit DW:FW ratio. These values are negatively correlated with total leaf area and a* and b* colorimetric coordinates. Separation between the control and LED red clusters is also possible along the PC 1. Control samples differ especially for the values of variables related to the phenolic compounds concentration. These variables are negatively correlated with the plant DW and with the DW:FW ratio of crowns and leaves. Finally, the PCA of data allowed the separation of blue light treated samples from both the control and the LED red ones, outlining the differential expression of certain yield-related parameters (such as fruit set and the fruit DW/FW ratio) and of quality parameters related to final fruit coloration.

Conclusions

This study addressed the potentiality of single color LED lighting systems for the cultivation of strawberry under protected conditions. It was possible to show that strawberry plants under red and blue LEDs are able to grow and yield fruits of standard quality. Interestingly, the use of blue light was able to cause positive effects on fruit set allowing a significant higher final yield. Blue and red LEDs did not alter the primary fruit quality traits (total sugars content, acidity and pulp consistency), whereas strawberry color and anthocyanins composition resulted partially modified. Based on these results, it appears that the use of blue light can be considered as a potential interesting tool to enhance strawberry yield in protected conditions. Ways of application (blue light alone or in combination with other light sources) and timing (duration and phenophases) must be further investigated in order to define the economical sustainability of this approach and to reach an optimization of the strawberry production management under controlled conditions.

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Addresses of authors:
Stefano Nadalini[1], Paolo Zucchi[2] and Carlo Andreotti[1,*]
[1] Free University of Bozen-Bolzano, Faculty of Science and Technology, Piazza Università 5, 39100 Bolzano-Bozen, Italy
[2] Fondazione Edmund Mach, Technology Transfer Centre, via E. Mach 1, 38010 San Michele all'Adige, Italy
* Corresponding author;
E-mail: carlo.andreotti@unibz.it, Tel.: +39 0471 017122