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Light Spectrum Variably Affects the Acclimatization of Grafted Watermelon Seedlings While Maintaining Fruit Quality

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Abstract: In many countries of Europe and Eastern Asia, watermelon production is mainly based on the use of grafted seedlings. Upon grafting, seedlings undergo a period of healing where artificial lighting is provided by light-emitting diodes in controlled chambers in order to accelerate and improve the healing process. The objective of our study was to test the effect of light quality on the final product (i.e., seedlings ready for transplanting) in the nursery, as well as to evaluate the possible implications on fruit quality after field cultivation. Narrow-band blue (B) and red (R) wavelengths, 64–36% R-B (36B), 76–24% R-B (24B), 88–12% R-B (12B), and 83–12% R-B plus 5% far-red (12B+FR) wavelengths were tested. 12B+FR enhanced the root dry weight, root architecture, and maximum photosynthetic rate, while RB combinations generally showed better root system development with increased blue portion. R light induced inferior root dry weight and quality indices (root/shoot and shoot-dry-weight/length ratios), lower gas exchange parameters, and chlorophyll content, but high shoot length and leaf area. B light led to inferior root architecture, lower stem diameter, leaf area, and maximum photosynthetic rate. Both R and B wavelengths showed decreased concentration of macronutrients and trace elements. After field cultivation, fruit quality (i.e., morphology and color), and valuable nutritive characteristics (i.e., phenolics, carotenoids, lycopene, antioxidants) maintained high quality irrespective of light treatments. Overall, 12B+FR performed well in almost all qualitative parameters including the morphology, the root development, and photosynthesis, while also maintaining high fruit quality.

Keywords: *Citrullus lanatus*; nursery; healing chamber; photomorphogenesis; root architecture; photosynthesis; marketable seedlings; crop production; lycopene; mineral elements



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1. Introduction

Watermelon (*Citrullus lanatus*), popular throughout the world, is a crop with economic and nutritional importance. The main regions of cultivation are located in Eastern Asia (China, South Korea, Japan), the Mediterranean (Greece, Spain, Italy, Turkey), and South America (Brazil) [1]. The watermelon crop is particularly valuable for Greece where the export value in 2020 reached 56 million euros, seventh among all countries in the world [2]. However, watermelon cultivation is subject to various abiotic (i.e., low or high temperatures, heavy metals, etc.) or biotic (i.e., soilborne pathogens, etc.) stresses. Such stress factors can be effectively fixed with vegetable grafting, a propagation technique where plants from different cultivars or even species are conjoined and form a seedling with desired traits [3,4]. Because of grafting, watermelon seedling production takes place in three stages: growth of to-be-grafted seedlings (scion and rootstock; stage I), wound healing upon grafting (stage II), and grafted seedling acclimatization (stage III).

The stage of acclimatization is important in order to develop seedlings with enhanced traits for successful field transplants. This stage is closely related to the second stage (wound healing) where histological modifications occur. Until recently, the healing stage was only performed in benches placed inside a greenhouse where environmental conditions were

insufficiently controlled, and successful healing was achieved after a considerable amount of time. The production of grafted watermelon seedlings is intensified from December to April, thus, for a commercial nursery with limited amount of space the healing time is crucial. Reduced healing time leads to greater meeting of needs for high-quality seedlings from the growers. Currently, healing of grafted seedlings usually takes place in growth chambers that provide defined environments, due to the high sensitivity of the procedure.

Light is provided by artificial light sources such as light-emitting diodes (LEDs) that offer several benefits including optional light quality and intensity, low thermal emission, and long operating lifetime among others [5,6]. Healing and acclimatization are considered the most critical stages in order for a grafted seedling to survive and thrive [7]. Our recent study highlighted the importance of selecting the optimum light wavelength for the rapid and successful healing of grafted watermelon seedlings [8]. However, the stage of acclimatization of watermelon has not yet been examined. Upon acclimatization, seedlings are considered as a “final product” which are purchased by growers for field cultivation. For nurseries involved with watermelon seedlings, a critical aspect of their production apart from high seedling quality is the overall fruit quality that reaches the market and eventually the consumer’s plate [9]. However, the latter is highly dependable on growers’ practices and environmental conditions during cultivation, while the effect of lighting conditions during the seedling production, if any, is not known. Light quality is a factor that imposes morphogenetic responses to plants. In particular, red and blue wavelengths are the most influential for plant growth and development, while far-red has also been shown to affect photomorphogenetic properties along with red wavelength [6].

The objective of our study was to examine the effect of LEDs with varying spectral compositions employed during the healing stage of grafted watermelon seedlings, on seedling acclimatization, and on the final product (i.e., marketable seedlings) of the nursery. Moreover, this study focuses on the potential after-effect of LEDs with different spectral composition on fruit morphology and phytochemical composition in terms of altering or maintaining their quality.

2. Materials and Methods

2.1. Plant Material and Stage I Production

Seeds were provided by HM. Clause SA (Portes-Les-Valence, France). Watermelon (*Citrullus lanatus*) hybrid Celine F1 was the scion segment, while interspecific squash (*Cucurbita maxima* × *C. moschata*) hybrid TZ-148 was the rootstock segment. Plug trays with 171 or 128 cells for scion and rootstock, respectively, were used. The substrate was 5:1:2 mixture of peat, perlite, and vermiculite. The seeds germinated after 72 (watermelon) or 48 h (squash) in a chamber with 95–98% relative humidity and 25 °C temperature. Upon germination, the seedlings to-be-grafted were grown in a plastic greenhouse for 10 days under 21.5 °C minimum temperature. Supplemental lighting (high-pressure sodium lamps, 18 h photoperiod, $100 \pm 10 \mu\text{mol m}^{-2} \text{s}^{-1}$, MASTER GreenPower 600W 400V E40, Philips Lighting, Eindhoven, The Netherlands) was only applied for watermelon scion growth.

2.2. Grafting, Stage II Production, and Light Conditions

Grafting was executed using the splice grafting method, and the root system was removed at the same time, a technique which is often used during the grafting procedure of cucurbits. The grafted seedlings were immediately planted in plug trays with 72 cells and filled with peat, perlite, and vermiculite (3:1:1).

The grafted seedlings were subjected to a healing procedure for seven days inside a healing chamber (89–98% relative humidity, 25 °C temperature, recirculating air) including sole artificial lighting. For the latter, light sources were LEDs with different radiation spectra. Specifically, LEDs emitted narrow-band blue (B; peak wavelength at 450 nm), or red (R; peak wavelength at 661 nm), 36% blue and 88% red (36B), 24% blue and 88% red (24B), 12% blue and 88% red (12B), and 12% blue, 83% red, and 5% far-red (12B+FR; FR peak at 725 nm) with photosynthetic photon flux density (PPFD) of $85 \pm 5 \mu\text{mol m}^{-2} \text{s}^{-1}$.

Spectral distributions of the light treatments are presented in Table 1. All light treatments had a photoperiod of 18 h day/8 h night and were placed at 30 cm above the plants.

Table 1. Wavelength distribution and important photobiological parameters of the light treatments tested. YPFD: yield photon flux density; PPS: phytochrome photostationary state. YPFD and PPS values were calculated according to Sager et al. [10].

Waveband	Light Treatment					
	B	36B	24B	12B	12B+FR	R
UV %; 380–399 nm	0	0	0	0	0	0
Blue %; 400–499 nm	100	36	24	12	12	0
Green %; 500–599 nm	0	0	0	0	0	0
Red %; 600–699 nm	0	64	76	88	83	100
Far-red %; 700–780 nm	0	0	0	0	5	0
YPFD ($\mu\text{mol m}^{-2} \text{s}^{-1}$)	63.8	73.5	75.4	77.2	73.9	79.1
PPS	0.51	0.88	0.89	0.89	0.88	0.89

2.3. Grafted Seedlings' Acclimatization in Stage III

Upon exiting the healing chamber, the trays containing seedlings were placed in a greenhouse for 7 days of acclimatization until they were considered commercial product. Minimum temperature was 21.5 °C, and HPS lamps were used for supplemental lighting (18 h day/8 h night photoperiod, PPF of $100 \pm 10 \mu\text{mol m}^{-2} \text{s}^{-1}$). For fertilization, permanent NPK (7:3:7) was included in the substrate along with Mg and trace elements (Osmocote Exact Mini, Geldermalsen, The Netherlands).

2.4. Determinations after Acclimatization

Upon visual examination by experienced personnel, it was determined that seedlings have reached the minimum marketable size after seven days of acclimatization in the greenhouse. At that time, 36 seedlings per treatment were sampled and evaluated as follows.

A digital caliper was used for shoot length and stem diameter determination. Dry weight of shoots and root systems were measured after 3 days at 72 °C. Moreover, two quality indices, the root-to-shoot ratio (R/S) and shoot dry weight-to-length ratio (DW/L), were also calculated from the values above. Leaf area was measured with LI-3000C (LI-COR biosciences, Lincoln, USA). Relative chlorophyll content was determined with a chlorophyll meter (CCM-200 plus, Opti-Sciences, Hudson, NH, USA). Parameters of the root architecture such as surface area, total length, tip number, and volume were measured in 5 seedlings per treatment with WinRHIZO Pro software (Regent Instruments Inc., Québec, QC, Canada). WinRHIZO software has been suggested [11] as a valuable tool for the detailed examination of the root system with view to understanding its manner of development. Maximum quantum yield of primary photochemistry of dark-adapted leaves (Fv/Fm) was determined with a Pocket-PEA fluorometer (Pocket-PEA, Hansatech Instruments, Norflock, UK). Gas exchange parameters (stomatal conductance (gs), maximum photosynthetic rate (Pmax), and transpiration rate (E)) were measured with a Li-6400XT (LI-COR biosciences, Lincoln, NE, USA) set at a CO₂ concentration of 400 ppm, temperature of 25 ± 1 °C, and $500 \mu\text{mol m}^{-2} \text{s}^{-1}$ PPF. A CR-400 Chroma Meter (Konica Minolta Inc., Tokyo, Japan) was used for leaf color measurements.

After seven days in the greenhouse, 120 seedlings (3 replications \times 40 seedlings) per light treatment were sampled, dried, and ground for nutrient content determination. Nitrogen was determined using the Kjeldahl method [12], by which samples were digested with H₂SO₄, distilled in boric acid (H₃BO₃), and titrated with hydrochloric acid (HCl). Phosphorus, potassium, calcium, magnesium, sodium, iron, zinc, manganese, and copper were extracted using HCl and determined by an inductively coupled plasma mass spectrometer (ICP-MS, ICAP6300, Thermo Scientific, Waltham, MA, USA). Boron was extracted using HCl and determined by a UV-VIS spectrophotometer (Shimadzu Scientific Instruments, Columbia, MD, USA) at 414 nm.

2.5. Field Cultivation

Upon acclimatization, seedlings were transplanted and cultivated for over two months until fruit harvest. The cultivation was conducted in 2018 and 2019, in a commercial farm at Vathylakkos, Thessaloniki, Greece (N 40.761; E 22.724). Both cultivations showed similar conclusions; thus, data from 2019 are presented. Data from 2018 are also presented in Supplementary Table S1. Transplantation took place on May 5th, peak flowering was approximately on May 31st, and fruits were harvested on July 10th. The soil was prepared prior to transplantation with usual plowing, crumbling, dry poultry manure incorporation, and irrigation practices. The soil was characterized as sandy clay loam (SCL); moderate to heavy type with moderate pH, low CaCO_3 , moderate organic matter content, and low salinity. Irrigation, fertilization, and weed and pathogen control were conducted according to normal local practices.

Fifteen plants from every light treatment were transplanted. Plant distance between and within rows were 3 and 1 m, respectively. Plants were arranged in a randomized complete block design (RCBD) with three replicates. Specifically, plants were transplanted in three rows containing one group of plants from every light treatment, and each group consisted of 5 plants in a row.

2.6. Determinations after Field Cultivation

After field cultivation and harvest, a number of quality parameters were measured in order to examine the possible after-effect of light wavelengths imposed during the healing stage, and to determine the fruit status after cultivation in the field. After about 40 days from peak flowering, fully developed fruits free of defects or symptoms from the middle plant (third plant) from each row and treatment were harvested for morphological and quality determinations. Specifically, upon weighting in order to have similar size, fruits were cut longitudinally and length, width, as well as rind thickness were measured with a ruler. Colorimetric parameters were obtained from the fruit center. Total soluble solids ($^\circ\text{Brix}$) were determined using a refractometer (PAL- α , Atago, Tokyo, Japan).

Total phenolic content and ferric reducing antioxidant power (FRAP) were determined in homogenized 5 g samples obtained from the whole fruit and extracted with 25 mL of 80% aqueous methanol. Total phenolic content was determined according to Singleton and Rossi [13], where 0.5 mL of methanolic plant extract, 2.5 mL of Folin–Ciocalteu's reagent, and 2 mL of 7.5% sodium carbonate solution were incubated for 5 min at 50 °C. Absorbance of the colored product was measured at 760 nm (Shimadzu Scientific Instruments, Columbia, MD, USA) versus a blank consisting of 0.5 mL of 80% aqueous methanol instead of methanolic plant extract. The results were expressed as mg of gallic acid equivalent per gram fresh weight.

FRAP was determined according to Benzie and Strain [14] where 0.1 mL of methanolic plant extract was added to 3 mL of working solution (CH_3COONa buffer solution, pH 3.6; TPTZ and FeCl_3) and incubated at 37 °C for exactly 4 min. Absorbance of the colored product was measured at 593 nm versus a blank consisted of 0.1 mL of 80% aqueous methanol. The results were expressed as μg of ascorbic acid equivalent per gram fresh weight.

Total carotenoid content and lycopene content were determined in homogenized 1-g samples obtained from the whole fruit and extracted with 25 mL of 80% aqueous ethanol. Absorbance of the colored product was measured at 445 and 503 nm versus a blank consisting of 80% aqueous ethanol. The results were expressed as μg of ascorbic acid equivalent per gram fresh weight.

2.7. Statistical Analysis

Statistical analysis was performed using IBM SPSS software (SPSS 23.0, IBM Corp.). Data were compared by one-way and two-way analysis of variance (ANOVA) at significance level $p = 0.05$, while mean comparisons were conducted using Tukey test at $\alpha = 0.05$.

3. Results and Discussion

3.1. Grafted Seedlings' Acclimatization

Figure 1 depicts acclimated grafted watermelon seedlings treated with the six different wavelengths during healing. Red and blue wavelengths are known to drive photosynthesis in the most efficient manner, subsequently leading to greater biomass production and accumulation [15]. In our study, shoot dry weight was significantly greater in 24B compared to 12B (Figure 2A). 12B+FR induced greater root dry weight compared to the rest of the LEDs, while 36B and 24B had greater values compared to B, R, and 12B (Figure 2B). A healthy and vastly developed root system is critical for the rapid and efficient establishment of seedlings after transplantation. Total root system was significantly longer under 12B+FR, R, and 36B, compared to 12B (Figure 2C). The root surface area which is the area of roots in contact with the soil, as well as the number of tips, were also enhanced under 12B+FR compared to 12B and B (Figure 2D,E), while 12B+FR and R induced significantly greater root volume than B and 12B (Figure 2F). Overall, it is evident that narrow-band R wavelength and bichromatic treatments with relatively high blue amount (36B and 24B) or with additional FR wavelength (12B+FR) enhanced important parameters of the root system architecture leading to rapid lateral roots formation. Red and far-red wavelengths are known to be related with the root system development due to their involvement with auxin distribution from the apical bud to the roots [16]. In a study with artichoke, the authors reported longer root formation when seedlings were treated with red light compared to blue wavelength [17].

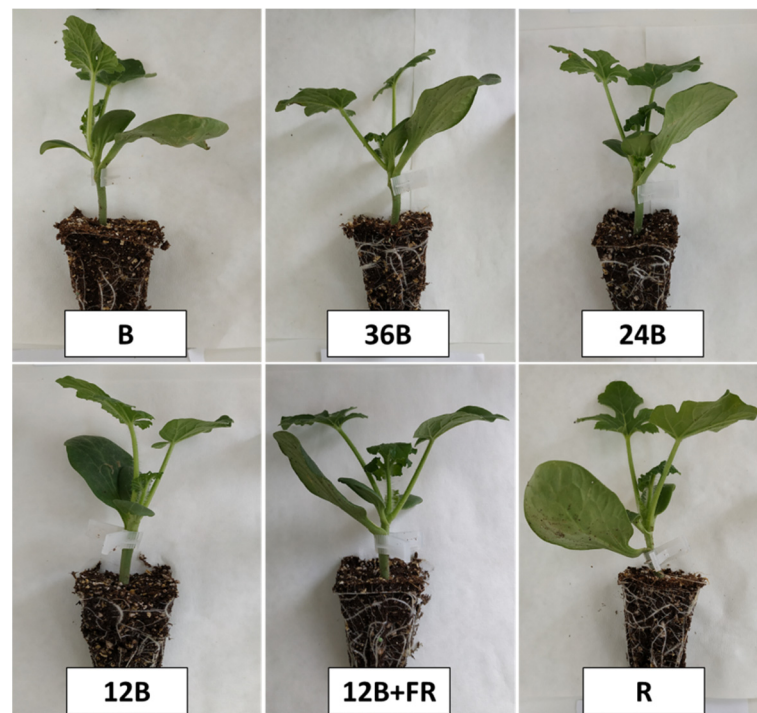


Figure 1. Grafted watermelon seedlings after healing in a chamber under six different light treatments for 7 days, and after acclimatization in a greenhouse for 7 days. B: narrow-band blue; 36B: 36% blue and 64% red; 24B: 24% blue and 76% red; 12B: 12% blue and 88% red; 12B+FR: 12% blue, 83% red, and 5% far-red; R: narrow-band red.

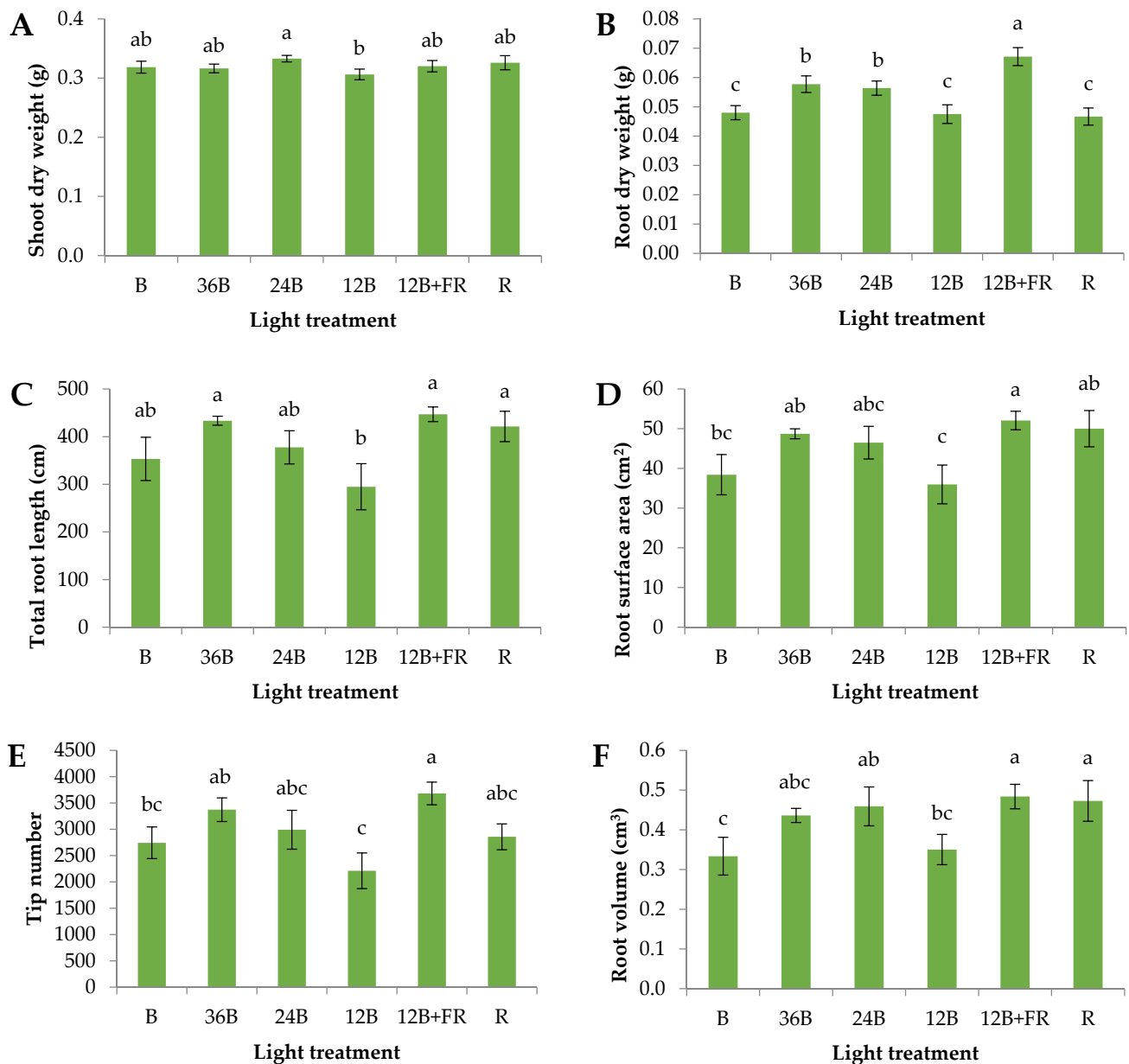


Figure 2. (A) Shoot dry weight, (B) root dry weight, and root architecture parameters such as (C) total root length, (D) root surface area, (E) tip number, and (F) root volume of grafted watermelon seedlings treated for seven days in a healing chamber with six light treatments, plus seven days in a greenhouse. Mean values (\pm SE) within a row followed by different letters are significantly different ($\alpha < 0.05$).

12B+FR also enhanced the R/S ratio compared to the other light treatments (Table 2). The increased R/S ratio is associated with better transplantation success due to the enhanced potential of a more expanded root system to absorb water and nutrients [18]. In addition, DW/L has been described as an essential quality parameter for seedlings of important vegetables such as tomato and grafted watermelon [1,19]. Higher values of the parameter are related to better quality; thus, it is a useful tool for fast and efficient seedling assessment before transplantation in the field. In this study, R led to significantly lower DW/L compared to the rest of the LEDs (Table 2) leading to the conclusion that narrow-band R is inefficient for the production of high-quality grafted watermelon seedlings.

Table 2. Developmental, physiological, and colorimetric parameters of grafted watermelon seedlings treated for seven days in a healing chamber with six light treatments, plus seven days in a greenhouse. R/S: root-to-shoot ratio; DW/L: shoot dry weight-to-length ratio; chl: chlorophyll; Fv/Fm: maximum quantum yield of primary photochemistry of a dark-adapted leaf. Mean values (\pm SE) within a parameter followed by different letters are significantly different ($\alpha < 0.05$).

Parameters	Light Treatments					
	B	36B	24B	12B	12B+FR	R
R/S ratio	0.15 \pm 0.01c	0.17 \pm 0.01b	0.17 \pm 0.01b	0.16 \pm 0.01bc	0.21 \pm 0.01a	0.13 \pm 0.01d
(DW/L) *1000	4.80 \pm 0.14a	5.01 \pm 0.13a	4.86 \pm 0.10a	4.93 \pm 0.12a	4.78 \pm 0.12a	4.55 \pm 0.14b
Chl. content	32.45 \pm 1.22a	31.09 \pm 1.09a	29.18 \pm 1.31ab	29.16 \pm 1.18ab	28.07 \pm 1.24ab	26.16 \pm 1.01b
Fv/Fm	0.822 \pm 0.002a	0.823 \pm 0.002a	0.823 \pm 0.002a	0.825 \pm 0.002a	0.823 \pm 0.002a	0.822 \pm 0.003a
Lightness	40.33 \pm 0.34a	40.40 \pm 0.43a	39.72 \pm 0.28a	39.43 \pm 0.31a	40.53 \pm 0.34a	40.31 \pm 0.27a
Chroma	21.94 \pm 0.39a	23.75 \pm 0.59a	23.25 \pm 0.43a	23.41 \pm 0.44a	23.88 \pm 0.57a	23.37 \pm 0.36a
Hue angle	129.58 \pm 0.23a	129.18 \pm 0.32a	129.26 \pm 0.21a	129.31 \pm 0.22a	129.18 \pm 0.30a	128.85 \pm 0.19a

Chlorophylls are probably the most important molecules related to plant life and development [20]. Chlorophylls a and b absorb light energy in the form of red and blue wavelengths, both of which are crucial for photosynthesis. In the present study, R led to significantly lower relative chlorophyll content than B and 36B (Table 2). Similar to our results, basil, pepper, spinach, and lettuce produced greater amounts of chlorophylls when treated with 91%/9% red/blue compared to narrow-band red light [21], proving the importance of blue light in chlorophyll regulation which is accomplished through the involvement of phot I and phot II phototropins [22].

Fv/Fm showed no differences among light treatments (Table 2), even though all means laid within the reported limits (0.78–0.86) for healthy seedlings with efficient photosynthetic apparatus [23]. This result proves that, among the tested light treatments, no damaging effect remains on seedlings until after the stage of acclimatization regardless of the light spectra imposed during the healing stage.

Color is usually a quality indicator in most agricultural products [24]. However, in our case, colorimetric parameters such as leaf lightness, chroma, and hue angle were not significantly affected by the different light treatments (Table 2). Initially, we expected a decrease in lightness under R treatment which also caused a significant drop in chlorophyll content. The two parameters are linearly related [25] but this was not evident in the grafted watermelon seedlings of our study.

After acclimatization, seedlings treated with R had a significantly greater shoot length compared to 36B and 12B (Figure 3A), while leaves were significantly more expanded in R, 36B, and 24B compared to B (Figure 3B). These two important quality indicators, shoot elongation and leaf expansion, are regulated by red and far-red wavelengths through phytochromes [26]; thus, they can be controlled depending on the provided light. This photoreceptor is related to shade avoidance responses as an answer to different red/far-red ratios of incident light. Similarly, Brassica microgreens (cabbage, mustard, arugula, and kale) formed larger leaves under narrow-band red than blue wavelength [27]. In a study with cucumber, red-blue treatments with decreasing blue portion led to longer shoots and more expanded leaves [28]. A valuable quality index of grafted watermelon seedlings, stem diameter, was significantly greater in 36B than B light treatment (Figure 3C). Similar to our results, Li et al. [29] reported greater stem diameter in tomato seedlings treated with narrow-band red or red-blue wavelengths compared to monochromatic blue light.

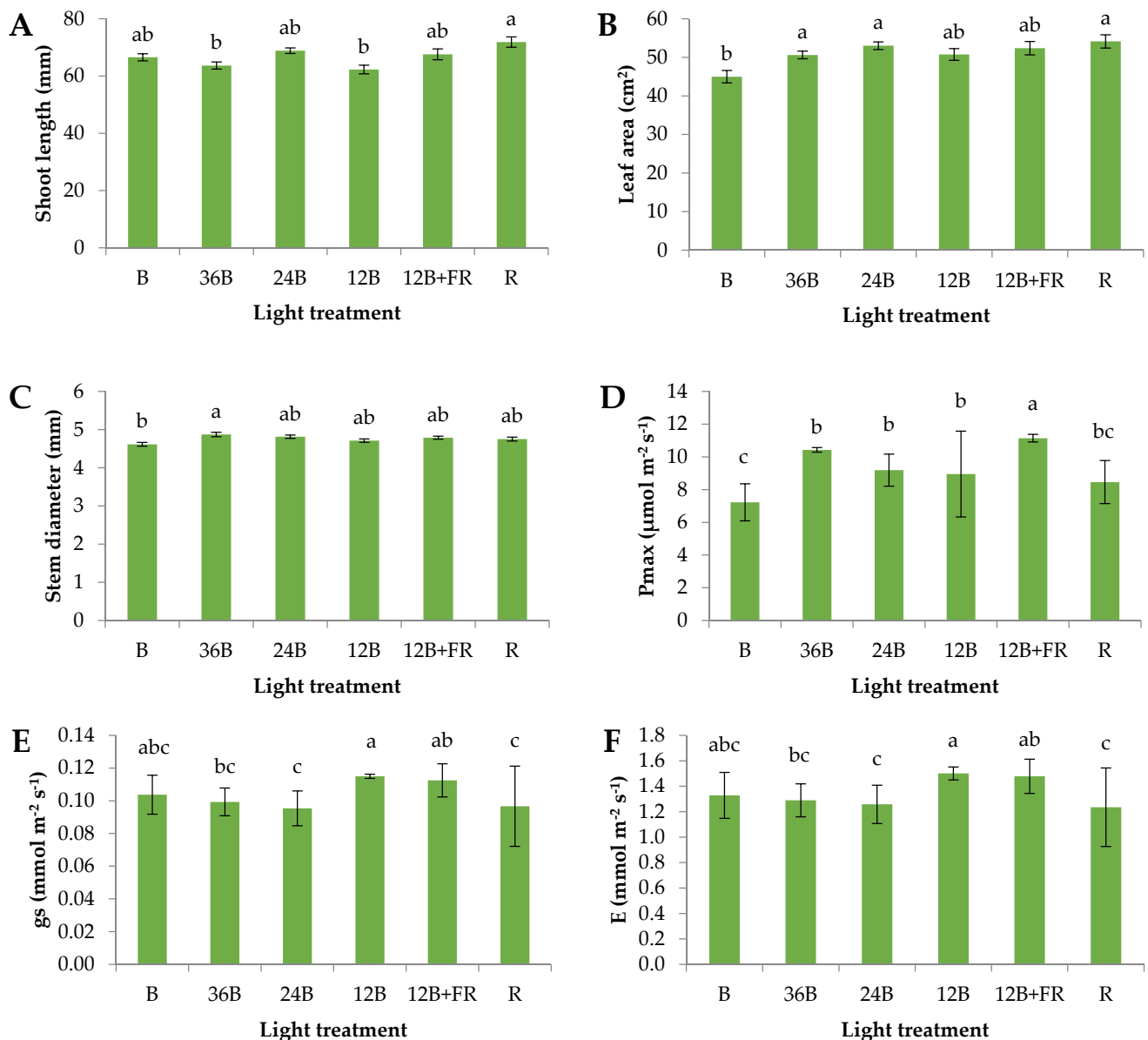


Figure 3. (A) Shoot length, (B) leaf area, (C) stem diameter, and gas exchange parameters such as (D) maximum photosynthetic rate, (E) stomatal conductance, and (F) transpiration rate of grafted watermelon seedlings treated for seven days in a healing chamber with six light treatments, plus seven days in a greenhouse. Pmax: maximum photosynthetic rate; gs: stomatal conductance; E: transpiration rate. Mean values (\pm SE) within a row followed by different letters are significantly different ($\alpha < 0.05$).

Pmax was enhanced under 12B+FR compared to the rest of the light treatments. Moreover, B showed the lowest values compared to 36B, 24B, and 12B (Figure 3D). Regarding gs and E, 12B had significantly greater values compared to 36B, 24B, and R (Figure 3E,F). Seedlings of another important cucurbit, cucumber, exhibited lower photosynthetic rate as well as indications of problematic photosynthetic apparatus under narrow-band red light [30]. In another study by our group, we found that Pmax, gs, and E of grafted watermelon seedlings immediately after exiting the healing chamber were inferior under R compared to bichromatic light treatments, especially 12B [31]. Narrow-band red light triggers the activity of phytochromes, but not other photoreceptors [32], suggesting a reduced potential to develop an efficient photosynthetic apparatus under solo red light. However, the addition of blue light (i.e., multichromatic light spectra) is known to reduce these dam-

aging effects [33]. Furthermore, Muneer et al. [34] reported that blue light enhances g_s and E in plant leaves. Here, R had rather low g_s values which led to lower E and subsequently reduced P_{max} . However, P_{max} is not always directly correlated to g_s and E as shown by B and 12B in this study.

Plants require the essential nutrient elements (macronutrients and trace elements) for normal development and morphogenesis [35]. These elements are absorbed by the root system and traverse the plant matrix of apoplastic and symplastic pathways until reaching their destination, the leaves, flowers, fruits, etc. In general, plants require a sufficient supply of macronutrients and trace elements to thrive and achieve maximum productivity [36]. Their nutrient content can efficiently be controlled by growth conditions including light quality. At the end of acclimatization, total nitrogen (N) was significantly greater in B and R compared to 36B, phosphorus (P) was enhanced in 36B, 24B, and 12B compared to B, R, and 12B+FR, while potassium (K) was greater under 36B and 12B compared to B, R, and 12B+FR (Table 3). Calcium (Ca) was significantly greater in 36B and 12B than R, magnesium (Mg) was enhanced under 36B than B, while sodium (Na) was greater in 36B and 24B than R (Table 3). Boron (B) was not significantly affected by the light treatments, while manganese (Mn) was greater under 12B+FR compared to R (Table 3). Zinc (Zn) and iron (Fe) were significantly enhanced under 12B+FR compared to the rest of the LEDs, while copper (Cu) was greater in 12B compared to B, R, and 12B+FR (Table 3). Overall, narrow-band R and B wavelengths showed decreased concentration of essential macronutrients and trace elements compared to the rest of the multichromatic light treatments. Nevertheless, blue light has been shown to regulate processes such as proton pumping and membrane permeability which are involved with the accumulation of nutrients in broccoli sprouts [37]. In addition, Gerovac et al. [38], working with kohlrabi, mizuna, and mustard microgreens reported a tendency (often significant) for increased macronutrients or trace elements under 74%/18%/8% red/green/blue compared with 74%/7%/9% red/far-red/blue or 87%/13% red/blue, respectively.

Table 3. Nutrient content of grafted watermelon seedlings treated for seven days in the healing chamber with six light treatments, plus seven days in the greenhouse. Mean values (\pm SE) within a nutrient followed by different letters are significantly different ($\alpha < 0.05$).

Parameters	Light Treatments					
	B	36B	24B	12B	12B+FR	R
Total N %	3.56 \pm 0.24a	3.12 \pm 0.05b	3.33 \pm 0.03ab	3.28 \pm 0.10ab	3.39 \pm 0.01ab	3.55 \pm 0.12a
P %	2.45 \pm 0.04b	2.84 \pm 0.04a	2.82 \pm 0.03a	2.83 \pm 0.06a	2.59 \pm 0.06b	2.48 \pm 0.14b
K %	5.29 \pm 0.10bc	5.80 \pm 0.15a	5.64 \pm 0.13ab	5.87 \pm 0.04a	5.32 \pm 0.12bc	4.96 \pm 0.25c
Ca %	2.72 \pm 0.02ab	2.84 \pm 0.12a	2.65 \pm 0.09ab	2.83 \pm 0.10a	2.77 \pm 0.08ab	2.50 \pm 0.11b
Mg %	0.95 \pm 0.02b	1.12 \pm 0.08a	1.01 \pm 0.06ab	1.01 \pm 0.05ab	1.03 \pm 0.02ab	1.00 \pm 0.01ab
Na %	0.05 \pm 0.01ab	0.06 \pm 0.01a	0.06 \pm 0.01a	0.06 \pm 0.01ab	0.05 \pm 0.01ab	0.05 \pm 0.01b
B ppm	51.86 \pm 1.44a	53.44 \pm 1.45a	53.59 \pm 1.78a	52.88 \pm 0.92a	51.77 \pm 1.24a	51.01 \pm 1.11a
Mn ppm	46.47 \pm 1.17ab	47.90 \pm 2.62ab	48.11 \pm 2.10ab	47.63 \pm 2.18ab	50.94 \pm 2.57a	44.50 \pm 1.17b
Zn ppm	74.74 \pm 1.50c	78.93 \pm 2.16bc	80.18 \pm 0.63bc	81.39 \pm 1.00b	89.99 \pm 0.36a	80.68 \pm 3.47b
Fe ppm	108.63 \pm 1.41c	115.37 \pm 1.01b	114.63 \pm 1.44b	115.47 \pm 2.16b	120.60 \pm 1.81a	109.43 \pm 1.28c
Cu ppm	15.45 \pm 0.75c	17.00 \pm 0.51abc	17.38 \pm 0.23ab	17.44 \pm 0.44a	15.73 \pm 0.38bc	15.28 \pm 0.81c

3.2. Field Cultivation

Fruits produced after field cultivation showed similar morphological, phytochemical, and colorimetric characteristics regardless of the light treatment that the seedlings were exposed to during healing. Specifically, morphological parameters such as fruit length, width, rind thickness, phytochemicals such as total soluble solids, total phenolic content, total carotenoid content, lycopene content, antioxidant capacity (displayed by FRAP), and colorimetric parameters such as lightness, chroma, and hue angle were not affected by the various light treatments during healing of the respective grafted watermelon seedlings (Table 4 for 2019 cultivation; Supplementary Table S1 for 2018 cultivation).

Table 4. Morphological, phytochemical, and colorimetric parameters of ripe watermelon fruits after field cultivation in 2019. The transplanted grafted seedlings were treated with six different light treatments during the healing stage. TSS: total soluble solids; TPC: total phenolic content; TCC: total carotenoid content; LC: lycopene content; FRAP: ferric reducing antioxidant power. Color was measured in the center of the fruit. Mean values (\pm SE) within a parameter followed by different letters are significantly different ($\alpha < 0.05$).

Parameters	Light Treatments					
	B	36B	24B	12B	12B+FR	R
Length (cm)	29.63 \pm 2.51a	32.00 \pm 1.61a	31.97 \pm 1.73a	30.97 \pm 2.29a	33.30 \pm 0.85a	31.83 \pm 0.68a
Width (cm)	21.33 \pm 1.17a	22.17 \pm 0.34a	21.77 \pm 0.90a	22.13 \pm 0.66a	23.37 \pm 0.13a	22.43 \pm 0.23a
Rind thick. (cm)	1.82 \pm 0.12a	1.58 \pm 0.12a	1.85 \pm 0.21a	1.78 \pm 0.13a	1.85 \pm 0.07a	1.65 \pm 0.17a
TSS ($^{\circ}$ Brix)	10.73 \pm 1.07a	10.17 \pm 0.57a	10.07 \pm 0.20a	10.03 \pm 0.24a	10.80 \pm 0.44a	9.93 \pm 0.28a
TPC (mg/g)	0.13 \pm 0.01a	0.14 \pm 0.02a	0.12 \pm 0.01a	0.14 \pm 0.01a	0.14 \pm 0.01a	0.15 \pm 0.02a
TCC (μ g/g)	35.09 \pm 1.44a	36.67 \pm 5.23a	36.79 \pm 1.83a	42.63 \pm 5.22a	42.42 \pm 2.52a	45.89 \pm 8.56a
LC (μ g/g)	33.81 \pm 1.45a	35.31 \pm 5.15a	35.60 \pm 1.87a	41.15 \pm 4.99a	40.92 \pm 2.14a	44.06 \pm 7.98a
FRAP (μ g/g)	35.52 \pm 3.20a	38.82 \pm 6.20a	35.18 \pm 2.59a	37.04 \pm 4.22a	40.36 \pm 0.93a	35.63 \pm 2.79a
Lightness	40.84 \pm 2.67a	40.76 \pm 2.49a	41.55 \pm 0.43a	41.03 \pm 3.10a	39.10 \pm 3.47a	42.00 \pm 1.12a
Chroma	27.48 \pm 0.18a	30.25 \pm 0.51a	30.90 \pm 1.32a	31.46 \pm 1.26a	33.46 \pm 1.64a	31.02 \pm 3.16a
Hue angle	34.82 \pm 1.51a	33.00 \pm 1.29a	33.22 \pm 0.12a	32.73 \pm 1.17a	32.84 \pm 0.42a	33.51 \pm 0.92a

Lycopene is a valuable carotenoid with great health benefits for humans, having a positive role in prostate cancer [39]. Watermelon is one of the most important sources of lycopene for humans, along with tomato. In fact, watermelon lycopene has been found to have considerably higher antioxidant activity compared to tomato lycopene [40]. In our case, different light wavelengths did not influence the biosynthesis and accumulation of lycopene, thus maintaining fruit quality. It is not surprising that light treatments during the healing stage did not affect fruit quality since different wavelengths were only imposed for 7 days, almost three months before fruit harvesting. The conclusion drawn from this part of the experiment was that different light wavelengths during healing of grafted watermelon seedlings do not alter the fruit quality in terms of morphology, phytochemical content, or color.

4. Conclusions

Nurseries involved with the development of grafted watermelon seedlings mainly focus on the production of high-quality final product-seedlings with high transplantation success and rapid growth upon acclimatization in the nursery. In our study, 12B+FR enhanced the root system development (dry weight and root architecture) and the maximum photosynthetic rate. Red-blue combinations showed variable responses with generally better root system development with increasing blue portion. Seedlings treated with narrow-band R light showed inferior root dry weight, and subsequent R/S ratio and DW/L, considerably lower gas exchange parameters and chlorophyll content, but high shoot length and leaf area. Narrow-band B induced the formation of inferior root system displayed by root architecture analysis, and lower stem diameter, leaf area, and maximum photosynthetic rate. Both R and B wavelengths showed a decreased concentration of macronutrients and trace elements compared to the rest of the light treatments. Furthermore, nurseries are also concerned about the overall fruit quality that reaches the market and eventually the consumers' plate. After field cultivation and fruit quality determination, it is concluded that different light wavelengths during the healing stage do not alter the morphology, phytochemical content, or color of fruits, thus maintaining their quality at high levels. Different light wavelengths were imposed only during healing of grafted seedlings, while fruits were harvested almost three months later. The differences observed after acclimatization were lost until harvest.

Supplementary Materials: The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/horticulturae8010010/s1>, Table S1: Morphological, phytochemical, and colorimetric parameters of ripe watermelon fruits after field cultivation in 2018. The transplanted grafted seedlings were treated with six different light treatments during the healing stage. TSS: total soluble solids; TPC: total phenolic content; TCC: total carotenoid content; LC: lycopene content; FRAP: ferric reducing antioxidant power. Color was measured in the center of the fruit. Mean values (\pm SE) within a parameter followed by different letters are significantly different ($\alpha < 0.05$).

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