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Effect of postharvest visible spectrum LED lighting on quality and bioactive compounds of tomatoes during shelf life

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ABSTRACT

The objective was to evaluate the effect of visible spectrum LED lighting during shelf-life on physicochemical quality and the main bioactive compounds of Kumato® cherry tomatoes. Tomatoes were stored 13 days at 5 °C under white (W), blue (B), blue + red (B + R), green + red (G + R), and green + far-red (G + FR) LED lights. Darkness (D) was used as control. Tomatoes under illumination showed higher weight losses and firmness decreases (30–35%). No chilling injury was observed. B + R lighting increased the carotenoid content by ~27%, while G + R and G + FR reported an increase ~30% in phenolics. B and B + R showed the highest increase in the phytochemical biosynthesis (lycopene and naringenin, as main carotenoid and flavonoid found) compared to D and W. Conclusively, illumination with B + R or B during shelf-life is recommended to enhance the main bioactive compounds. G + R and G + FR, also reported to be good elicitors of the phenolics and carotenoids biosynthesis.

1. Introduction

Tomato (*Solanum lycopersicum*) is one of the most widespread and economically valuable vegetables worldwide, being the production in 2020 of 183 million tonnes (FAO, 2022). In recent years, several breeding programmes have been carried out to extend the shelf-life of tomatoes (Panjai, Noga, Hunsche, & Fiebig, 2019).

Light, in the range of photosynthetically active radiation (PAR), plays several roles in plant life, since it is a source of energy for carbohydrate synthesis, acts as a morphogenic signal to regulate several processes, and stimulates a number of light-dependent reactions (Berkovich et al., 2017). In fact, the type and quantity of available radiation influences many physiological, morphogenetic and reproductive processes of plants, being also the main driver for the production of secondary metabolites (Zhang, Bian, Yuan, Chen, & Lu, 2020). Light Emitting Diode (LED)-based lighting technology has expanded the possibilities to analyse the effects of lighting parameters on physiological processes in plants and to explore the effects of light spectral quality on metabolism and photo-oxidative processes (Rodríguez, 2019). Blue LED light (67 W m^{-2}) has been applied to tomato plants increasing their total dry weight (Xiaoying, Shirong, Taotao, Zhigang, & Tezuka, 2012) and even blue, red, and far-red (6.5, 5, and 1.5 W m^{-2}, respectively) LED lighting during cultivation has had positive effects on harvested fruit (Duchovskis and Samuoliene, 2010).

The effect of LED lights on tomato storage is being actively studied. Ngcobo, Bertling, and Clulow (2020) recently reported that application of LED lights on immature tomato alters its colour and phytochemical composition. Although it has been reported that tomatoes stored below 13 °C may suffer from chilling injury (Rai, Kumari, & Vashistha, 2022), a previous work has shown that UV or even LED lighting prior to tomato storage can reduce or even prevent the occurrence of chilling injury (Baenas et al., 2021).

Parallelly, in the Solanaceae family, Martínez-Zamora, Castillejo, and Artés-Hernández (2021) demonstrated that the application of a photoperiod of 14 h of fluorescent illumination + 10 h under blue (450 nm) and red (660 nm) LEDs (576 kJ m^{-2}) for 4 days at 20 °C in red bell peppers resulted in an increased carotenoids accumulation (33%) during shelf-life when compared to other photoperiods conventionally used in supermarkets.

Specifically in tomatoes, Liu, Zabaras, Bennett, Aguas, and Woonton (2009) showed that a daily 24.3 kJ m^{-2} red light treatment enhances lycopene accumulation with minimum effects on the colour, hardness, or total soluble solids during a postharvest storage of 21 days at 13 °C, which indicates that red light is a regulator of carotenoid synthesis and

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accumulation. Also, Nájera, Guil-Guerrero, Enríquez, Álvaro, and Urrestarazu (2018) have shown as higher proportions of red zones (red and far red) of the visible spectrum in postharvest LED lighting induced lycopene synthesis by 41% compared to darkness and by 24% relative to other LED lighting conditions for 6 days at 19 °C. Similar results were shown by Panjai et al. (2019) after a continuous 113 μmolm^{-2} s^{-1} red illumination for 14–20 days at 20 °C regarding the biosynthesis and accumulation of lycopene and β-carotene. Furthermore, the daily exposition for 6 or 12 h day^{-1} to the same red illumination showed comparable results with regards to the total phenolic and flavonoid content, as well as its hydrophilic antioxidant capacity.

The main objective of this work was to evaluate the effect induced by a continuous postharvest illumination with visible spectrum LED lights on the main quality attributes and bioactive compounds of cherry tomatoes during a refrigerated shelf-life period. Therefore, we would be able to elucidate if combining different regions of the visible light spectrum are able to induce changes in the secondary metabolism, which may result in the synthesis of bioactive antioxidant compounds, such as phenolic acids, flavonoids, and carotenoids, with potential health benefits.

2. Material & methods

2.1. Plant material

Kumato® cherry tomatoes (*Solanum lycopersicum*) were harvested and supplied by G's España Holdings SL (Torre-Pacheco, Murcia, Spain). The KM5512 variety from Syngenta seeds (Torre-Pacheco, Murcia, Spain) was used in the experiment. The main quality attributes of 50 representative fruits were monitored (Table 1). Fruits with a maturity index of 7.03 ± 0.4 were selected for the experiment and were visually assessed to ensure the absence of any visible damage.

2.2. Postharvest handling

Ten kg of tomatoes were disinfected with sodium hypochlorite (1 min; 100 ppm; pH = 6.5; 5 °C) and then rinsed 1 min. Once dewatered and superficially dried with filter paper, they were separately placed on caps to avoid overlapping, and they were packed inside TR-750 mL trays (Sena S.A., Guipúzcoa, Spain) which were thermally sealed (BP 40/E, Beford technitrans, Saint-Clément, France) with a 25 μm macro-perforated oriented polypropylene film (Plásticos del Segura, Murcia, Spain) to avoid weight loss. The average pore size of the film was 8.6 ± 0.4 mm and the cadence was 32 ± 0.6 mm. The average weight of the trays containing the fruits was 102.5 ± 7.2 g.

2.3. Lighting treatments and storage conditions

Four continuous lighting treatments were assayed in 30 × 60 × 60 cm (width × length × height) self-made metal containers inside a cold room, where the samples were approximately placed 20 cm from the light source, although such distance was slightly modified to reach the required intensities to be similar in all treatments (Fig. 1).

- Darkness (**D**): no light was applied. It was used as control.
- White (**W**): fruits were disposed under white LED lighting (full spectrum) with an intensity of 20 W m^{-2}, as it is conventionally used for storage in the food industry or supermarkets.
- Blue (**B**): fruits were disposed under blue LED lighting with an intensity of 21.5 W m^{-2}.
- Blue + Red (**B + R**): fruits were disposed under a red and blue LED lighting combination with an intensity of 10.9 W m^{-2} and 11.3 W m^{-2}, respectively, with a total intensity of 22.2 W m^{-2}.
- Green + Red (**G + R**): fruits were disposed under a green and red LED lighting combination with an intensity of 11 W m^{-2} and 10.2 W m^{-2}, respectively, with a total intensity of 21.2 W m^{-2}.
- Green + Far-Red (**G + FR**): fruits were disposed under a green and far-red LED lighting combination with an intensity of 11 W m^{-2} and 11.3 W m^{-2}, respectively, with a total intensity of 22.3 W m^{-2}.

Regarding LED treatments applied, B and R have been described as the most studied elicitors from the visible light spectrum (Kong, Zhao, Ma, Liang, & Zhao, 2021; Ntagkas et al., 2020). For that reason, single B was applied while R was combined to different regions of the visible light spectrum. The selection of these treatments was based on our previous results in bell pepper (Martínez-Zamora, Castillejo, & Artés-Hernández, 2021) and those obtained by Panjai et al. (2019) and Baenas et al. (2021), who demonstrated that red lighting is useful to enhance the accumulation and synthesis of carotenoid compounds in tomatoes, while avoiding the chilling injuries during refrigerated storage.

The trays within containers were kept under illumination treatments in a 5 m^3 cold room at 5 °C and a relative humidity of 85%. The tomatoes were placed with the stem scar on one side inside the trays. LED lights were purchased from LEDMurcia S.L. (Murcia, Spain). The wavelengths used were: Red (R; peak at 630 nm), Blue (B; peak at 465 nm), Green (G; peak at 525 nm), and Far Red (FR; peak at 670 nm). The LEDs were placed on top of the containers illuminating 24 h d^{-1} during the whole shelf-life period. A constant Photosynthetic Photon Flux Density for W, B, B + R, G + R, and G + FR of 96, 103.5, 106.6, 101.8, and 107 μmol m^{-2} s^{-1}, respectively, was measured using a DO 9721 quantum photoresist data logger (Delta Ohm, SRL, Venice, Italy). Although the distribution in the chambers was homogeneous, the tomato trays were rotated every day. The spectral parameters were determined by the supplier with an illuminance spectrophotometer (CL-500A, Konica Minolta, Chiyoda, Tokyo, Japan), and are presented in Fig. 1.

The experimental design (Fig. 1) consisted of 6 containers with 12 trays per container. Each container represented one light treatment. Each tray contained 6 Kumato® cherry tomatoes. Measurements were taken at the top half of each tomato (n = 18), for which a longitudinal cut was made. These halves were formed by the seeds, the pericarp, and the skin. The experiment lasted 13 days at 5 °C with sampling days on 0,

Table 1
Main physicochemical quality parameters of studied Kumato® cherry tomatoes (n = 50).

Initial quality and visual characterisation		
Weight (g)		14.6 ± 1.8
Caliber (mm)	**Longitudinal**	32.9 ± 1.3
	Equatorial	28.9 ± 1.5
Colour	**L***	39.1 ± 0.6
	a*	2.6 ± 1.9
	b*	18.9
	Chroma	19.3 ± 0.8
	°Hue	82.3 ± 9.6

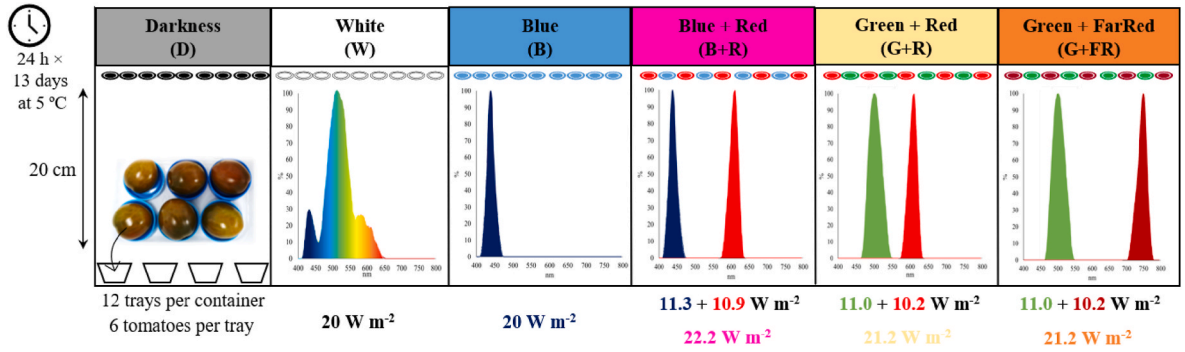

Fig. 1. Experimental design.

3, 6, 10, and 13. In each container there were 3 replicates for each sampling day (12 trays per lighting treatment). Each replicate consisted of 3 trays with 6 individually numbered fruits per tray (n = 18).

2.4. Determination of the main physicochemical quality attributes

Quality characterisation was assayed at harvest as previously described by (Castillejo, Martínez-Hernández, Gómez, Artés, & Artés-Hernández, 2016; Martínez-Zamora, Castillejo, & Artés-Hernández, 2021). Longitudinal and equatorial fruit size was measured, and results were expressed in mm. Tomatoes were individually weighed at harvest and during refrigerated storage to determine weight losses by difference. The main quality attributes determination was carried out on each sampling day. Colour was measured with a Konica Minolta CR-400 colorimeter (Tokyo, Kanto, Japan) in the CIEL*a*b* colour space. Colour variations (ΔE) over the shelf life using the formula described by Torres-Sánchez, Martínez-Zafra, Castillejo, Guillamón-Frutos, and Artés-Hernández (2020). Tomato firmness was measured using a texturometer (Brookfield, CT3-4500, Toronto, ON, Canada) at 20 °C following the method described by Castillejo, Martínez-Zamora, and Artés-Hernández (2022). Such physicochemical determinations were made in triplicate where each replicate was a pooled sample of 6 top longitudinal tomato halves ($n = 18$).

As mentioned above, the fruits were placed on caps separately, with the top of the tomato receiving the highest dose of illumination. Therefore, fresh samples of the top half of all tomatoes were frozen for further analyses (individual phenolic compounds, flavonoids, and carotenoids). In addition, physicochemical analyses were carried out on this upper part of the fruit, for which it was necessary to extract the juice with a Robot Coupe J80 Ultra (Vincennes, Île-de-France, France). A digital refractometer (Atago N1; Tokyo, Kanto, Japan) was used to measure the Total Soluble Solids Content (SSC). A pH-meter (GLP21, Crison; Alella, Cataluña, Spain) was used to measure the pH. Titratable acidity (TA) was determined by acid-base titration using a MPT-Titrino798 from Metrohm AG (Herisau, Switzerland) (Castillejo et al., 2022). The maturity index (MI) was calculated as SSC/TA ratio. These physicochemical determinations were carried out in triplicate where each replicate was a juice of 6 individual longitudinal tomato halves ($n = 3$).

2.5. Determination of individual phenolic compounds and flavonoids

The frozen samples were ground to fine powder prior to analysis using liquid nitrogen, with a basic grinder (IKA, A 11 basic, Berlin, Germany) at 12,700 g for 10 s. For the extraction, 1 g of each sample was weighed in triplicate into 50-mL tubes and 10 mL of methanol:water (80:20, v:v) was added to each and homogenised with Ultraturrax (IKA, Berlin, Germany). The extraction was carried out following the method described by Castillejo et al. (2022). The UHPLC (Shimadzu, Kyoto, Japan) used was equipped and used as described by Castillejo et al. (2022). Phenol and flavonoid peaks were identified by comparison of the retention time and spectra with standards. The chromatographs were recorded from 280 to 360 nm. Each sample was analysed in triplicate where each replicate was a frozen powder of 6 top longitudinal tomato halves ($n = 3$) and the results were expressed as g kg^{-1} fresh weight (fw).

2.6. Extraction and analyses of carotenoids

To carotenoid analysis, 1 g of tomato frozen powder was homogenised with 5 mL of chloroform:dichloromethane (2:1, v/v) in a basic grinder (IKA A11, Staufen, Germany). The extraction was carried out following the method described by Martínez-Zamora, Castillejo, and Artés-Hernández (2021). The filtered extract (0.2 μm PTFE) was stored into vials at −80 °C until UHPLC analyses. The UHPLC (Shimadzu, Kyoto, Japan) used was equipped and used as described by Martínez-Zamora, Castillejo, Gómez, and Artés-Hernández (2021). The peaks areas were recorded at 476 nm. Carotenoids were quantified as equivalents of β-carotene equivalents and results were expressed as mg kg^{-1} fw. Each sample was analysed in triplicate where each replicate was a frozen powder of 6 top longitudinal tomato halves ($n = 3$).

2.7. Statistical analysis

Statistical analysis was performed in Statgraphics Plus software (v. 5.1 Stat- Technologies Point technologies. Inc., Warrenton, VA, USA). The experiment was a two-factor (light treatment × storage time) design subjected to analysis of variance (ANOVA). Statistical significance was assessed at the level of $p < 0.05$, and Tukey's multiple range test was applied to separate means.

3. Results and discusion

3.1. Physicochemical quality changes and fungal growth during shelf-life

The mean weight of the fruits used in the experiment was 14.6 ± 1.8 g, with a mean diameter of 30.9 mm, and commercial characteristic colour of this Kumato® cherry variety (Table 1). Such quality attributes agree with previous findings reported by Giosanu and Vîjan (2016) for this variety, and colour is quite similar to that reported by Li et al. (2022) and Mun et al. (2021), who compared different tomato varieties, including the standard Kumato®.

The water loss due to physiological process of the fruit, and the vapor pressure deficit between the fruit tissues and the environment, is one of the main symptoms of senescence processes leading to a loss of consistency and, therefore, fruit weight. Fig. 2 shows how weight losses increased throughout the refrigerated storage, especially on those cherry tomatoes stored under lighting. Although these weight losses could also be related to possible chilling injury, no visual damage to the tomato skin was observed. Weight losses did not exceed 5%, mainly due to their

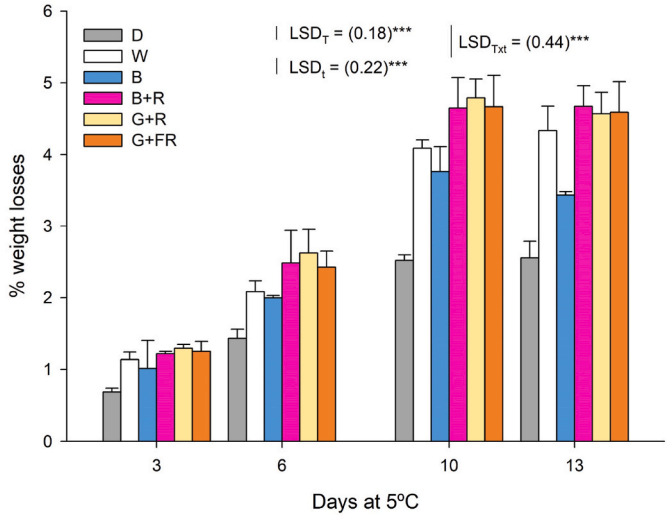

Fig. 2. Weight losses (%) of Kumato® cherry tomatoes stored during 13 days at 5 °C under several illumination treatments ($n = 3$: determinations were made in triplicate where each replicate was a pooled sample of 6 tomatoes).

high volume-to-surface area ratio, and from the 3rd day at 5 °C the lowest dehydration was recorded in tomatoes under darkness conditions (D = 0.69 ± 0.05%). However, the remaining light treatments induced higher weight losses, which increased exponentially over the shelf-life period.

Light treatments increased weight losses by ~70% compared to darkness conditions, without relevant differences among them. Another factor to consider may be the slight temperature increase (~2 °C) inside the trays kept under LED lights, which was monitored during the shelf-life and contributed to the slight warming of the fruits. Also, weight losses were ~25% lower under a continuous illumination with blue LEDs than under the other illumination treatments assayed after 13 days at 5 °C. Such higher weight losses may have been stimulated by the abiotic oxidative stress induced during a continuous illumination, and to the heat transmitted by lighting, although this is much lower than the one transmitted by fluorescent lamps. Thus, light combinations with higher wavelengths (R and FR) induced significant increases of weight loss, as a percentual decrease of the biomass, which have been associated with retained photosynthetic activity of plant tissues resulting in higher water loss through transpiration, as it was observed in fresh-cut red chard and rocket baby leaves (Pennisi et al., 2021).

Firmness is a very important quality attribute in tomato fruits during postharvest. Light and temperature are the main environmental factors that could induce internal changes in the cell wall, and in the cell membrane components, causing this turgor loss. In this way, and since firmness is closely related to dehydration, all tomatoes subjected to continuous illumination (W, B, B + R, G + R, and G + FR) reported a less firmness than those stored in darkness (Table 2). In this sense, when the tomatoes under darkness conditions lost 21% of their initial firmness after 13 days at 5 °C, W, B, B + R, G + R, and G + FR experimented a higher decrease by 33.7, 30.6, 30.7, 34.6, and 35.8%, respectively. After 10 days at 5 °C, the fruits under LEDs lights with higher wavelengths (R and FR) showed 15–21% higher firmness than under D, which was directly correlated to weight losses ($R = -0.7577$; $p < 0.001$). This trend was observed since the 10th day of shelf-life, contrasting results obtained by Ntagkas et al. (2020), who did not observed firmness differences in tomato fruits (*Solanum lycopersicum* cv. Vimoso) stored for 15 d at 18 °C in darkness or under continuous LED illumination (350–500 μmol m^{-2} s^{-1} blue, green, red, far-red, or white), which could be probably due to the higher temperature assayed. In this case, the authors concluded that the light was insufficient to affect the lignin pathway to decrease the firmness of the fruit. By contrast, Martínez-Zamora, Castillejo, and Artés-Hernández (2021) reported a decrease in firmness of red bell peppers by 53% after 6 days 7 °C + 4 days 20 °C, compared to that at harvest, which was directly related to the dehydration induced by the higher temperature during the retail sale period, since no differences were found between bell peppers under a photoperiod during the retail sale with 14 h fluorescent + 10 h blue and red LED lights and a continuous fluorescent lighting.

The remaining physicochemical quality parameters measured are also shown in Table 2. Obtained results showed that the illumination conditions studied did not alter such quality parameters of Kumato® cherry tomatoes. The mean values of SSC were 7.02 ± 0.26 °Brix, while the mean pH values were 4.11 ± 0.04, both with no relevant differences between treatments and/or day of analysis. By contrast, TA after

Table 2
Physicochemical quality changes of Kumato® cherry tomatoes stored during 13 days at 5 °C under several illumination treatments ($n = 3^{\dagger}$: determinations were carried out in triplicate where each replicate was a juice of 6 top longitudinal tomato halves; $n = 18^{\ddagger}$: determinations were made in triplicate where each replicate was a pooled sample of 6 top longitudinal tomato halves).

Illuminating treatments	Days at 5 °C	TSS† (°Brix)	pH†	TA† (g citric acid/100 mL)	ΔE‡	MI‡	Firmness‡ (N)
	0	7.07	3.90 c/b/b/c/c/c	1.01 -/a/a/a/a/a		7.03 -/b/c/c/b/c	30.4 a/a/a/a/a/a
Darkness	3	7.13	4.00 b	0.90	2.38	7.94	26.9 ab
	6	7.30	4.07 ab	0.89	3.17	8.46	24.9 b
	10	7.07	4.10 Ba	1.05 A	2.42 C	6.80 D	25.1 Ab
	13	6.87	4.13 a	0.84	3.53 C	8.31	24.0 b
White	3	6.77	4.13 a	0.83 ab	1.99 d	8.14 ab	26.7 ab
	6	7.07	4.10 a	0.86 ab	3.75 c	8.37 ab	23.6 bc
	10	6.90	4.20 Aa	0.86 Bab	5.60 Ab	8.06 CDab	21.2 BCc
	13	7.03	4.20 a	0.73 b	7.52 Aa	9.73 a	20.1 c
Blue	3	6.73	4.07 ab	0.80 bc	2.37 b	8.42 bc	26.7 ab
	6	7.17	4.03 ab	0.89 ab	2.17 b	8.07 bc	25.8 abc
	10	6.93	4.10 Ba	0.82 BCbc	4.79 Ba	8.51 CDb	23.9 ABbc
	13	7.07	4.20 a	0.70 c	5.28 BCa	10.12 a	21.1 c
Blue+Red	3	7.00	3.97 c	0.86 b	2.38 b	8.16 bc	25.3 ab
	6	7.37	4.10 b	0.79 b	3.34 b	9.42 a	21.4 b
	10	6.83	4.13 ABab	0.74 BCDb	5.71 ABa	9.22 BCab	19.8 Cb
	13	7.03	4.20 a	0.81 b	7.08 ABa	8.71 ab	21.0 b
Green+Red	3	6.80	4.03 b	0.81 ab	1.98 c	8.44 ab	28.2 a
	6	7.20	4.13 ab	0.71 b	2.99 b	10.19 ab	22.6 b
	10	6.87	4.20 Aa	0.60 Db	5.65 ABa	11.45 Aa	21.1 BCb
	13	7.00	4.23 a	0.72 b	6.18 ABa	9.97 ab	19.9 b
Green+FarRed	3	6.83	4.07 b	0.76 b	1.98 b	9.02 b	25.0 bc
	6	7.23	4.10 b	0.77 b	2.98 b	9.45 ab	25.1 b
	10	6.87	4.17 ABab	0.67 CDb	5.20 ABa	10.33 ABab	21.1 BCcd
	13	7.27	4.23 a	0.68 b	6.26 ABa	10.79 a	19.5 d

A, B, C, and D denote significant differences ($p<0.05$) among light treatments at the same sampling day. a, b, c, and d denote significant differences ($p<0.05$) among sampling time under the same light treatment. Each letter in the day 0 row represents significant differences when different from light treatments in the order shown in the table.

harvesting was of 1.01 ± 0.06 g citric acid 100 mL^{-1}, which was partially reduced during the postharvest storage, especially under lighting influence ($p < 0.05$). Generally, the TA values slightly decreased during the shelf-life period as a consequence of maturation, which induced an increase of the MI ($R = -0.9455$; $p < 0.001$). After harvesting, the tomatoes reported a green ripe colour, typical of this variety, and a MI (SSC/TA) of 7.03 ± 0.35. Since tomatoes are climacteric fruits, ripening processes continued throughout the postharvest period in all treatments, however the low storage temperature can inactive the metabolism preventing tomato ripening. It was appreciated a faster ripening under conditions of combined illumination with red light (B + R, G + R and G + FR), reaching MI values of 9.22 ± 0.14, 11.45 ± 1.38, and 10.33 ± 0.41 at day 10, with significant differences with the remaining treatments on the same day.

Our results agree with Liu et al. (2009), who observed minimal effects on colour or SSC during postharvest storage (21 days at 12–14 °C) of tomatoes treated with UV-C and red light (67 W m^{-2}). By the contrary, Kong, Zhao, et al. (2021) showed as SSC initially increased and then decreased again in samples under darkness, red (638 nm), green (520 nm), and white at a light intensity of 30 W m^{-2}, except for tomatoes treated with blue light (442 nm). In this case, tomatoes under blue light initially reported 5.7 °Brix which were continuously increased for 7 days storage by 23%, obtaining the final value of 7.6 °Brix.

Colour changes are probably the most relevant indicator of tomato maturation, because its green colour associated to the chlorophylls content in the chloroplast changes to reddish tones due to the increase in the carotenoid concentration (Li et al., 2022). The mean L* value at harvest was 39.14 ± 0.64, which slightly decreased throughout the shelf-life period to values between 37.87 ± 0.86 and 38.34 ± 0.78 (data not shown). The parameter a* (colour coordinates varying from red (positive values) to green (negative values)) after harvest was 2.62 ± 3.43, which was increased in the illumination treatments compared to D, reaching values of 9.75 ± 2.66 in B samples, 9.36 ± 1.77 in B + R, 8.42 ± 1.70 in G + R, and 8.53 ± 1.93 in G + FR on day 13 (data not shown), associated to a higher ripening of the tomatoes. The b* values (colour coordinates varying from yellow (positive values) to blue (negative values)) were initially 18.92 ± 0.36, which did not significantly change during the refrigerated storage (data not shown). The calculated colour differences (ΔE) are presented in Table 2. No relevant differences during shelf life were found in fruits stored in darkness, which could be an indicator of chilling injury preventing proper ripening. However, fruits under lighting treatments reported a progressive increase of the ΔE value over the storage days. On days 10 and 13, tomatoes stored under B, B + R, G + R, and G + FR illumination conditions obtained the highest values, which indicated a direct relation to the higher accumulation of carotenoids in these samples as it is subsequently explained. This shows that tomatoes stored at 5 °C and under LED lighting continue to ripen and chilling injury is reduced. This trend was also observed in tomatoes stored under continuous red light (113 μmol m^{-2} s^{-1}) for 21 days at which facilitated the development of tomato colour (Panjai et al., 2019).

The studied light treatments did not show any visible fungal growth, which remained quite constant during the shelf-life period below 3.9 log CFU g^{-1} for mould and yeast (data not shown).

3.2. Bioactive compounds changes during shelf-life

3.2.1. Carotenoid content

The most widely known antioxidant compounds in tomato varieties are the carotenoids, especially lycopene, which is the main responsible compound of its reddish colour (Vega-López et al., 2022). This tomato variety is characterized by a darker colour, near the dark green when it matures, which is related to the decrease of red pigments, as lycopene, in comparison with the conventional tomato types. In fact, Mun et al. (2021) have recently shown as red tomato and cherry tomato have approximately twice as much lycopene as the Kumato® variety.

In this sense, the total carotenoid content at harvest of Kumato® cherry tomatoes was 804 ± 38 mg kg^{-1} fw, from which the 66.8% was lycopene, 21.9% lutein, 9.5% β-carotene, and the remaining 1.8% was neoxanthin (Table 3). The high content of the mentioned compounds is related to a dark red fruit colour according to a study by Flores, Sánchez, Fenoll, and Hellín (2017). No relevant changes were appreciated in the biosynthesis of minor carotenoids, as neoxanthin and lutein (greener pigments) under the different light treatments studied. By contrast, the pigments responsible for the orange (β-carotene) and the reddish colour (lycopene) were stimulated by the incidence of LED lighting, especially under B + R, B, G + R, and G + FR conditions, in this order.

The total carotenoid content (Fig. 3.R) is mainly increased by the high content of lycopene in such fruits (Fig. 3.L), which represented the ~67% of the total carotenoid content. These compounds were mainly bio-stimulated with a continuous illumination under B + R lights, since it is demonstrated the efficacy to increase the first stages of the carotenogenesis, where carotenes are biosynthesized (lycopene and β-carotene). In this case, this effect was not triggered to the lower stages of the carotenoid biosynthesis chain, where xanthophylls (minor compounds in tomato) are produced, such as lutein and neoxanthin. Furthermore, phytoene synthase (PSY) as main precursor of carotenoids is essential for the conversion of geranyl diphosphate to phytoene, which directly depends on light stimuli provided by the extremes of the visible spectrum, mainly blue and red regions (Tian, Li, Shah, & Gong, 2015). When the fruits and vegetables photoreceptors of blue light (cryptochromes) receive this stimulus, they activate the transcription factors (protein constitutive photomorphogenesis 1 and elongated hypocotyl 5) in charge of triggering the genetic chain to activate PSY (Artés-Hernández, Castillejo, & Martínez-Zamora, 2022; Tian et al., 2015). In the opposite region of the visible spectrum, red and far-red wavelengths control the phytochrome interacting factors to regulate the photomorphogenesis of these compounds (Artés-Hernández et al., 2022; Llorente, Martinez-Garcia, Stange, & Rodriguez-Concepcion, 2017; Tian et al., 2015).

These results agree with our previous findings in red bell peppers, belonging to the Solanaceae family, which were illuminated with a photoperiod of 14 h Fluorescent lights +10 h with B + R for 4 d at 20 °C at the end of the shelf-life during a simulation of the retail sale period in a supermarket (Martínez-Zamora, Castillejo, & Artés-Hernández, 2021). Furthermore, previous research developed in tomatoes have shown as red light treatments enhances carotenoid accumulation at 13 °C (Liu et al., 2009), 19 °C (Nájera et al., 2018) and 20 °C (Panjai et al., 2019), which is the recommended storage temperature for these fruits. Nevertheless, to ensure the shelf-life during transport through long distances, refrigeration temperatures are frequently used. In this sense, although long expositions to temperatures below the threshold value decrease the synthesis of carotenoids, induces free radical formation, and reduced colour formation (Farneti, Cristescu, Costa, Harren, & Woltering, 2012), the application of LED lighting (blue and red) has demonstrated to be a good option to reduce the chilling injuries (Baenas et al., 2021; Kong, Wen, Jiao, Liu, & Xu, 2021), being a good way to minimize the metabolism to extend the shelf-life enabling their transport.

The descent of carotenoid accumulation in our tomato cherries under darkness conditions may be explained by the low storage temperatures (Farneti et al., 2012), which is countered by the effect of LED lighting (B, B + R, G + R, and G + FR) (Fig. 3). Although these results are not always consistent over time, this observed trend is corroborated by other authors. W light did not report significant improvements compared to D. This effect could be explained by the fact that W light spectrum did not present enough content of B and R light (Fig. 1). In our monochromatic spectrum the wavelength ranged from 410 to 470 nm for blue and 570–640 nm for red. The blue and red region in our W light spectrum was below 30 and 26% of the relative spectral response. In this sense, the full spectrum LED, with higher proportion of R light, stimulated the synthesis of bioactive compounds in tomatoes, specially of the carotenoid content (Nájera et al., 2018). However, in our previous works we have demonstrated as the combination of B + R light, in similar

Table 3
Carotenoids content (mg kg^{-1} fw) changes of Kumato® cherry tomatoes stored during 13 days at 5 °C under several illumination treatments (n = 3: analyses were made in triplicate where each replicate was a frozen powder of 6 top longitudinal tomato halves).

Illuminating treatments	Days at 5 °C	Neoxanthin	Lutein	β-carotene	Lycopene
	0	15.0 ab/-/-/-/-/-	176 -/-/-/-/-/a	76 ± 0 b/a/b/b/b/c	537 ab/abc/-/c/b/b
Darkness	3	14.9 b	166	86 a	586 ABa
	6	15.2 ab	157	76 Bb	571 a
	10	15.4 a	168	75 Bb	488 Cbc
	13	15.0 ab	149	68 Bc	441 BCc
White	3	16.0	154	89	641 ABa
	6	15.9	149	83 AB	630 ab
	10	16.5	145	83 AB	491 BCc
	13	14.8	138	75 AB	509 ABbc
Blue	3	15.2	152	83 ab	605 AB
	6	16.2	180	91 Aba	620
	10	14.6	147	87 ABab	575 ABC
	13	15.0	164	80 Aab	596 A
Blue+Red	3	15.4	145	87 ab	648 Aab
	6	14.8	163	94 Aa	682 a
	10	14.9	154	95 Aa	607 ABbc
	13	15.2	162	78 Ab	551 Ac
Green+Red	3	16.2	164	84 ab	551 Bb
	6	15.4	155	77 ABa	652 ab
	10	15.4	154	86 Aba	638 Aa
	13	15.5	159	84 Ab	529 ABc
Green+FarRed	3	15.1	130 b	81 bc	560 ABb
	6	15.7	161 ab	87 ABab	621 ab
	10	15.1	151 ab	92 Aa	662 Aa
	13	15.6	156 ab	84 Ab	368 Cc

A, B, C, and D denote significant differences ($p<0.05$) among light treatments at the same sampling day. a, b, c, and d denote significant differences ($p<0.05$) among sampling time under the same light treatment. Each letter in the day 0 row represents significant differences when different from light treatments in the order shown in the table.

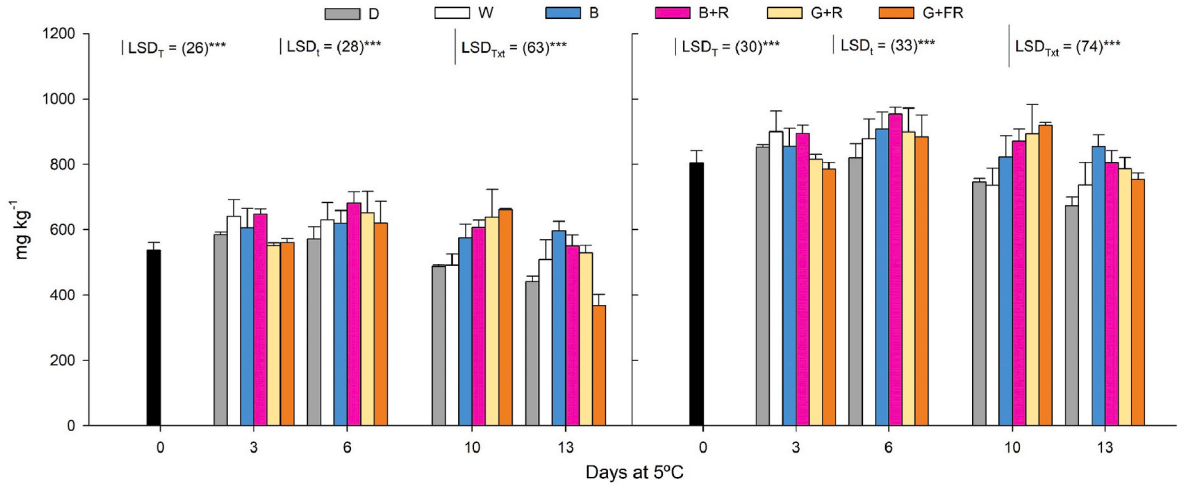

Fig. 3. Total lycopene content (mg kg^{-1} fw; left) and total carotenoid content (mg kg^{-1} fw; right) of Kumato® cherry tomatoes stored during 13 days at 5 °C under several illumination treatments (n = 3: analyses were made in triplicate where each replicate was a frozen powder of 6 top longitudinal tomato halves).

conditions as hereby described, affected the synthesis of carotenoids in bell peppers (Martínez-Zamora, Castillejo, & Artés-Hernández, 2021) and carrot sprouts (Martínez-Zamora, Castillejo, Gómez, & Artés-Hernández, 2021).

As it was early described, the use of such illumination preserved the carotenoid content during a refrigerated shelf-life (5 °C), by avoiding the breakdown of carotenoids or stimulating their synthesis (Rai et al., 2022). Therefore, chilling injury symptoms were not visible in our tomatoes during 13 days at 5 °C, but primary symptoms were observed due to the metabolic change of reduced carotenoid biosynthesis.

3.2.2. *Phenolic acid and flavonoid content*

One of the most relevant substances responsible of the antioxidant activity of tomatoes are the phenolic acids and flavonoids. The total polyphenols content of Kumato® tomatoes at harvest was 243.7 mg kg^{-1} fw (Fig. 4), which was determined by the sum of individual phenolic acids and individual flavonoids, accounting for 2 and 98%, respectively, of the total polyphenols content.

As shown in Table 4, the main phenolic acids identified were gallic, chlorogenic, caffeic, ρ-coumaric, and ferulic acids, with chlorogenic acid being the predominant, followed by caffeic acid, both representing more than 70% of the total phenolic acids. The individual phenolic acids identified are in agreement with Slimestada and Verheulb (2009), where chlorogenic acid and its derivates are reported to be the main phenolic compounds found in tomato. The individual flavonoids identified were rutin, kaempferol, and naringenin, being the latter one the main flavonoid found, and accounting for 62% of the total flavonoid content (Table 5). Anton et al. (2017) and Slimestada and Verheulb (2009) also reported that naringenin is the main flavonoid found in tomato.

The initial individual phenolic acids content at harvest was 5.85 mg kg^{-1} fw. After 3 days at 5 °C, the total phenolic acid content was 15.4% higher in tomatoes stored under G + R illumination than under darkness conditions. However, these increases were not maintained during the 13 days at 5 °C. This increase was related to the increase in caffeic acid, which reported a 35% increase compared to tomatoes stored under darkness. The content of the remaining phenolic acids identified was

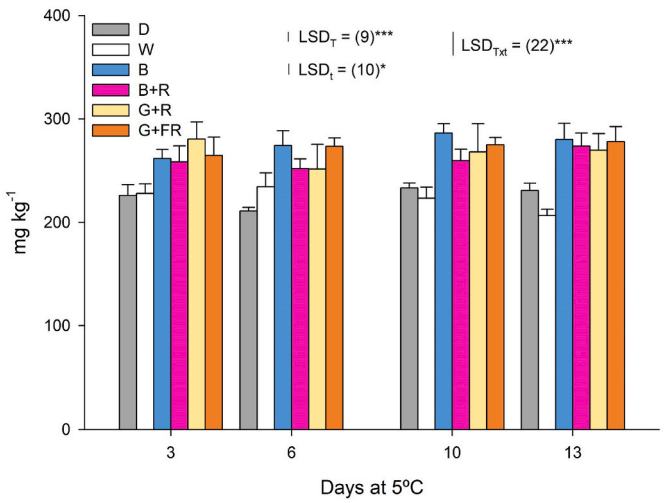

Fig. 4. Total phenolic content (mg kg^{-1} fw) of Kumato® cherry tomatoes stored during 13 days at 5 °C under several illumination treatments (n = 3: analyses were made in triplicate where each replicate was a frozen powder of 6 top longitudinal tomato halves).

maintained for 13 days at 5 °C, without significant differences compared to D. Panjai, Noga, Fiebig, and Hunsche (2017) reported a 44% increase in green-stage tomatoes after 5 days at 20 °C under red light (113 μmol m^{-2} d^{-1}). Blue and red lights have been widely studied for being included in PAR spectral range, the incidence of these lights on plant tissues activates cryptochromes and phytochromes, respectively, by regulating the synthesis of phenolic compounds as secondary metabolites. Furthermore, green light has also a positive effect on plants, in where cryptochromes absorb this type of light, triggering a genetic response that stimulates the synthesis of secondary metabolites (Artés-Hernández et al., 2022).

The content of individual flavonoids identified was stimulated by different visible spectrum illuminations, with or without combination. The initial individual flavonoid content in harvested tomatoes was 237.9 mg kg^{-1} fw, from which 62.3% was related to the naringenin content. W illumination did not report a positive effect on the naringenin content, showing a decrease of 35% after 10 days at 5 °C compared to initial day, without significant difference compared to D. However, after 3 days at 5 °C, the naringenin content of tomatoes stored under B, B + R, G + R, and G + FR was 18, 25, 29, and 32% higher than under darkness, respectively. The following major flavonoid found was rutin, accounting for 24%, which was 17 and 25% higher after 3 days at 5 °C in tomatoes stored under B and G + R continuous illumination compared to D. The minor flavonoid identified was kaempferol, accounting for 14%, which was preserved during the continuous exposure to the illumination conditions assayed (B, B + R, G + R, and G + FR). In addition, rutin and kaempferol contents were maintained during the refrigerated storage without significant differences compared to the values at harvest. By contrast, longer continuous exposure to visible spectrum lights, with or without combinations, increased the naringenin content of tomatoes stored at 5 °C. However, since day 10, the naringenin content was maintained up to 13 days. As for naringenin, the same trend was observed for the total flavonoid content due to its high content in relation to the total. From a general point of view, the combined light spectrum of green with red (G + R) and far-red (G + FR) recorded 24 and 18% higher flavonoid levels, respectively, after 3 days at 5 °C than D and W light treatments. However, after 6 days onwards, all studied visible spectra improved the total flavonoid content.

Fig. 3 shows how all illumination treatments induced a higher accumulation of total phenolics, in where just 6 days at 5 °C under these light conditions was enough to achieve the maximum increases (30, 19, 19, and 30%) in tomatoes stored under B, B + R, G + R, and G + FR, respectively, compared to D. However, after 13 days at 5 °C, these increases were slightly reduced but still above the initial observed increases. No differences were observed during 13 days at 5 °C among W and D treatments, in where even less accumulation in some sampling days was found. In this sense, higher light intensity, as an abiotic stress, triggers a higher photosynthetic rate by increasing the plant metabolism, which leads to an increase in phytochemical compounds, which are used by the plant to protect itself from the high amount of reactive oxygen species (Ntagkas et al., 2020). Recently in other vegetables as rocket sprouts, the phenolic acids and the flavonoid biosynthesis were enhanced by ~25%,~30%, and ~55% under photoperiods with White, Blue, and Red LEDs, respectively, compared to darkness (Martínez-Zamora, Castillejo, & Artés-Hernández, 2022).

Besides the light presence, its quality is very important, so the specific region of the emitted spectrum should be as narrow as possible and

Table 4
Phenolic acids content (mg kg^{-1} fw) changes of Kumato® cherry tomatoes stored during 13 days at 5 °C under several illumination treatments (n = 3: analyses were made in triplicate where each replicate was a frozen powder of 6 top longitudinal tomato halves).

Illuminating treatments	Days at 5 °C	Gallic acid	Chlorogenic acid	Caffeic acid	*p*-coumaric acid	Ferulic acid	Total phenolic acids
	0	0.73 ab/b/c/b/c/b	2.18 a/a/a/a/a/a	1.94 a/a/a/a/a/a	0.55 -/-/-/b/b/-	0.46 -/-/b/c/b/-	5.85 a/a/a/a/a/a
Darkness	3	0.73 Bab	1.93 ABb	1.63 BCab	0.57 ABC	0.48 BC	5.32 BCab
	6	0.71 b	1.97 Bab	1.31 bc	0.54	0.48 AB	5.01 ABb
	10	0.70 Bb	1.76 Bb	1.17 Abc	0.57	0.53 AB	4.71 Bb
	13	0.81 Ca	1.76 ABb	1.03 Ac	0.56 B	0.50 B	4.65 b
White	3	0.92 Aa	1.76 BCc	1.31 BCb	0.53 C	0.43 C	4.96 CDb
	6	0.82 Aab	2.22 Aa	1.25 b	0.54	0.48 AB	5.31 Aab
	10	0.84 ABab	2.08 Aab	0.99 Bb	0.55	0.47 B	4.92 ABb
	13	0.96 ABa	1.94 Abc	0.92 Bb	0.55 B	0.49 B	4.86 b
Blue	3	0.88 ABa	1.98 ABbc	1.81 ABa	0.58 AB	0.57 Aa	5.79 ABa
	6	0.81 bc	2.06 ABab	1.36 ab	0.55	0.52 ABab	5.29 Aab
	10	0.93 Aa	2.05 Aab	1.17 Ab	0.58	0.58 Aa	5.28 Aab
	13	0.90 BCa	1.84 Ac	0.97 ABb	0.57 B	0.55 Ba	4.81 b
Blue+Red	3	0.86 ABab	2.01 Aab	1.26 Cb	0.56 ABCab	0.50 Bbc	5.18 BCDab
	6	0.74 b	1.74 Cc	1.32 b	0.57 ab	0.56 Aab	4.91 ABb
	10	0.82 ABab	1.87 Bbc	0.91 Bb	0.55 b	0.50 ABbc	4.65 Bb
	13	0.98 ABa	1.46 Bd	1.04 Ab	0.60 Aa	0.65 Aa	4.68 b
Green+Red	3	0.78 ABbc	2.00 ABab	2.20 Aa	0.60 Aa	0.61 Aa	6.14 Aa
	6	0.70 c	1.68 Cc	1.24 Ab	0.52 b	0.43 Bb	4.59 Bb
	10	0.87 Ab	1.76 Bbc	0.98 Bb	0.56 ab	0.51 ABb	4.67 Bb
	13	1.03 Aa	1.64 ABc	0.97 ABb	0.56 Bab	0.51 Bb	4.71 b
Green+FarRed	3	0.75 Bb	1.56 Cb	1.24 Cb	0.55 BC	0.48 BC	4.56 Db
	6	0.76 b	1.63 Cb	1.13 Ab	0.55	0.50 AB	4.57 Bb
	10	0.94 Aa	1.75 Bb	1.01 Bb	0.55	0.50 AB	4.74 Bb
	13	0.94 ABa	1.69 ABb	1.00 ABb	0.56 B	0.52 B	4.69 b

A, B, C, and D denote significant differences ($p<0.05$) among light treatments at the same sampling day. a, b, c, and d denote significant differences ($p<0.05$) among sampling time under the same light treatment. Each letter in the day 0 row represents significant differences when different from light treatments in the order shown in the table.

Table 5
Flavonoids content (mg kg^{-1} fw) changes of Kumato® cherry tomatoes stored during 13 days at 5 °C under several illumination treatments (n = 3: analyses were made in triplicate where each replicate was a frozen powder of 6 top longitudinal tomato halves).

Illuminating treatments	Days at 5 °C	Rutin	Kaempferol	Naringenin	Total flavonoids content
	0	56.0 -/-/b/b/b/-	33.7 b/-/b/-/-/-	148.2 a/a/b/-/-/b	237.9 a/a/b/-/-/b
Darkness	3	56.9 C	35.4 ab	128.6 Cbc	220.9 Bab
	6	55.2 AB	36.8 Aa	114.1 Dc	206.1 Cb
	10	59.0 AB	36.8 a	132.7 Bab	228.6 BCab
	13	57.3 B	35.7 a	133.3 BCab	226.3 Bab
White	3	49.8 C	35.1	138.3 BCa	223.2 Bab
	6	56.3 AB	35.5 AB	137.4 CDa	229.2 BCab
	10	54.1 B	35.2	129.0 Bab	218.3 Cab
	13	54.5 B	34.7	112.4 Cb	201.7 Bb
Blue	3	66.4 ABa	37.8 a	151.8 ABCab	255.9 ABab
	6	60.3 ABab	36.3 ABab	172.5 Abab	269.1 Aab
	10	64.6 Aab	36.0 ab	180.5 Aa	281.2 Aa
	13	60.4 Bab	34.8 ab	180.2 Aa	275.5 Aa
Blue+Red	3	57.0 BCb	35.5	161.0 AB	253.5 AB
	6	63.5 Aab	36.0 AB	147.7 BC	247.2 AB
	10	55.8 Abb	34.3	164.9 A	255.0 ABC
	13	69.8 Aa	37.0	162.3 AB	269.2 A
Green+Red	3	71.2 Aa	37.1	166.1 AB	274.4 A
	6	48.6 Bb	32.8 B	165.6 ABC	247.0 AB
	10	56.0 Abb	34.8	172.6 A	263.4 AB
	13	56.1 Bb	35.7	173.4 A	265.2 A
Green+FarRed	3	56.1 C	34.7	169.3 Aab	260.1 Aab
	6	57.0 AB	34.2 AB	177.8 Aa	269.0 Aab
	10	54.9 AB	34.3	181.2 Aa	270.4 Aab
	13	57.5 B	36.0	179.9 Aa	273.5 Aa

A, B, C, and D denote significant differences ($p<0.05$) among light treatments at the same sampling day. a, b, c, and d denote significant differences ($p<0.05$) among sampling time under the same light treatment. Each letter in the day 0 row represents significant differences when different from light treatments in the order shown in the table.

associated with a specific colour (Artés-Hernández et al., 2022).

3.2.3. Total antioxidant compounds

As the sum of the identified main compounds with potential antioxidant activity (phenolic acids, flavonoids, and carotenoids), we have included the total content of antioxidant compounds in tomato fruits stored for 13 days at 5 °C under different lighting conditions (Fig. 5). The 75–77% of the total bioactive compounds were carotenoids, while the remaining antioxidants were phenolic compounds, mainly flavonoids (22–24%) and in a residual value the phenolic acids (0.4–0.5%).

In this way, following the same behaviour that the previously described individually content, the sum of the total antioxidant compounds was significantly higher in the tomatoes illuminated with B + R (1142.1 mg kg^{-1} fw; +14–19% compared to D during the refrigerated storage; $p < 0.05$) and B (1136.2 mg kg^{-1} fw; +13–25%; $p < 0.05$), followed by G + R (1116.6 mg kg^{-1} fw; +11–18%; $p < 0.05$), G + FR (1108.8 mg kg^{-1} fw; +11–22%; $p < 0.05$), W (1035.6 mg kg^{-1} fw; +3–7%; $p > 0.05$), and D (998 mg kg^{-1} fw) in this order. The proposed alternative LED illuminations showed ~12% more bioactive compounds than W and D, although when the tomatoes ripening was increased, after 10 and 13 days, these values increased up to ~22%. This effect may be explained since W light spectrum did not have enough R (Fig. 1), as cited above, since previous authors have demonstrated as the presence of R LEDs affects directly to the synthesis of carotenoids in bell peppers (Martínez-Zamora, Castillejo, & Artés-Hernández, 2021), carrot sprouts (Martínez-Zamora, Castillejo, Gómez, & Artés-Hernández, 2021), and different tomato varieties (Nájera et al., 2018).

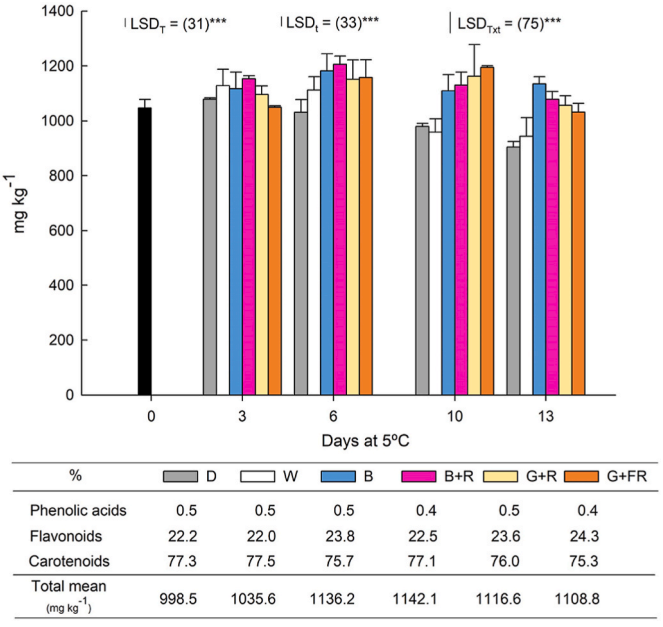

Fig. 5. Total antioxidant compounds (mg kg^{-1} fw) of Kumato® cherry tomatoes stored during 13 days at 5 °C under several illumination treatments (n = 3: analyses were made in triplicate where each replicate was a frozen powder of 6 top longitudinal tomato halves).

Therefore, a continuous illumination with the alternative lights did not report significant differences among them, single or combined, but they always showed higher accumulations of bioactive compounds regarding white heterochromatic lights and darkness.

According to these results, the decrease of antioxidant compounds under darkness conditions could be due to the induction of free radical formation and the decrease of the synthesis of antioxidant compounds, such as carotenoids, phenolic acids, or flavonoids (Farneti et al., 2012). Therefore, the application of visible spectrum LED lighting (B, B + R, G + R, and G + FR) resulted in an interesting tool to minimize losses of bioactive compounds during a refrigerated storage (Fig. 5). Another point to note is that these increases could be due to the concentration of phytochemicals, as the weight loss of tomatoes under visible spectrum LED light was greater than under darkness and white light. Therefore, a postharvest illumination of specific zones of the visible spectrum is recommended to avoid the breakdown of phytochemicals and/or to increase their biosynthesis.

4. Conclusions

A postharvest LED illumination with specific wavelengths of the visible spectrum improved the bioactive quality of Kumato® cherry tomatoes during a refrigerated shelf-life period without relevant changes in the physicochemical quality. Although a continuous high intensity illumination is not commercially viable during shelf-life, our

results are a first step for future research in where several parameters and conditions should be still elucidated and optimized. Further investigations on the effect of such lighting treatments during a photoperiod, in where a conventional light is applied during the day, and such alternative lighting treatments during nights (instead of darkness) seems worthy to perform. Moreover, light intensity optimization and/or supplementation with other wavelengths during the photoperiod seems also an interesting issue to study to give a step forward. Therefore, the next step should be to elucidate if it is still possible to increase the biosynthesis of the main antioxidants compounds, by activating their defence mechanisms, improving their bioactive compounds content, in such commercial illumination conditions to be able to transfer the results to the industry in the supply chain.

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CRediT authorship contribution statement

Lorena Martínez-Zamora: L.M.Z., Conceptualization, Methodology, Formal analysis, Investigation, Software, Validation, Writing – original draft, Writing – review & editing, Supervision. **Noelia Castillejo:** N.C, Conceptualization, Methodology, Formal analysis, Investigation, Validation, Data curation, Writing – original draft, Writing – review & editing. **Francisco Artés–Hernández:** F.A.H, Conceptualization, Methodology, Validation, Resources, Writing – original draft, Writing – review & editing, Visualization, Project administration, Funding acquisition.

Declaration of competing interest

The authors declare no conflict of interest.

Data availability

The authors are unable or have chosen not to specify which data has been used.

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