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Effects of UV-C, red light and sun light on the carotenoid content and physical qualities of tomatoes during post-harvest storage

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ABSTRACT

Mature-green (breaker-stage) tomatoes were harvested and treated daily with short bursts of UV-C, red light or sun light for up to 21 days. Control untreated tomatoes were kept in the dark for the same period. The effects of the treatments on the levels of the major tomato carotenoids, skin colour, tissue firmness and total soluble refractive solids were evaluated throughout storage. Results indicated that the concentration of lycopene in tomato exocarp was significantly increased after 4 days and dramatically enhanced by UV-C or red light treatments. However, the concentration of β-carotene was not affected by UV-C or red light treatments, and decreased by sun light treatment during 21 days of storage, compared to the control samples. The colour (a^* and b^* values) and force required to penetrate the tomatoes was, to a small but significant extent, influenced by the light treatments. However, the total soluble refractive solids of all tomato samples remained the same throughout storage. The findings reported here could be employed to improve tomato nutritional qualities lycopene content without inducing significant changes to the physical properties of tomatoes during post-harvest storage.

1. Introduction

Tomato is a climacteric fruit and continues to ripen after harvest. During ripening, the green pigment chlorophyll degrades and carotenoids are synthesized. Carotenoids, particularly lycopene and β-carotene, represent the primary components of ripe fruit pigmentation in tomato pericarp and are responsible for the characteristic colour of ripe tomatoes, conferring deep red and orange colours, respectively. These carotenoids largely influence the quality perception of fresh tomatoes. For fresh tomatoes, texture and colour are the most important quality attributes, which directly relate to their marketing value (Tijskens & Evelo, 1994).

The content of carotenoids in tomatoes is important, not only due to the colour they impart, but also due to their acknowledged health benefits. Several epidemiological studies have reported that the dietary intake of carotenoids reduces the incidence of degenerative diseases, including heart disease and cancer. Therefore, considerable work has been conducted to increase their levels in tomatoes through breeding programmes or ripening intervention technologies during post-harvest storage (Alba, Cordonnier-Pratt, & Pratt, 2000; Liu et al., 2003; Rosati et al., 2000).

Early studies indicated that phytochromes mediate light-induced carotenoid biosynthesis in tomato by conducting red and far-red light during ripening (Khudairi & Arboleda, 1971; Thomas & Jen, 1975). Alba and co-workers (2000) reported that red light treatments (six 40 W Gro-lux lamps) increased lycopene accumulation 2.3-fold in tomatoes and that red light-induced lycopene accumulation was reversible by far-red light treatment. They concluded that the accumulation of lycopene was under the control of fruit-localised phytochromes. Other studies have shown that red light treatment increases the carotenoid content and red colour of tomatoes, with varying effects on tomato firmness (Lee, Bunn, Han, & Christenbury, 1997).

Ultraviolet (UV) radiation (100–400 nm) can effectively penetrate into the plant tissues and be absorbed. Maneerat, Hayata, Muto, and Kuroyanagi (2003) reported that UV-A irradiated tomatoes show normal colour development and fruit ripening without any physiological disorder. Additional studies have reported that low-dose UV-C can induce resistance to Rhizopus soft rot, delay ripening, improve firmness and extend the shelf-life of tomatoes (Liu et al., 1993; Luckey, 1980; Stevens et al., 1996, 1998 and 2004; Wilson et al., 1994). Similarly, Maharaj, Arul, and Nadeau (1999) have reported that UV-C irradiation at 3.7 kJ/m^2 and 24.4 kJ/m^2 delays the development of tomato tissue colour and softening. Barka, Kalantari, Makhlouf, and Arul (2000) also reported that treatment of green tomatoes with UV-C light (peak output of 254 nm) reduces cell-wall degrading enzyme activity. Although UV light seems to have a physiological effect on tomatoes, to our knowledge there are no reports on the effect of UV light treatment on the concentration of the major carotenoids in tomatoes.

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In nature, carotenoids mainly occur in the all-trans configuration (Chandler & Schwartz, 1987). In this paper, all-*trans*-lycopene and all-*trans*–carotene are subsequently referred to as lycopene and β-carotene, respectively. The aim of the present work was to investigate the effects of UV-C (254 nm), red light (610–750 nm) and total sun light (100–10^6 nm) on tomato carotenoids (lycopene and β-carotene) and selected tomato qualities (colour, hardness, and total soluble refractive solids) during post-harvest storage under semi-industrial conditions.

2. Materials and methods

2.1. Tomato fruit

Tomatoes (*Lycopersicon esculentum* cv. Red Ruby) of mature green colour (breaker-stage) were sourced from Shepparton, North East Victoria, Australia. Fruit diameters were 55–60 mm. Tomatoes were kept at ambient temperature during transportation.

2.2. Storage

Two days after harvest, tomatoes were evenly placed without touching each other onto plastic trays one layer thick. Based on Australian industry practice, the tomatoes were stored for 21 days in the dark between 12 and 14 °C with fans continuously circulating air across the tomatoes.

2.3. Light treatment

A box (1.265 m deep, 1.265 m wide and 0.5 m deep) was constructed from white melamine- covered particle board (13mm thick) containing six 36 W 'red' Gro-lux lamps (Sylvania Australia, NSW, Australia) and six 36 W germicidal lamps (Philips Australia, NSW, Australia). The electrics were mounted at the top, so the lights could be operated in isolation. The six 36 W Gro-lux lamps had an irradiance of 15.5 W/m^2 when measured over the region of 610–750 nm by Spectroradiometer (IL1700, International Light Technologies, MA, USA). The six germicidal UV-C lamps had an irradiance of 22.8 W/m^2 when measured at 254 nm by UV radiometer (IL1700, International Light Technologies, MA, USA).

Every day, the tomato trays were placed into the box. The protocol for UV-C light treatment was based on the study of Maharaj et al. (1999). The tomatoes were exposed to all six germicidal UV-C lamps for 5 minutes, turned over and exposed again for 5 min, equalling a total daily treatment energy of 13.7 kJ/m^2.

The method of red light treatment was based on the study by Alba et al. (2000), however, the tomatoes were exposed to all six Gro-lux lamps for 12 min, turned over, and exposed again for a further 12 min, equalling a total daily treatment energy of 24.3 KkJ/m^2. Control tomatoes were placed into the box with lights off for 12 min, turned over, and then left for a further 12 min.

Every day (around 12:00pm), the tomato trays were placed outside for sun light treatment. As there are few publications on the effect of sun light treatment on tomato carotenoid content, the tomatoes were exposed to sun light for a total time of 60 min. Tomatoes were exposed to sun light for 30 min, turned over and exposed again for a further 30 min. Using half hour direct exposure sun light data (for Tullamarine Airport, Victoria, Australia) sourced from the Australia Bureau of Meteorology, daily sun light treatment energy was, on average, 1.66 ± 1.52 MJ/m^2. The temperature during the sun light treatment was recorded and was between 26 and 31 °C.

2.4. Colour

At 0, 4, 15 and 21 days of treatment, tomato surface colour values were measured using a Minolta Chromometer Model CR 200 (Konica Minolta, Tokyo, Japan) and average readings at 9 predetermined points on the equator of each fruit recorded. Nine tomatoes were measured for each treatment. The instrument was calibrated against a standard white tile ($Y = 93.9$, $x = 0.313$, $y = 0.321$). Hunter a^*, b^* and L^* values were obtained, and colour was expressed as the a^*/b^* ratio. The a^*/b^* ratio is the ratio of yellow–red to blue–green components of colour and represents the colour index related to colour variation during tomato ripening (Francis & Clydesdale, 1975).

2.5. Hardness

At 0, 4, 15 and 21 days of treatment, the pressure required to penetrate each tomato in two locations was determined using a fruit penetrometer (Alba et al., 2000). Nine tomatoes were measured for each treatment at each sampling time point.

2.6. Total soluble refractive solids

At 0, 4, 15 and 21 days of treatment, the total soluble refractive solids were determined, in duplicate, with a refractometer, and expressed as a percentage Brix. Nine tomatoes were measured for each treatment. This parameter is an index of soluble solids concentration in the tomato and is shown as g solids per 100 ml (Giovanelli, Lavelli, Peri, & Nobili, 1999).

2.7. Lycopene and β-carotene analysis

For carotenoid analysis, four 2 cm^2 pericarp sections were cut from the equatorial regions of three tomatoes. The sections from the three tomatoes (twelve sections in total) were combined, weighed and immediately frozen with liquid nitrogen. Samples were then stored at −18 °C, freeze dried, and ground with a mortar and pestle. Samples of tomatoes treated with sun light for 15 days were not taken and analysed. All other samples were prepared and analysed in triplicate.

Carotenoids were extracted using a combination of solvent and solid-phase extraction (SPE). Dried tomato powder was mixed with 10 ml of 1:1 cyclohexane and dichloromethane (containing 0.1% BHT) on a shaker (Heidolph Multi Reax) for 20 min at 1400 rpm. The mixture was centrifuged at 2000 rpm for 5 min, using an Orbital 420 centrifuge (Clements Medical Equipment Pty Ltd., NSW, Australia). The extract was decanted into a test-tube and the pellet was re-extracted using the same technique. The two extracts were combined and placed under nitrogen at 30 °C until dry. Re-extraction of previously extracted material showed that 98% of all-*trans*-lycopene and all-*trans*-β-carotene was isolated from the matrix after two extractions. The dried extract was then reconstituted with 2 ml of cyclohexane containing 0.1% BHT. An aliquot (1 ml) was loaded onto a pre-conditioned silica cartridge (500 mg capacity, 4 ml volume, Altech, Baulkham Hills, NSW, Australia). The cartridge was washed with 1 ml of 0.1% BHT in cyclohexane. The retained carotenoids were eluted with 5 ml of 15% (v/v) dichloromethane in cyclohexane and collected in a test-tube. The eluant, containing on average, 97.4 ± 1.2% (mean ± std deviation, $n = 3$) of lycopene and 96.2 ± 1.9% (mean ± std deviation, $n = 3$) of β-carotene, was then filtered through a 0.22 μm filter for quantitative determination of carotenoids.

Carotenoids were quantified using a Shimadzu HPLC system equipped with two high-pressure LC-10ADVP pumps, a SIL-10ADVP auto sampler (250 μl sampling loop), a CTO-1-ADVP column oven and a SPD-M10ADVP photodiode array detector (Shimadzu Inc., Rydalmere, NSW, Australia). A YMC Carotenoid column (C30) was used for the separation of the carotenoids: 4.6 mm i.d × 250 mm length, 5 μm particle size (Waters Associates, Chippendale, NSW, Australia). The mobile phases used were

methanol, MTBE, water (81:15:4) (A) and methanol, MTBE, water (6:90:4) (B) (Schieber, Marx, & Carle, 2002) at a flow rate of 1.5 ml min^{-1}. Analytes were eluted using a linear gradient: 0% to 100% B over 10 min, followed by 100% B for a further 6 min. Detection was achieved at 473 nm for lycopene and 452 nm for β-carotene. Analytes were identified by comparison of their elution times with those of authentic standards (Sigma–Aldrich, Castle-Hill, NSW, Australia). Quantification was achieved by the use of external calibration curves (76–1220 ng on column, r^2: 0.9999 for lycopene; 22–430 ng on column, r^2: 0.9996 for β-carotene).

2.8. Statistical analysis

All results are expressed as the mean, plus or minus the standard error of the mean. The significant difference between samples was analysed by means of one-way ANOVA and Student's *t*-Test. All of the statistical analyses were performed using Microsoft Excel 2003 service pack 2 (Microsoft, WA, USA). The threshold *p*-value chosen for statistical significance was $p < 0.05$.

3. Results and discussion

3.1. Chromatographic separation of carotenoids

Reverse-phase HPLC is fast becoming the method of choice for the separation and determination of carotenoids from food matrices. The commercially available C_{30}-based YMC column has the ability, in combination with the right mobile phase, to separate most *cis*- and *trans*- forms of the major carotenoids within a relatively short run time (Fig. 1).

3.2. The effects of storage time on the levels of lycopene

The lycopene contents of tomatoes, untreated (stored in the dark only) and treated with UV-C, red light or sun light during 21 days of storage are presented in Fig. 2. There were various effects of storage time on the lycopene contents of light treated and untreated tomatoes during this period.

The accumulated lycopene contents of all tomatoes (untreated, red light, UV-C and sun light treated) did not change significantly (average 29 ± 6 g/g dry mass) during the first 4 days of storage ($p > 0.05$).

Between days 4 and 21, lycopene levels in untreated tomatoes increased, attaining maximal levels of 85 ± 15 g/g dry mass at day 15, which represented an increase of 3.5 fold, compared to that at day 4. The results were similar to those observed by Alba and co-workers (2000), who found that lycopene accumulated over 16 days when tomatoes were stored in the dark.

Between days 4 and 21, the lycopene content of the sun light-treated tomatoes increased 2.4-fold. The lycopene content of the UV-C and red light treated tomatoes increased by 6- and 9-fold over the same period, respectively.

The results obtained from this study showed that the levels of lycopene started increasing after 4 days of storage in all tomato samples (light- treated and untreated), which was comparable to results obtained by Fraser, Truesdale, Bird, Schuch, and Bramley (1994). The results obtained were also consistent with the study of Schofield and Paliyath (2005), who reported that tomato disc

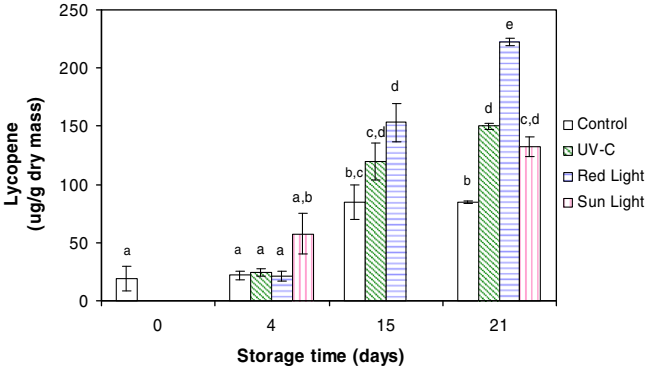

Fig. 2. Concentration of lycopene in untreated (Control), UV-C light-, red light- and sun light- treated tomatoes after 0, 4, 15 and 21 days of treatment and storage. The error bars represent standard errors of the means ($n = 3$).

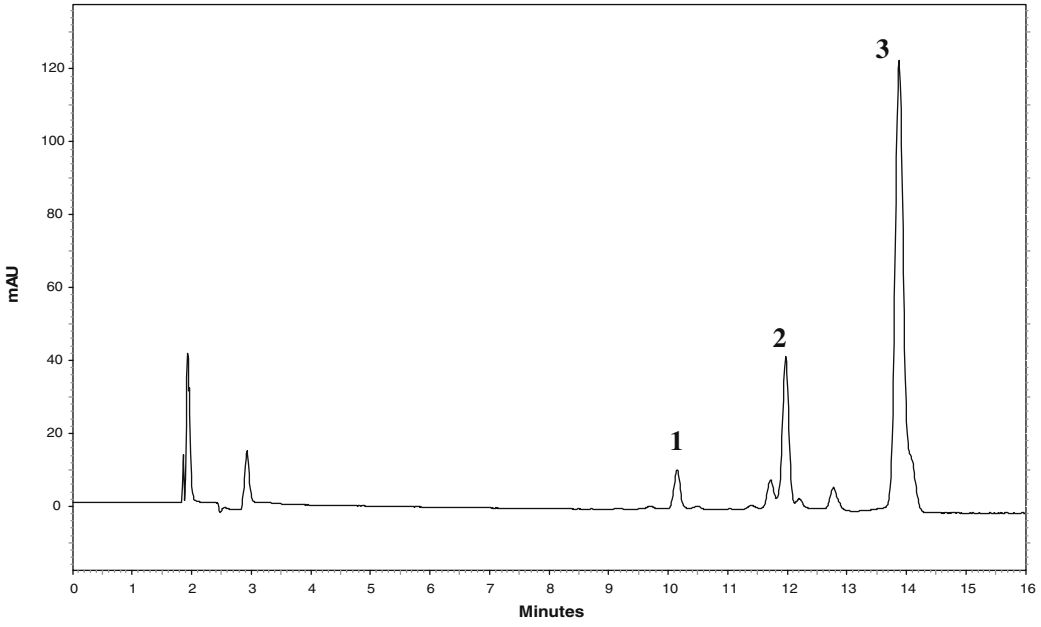

Fig. 1. Typical HPLC trace ($\lambda = 452$ nm) of a tomato extract obtained using the conditions described herein. Major peaks corresponding to all-*trans*-β-carotene (1), 13-*cis*-lycopene (tentative identification) (2) and all-*trans*-lycopene (3) are shown.

carotenoid levels did not change during the first 4 days of storage, and started increasing after 4 days of incubation in darkness or when exposed to either red light or red light followed by far-red light. This observation implies that light is not essential for the induction of lycopene synthesis in tomatoes, at least after the immature-green stage, but is an important factor influencing lycopene accumulation in tomatoes (McCollum, 1953). Therefore, it appears that lycopene accumulation in tomato pericarp consists of both light-independent and light-dependent components (Raymundo, Chichester, & Simpson, 1976).

3.3. The effects of light treatments on the level of lycopene during storage

The lycopene contents of the tomatoes were increased greatly by red light treatment, compared to those of control tomatoes after 4 days of storage (Fig. 2). The lycopene contents of tomatoes subjected to red light treatment were increased by 1.8-fold at the 15th day and by 2.6-fold at the 21st day of storage ($p < 0.05$), compared to that of control tomatoes. These increases were greater than those reported by Schofield and Paliyath (2005). In their study, only a 50% enhancement of total carotenoid accumulation was observed in discs exposed to red light.

The UV-C treatment also increased the lycopene content of tomatoes during storage (Fig. 2). The lycopene contents of UV-C treated tomatoes increased by 1.4- and 1.8-fold at days 15 and 21 of storage, respectively, when compared to that of untreated tomatoes. Low-dose UV-C light has been linked with elevated disease resistance and extension of tomato shelf-life in many previous studies (Barka et al., 2000; Liu et al., 1993; Maharaj et al., 1999; Stevens et al., 1998; Stevens et al., 2004). To the best of our knowledge, this is the first published report that describes a UV-C light-induced increase in tomato lycopene content.

The sun light treated tomatoes had a 1.5-fold increase in lycopene content at day 21 of storage, when compared to that of untreated control tomatoes (Fig. 2). The increase observed after 21 days of sun light treatment was similar to that observed after 21 days of UV-C treatment. These results were similar to those reported by McCollum (1953) with sun light treated detached tomatoes. The results obtained in this study indicate that the lycopene content of stored tomatoes is enhanced by daily red light, UV-C or sun light treatments. Red light treatment has a greater effect on tomato lycopene content than has UV-C or sun light treatment. In this study, sun light had the least impact on tomato lycopene content. This indicates that light (especially at red or UV-C wavelengths) might be a specific regulator of carotenoid synthesis and accumulation in tomatoes during post-harvest storage.

3.4. The effects of light treatments on the level of β-carotene during storage

The β-carotene contents of tomatoes, untreated (stored in the dark only) and treated with UV-C, red light or sun light during 21 days of storage are presented in Fig. 3. The β-carotene contents of all tomatoes (untreated, red light and UV-C treated), except sun light- treated, did not change significantly (average of 12 ± 1 g/g) during 21 days of treatment and storage ($p > 0.05$). The β-carotene content of sun light- treated tomatoes declined significantly ($p < 0.05$) after 4 days of treatment and remained at this low level throughout the 21 days of treatment and storage (average of 2 ± 0.3 g/g). The results were similar to those reported by Thiagu and co-workers (Thiagu, Onwuzulu, & Ramana, 1993), who showed that β-carotene increased up to the light-pink stage and declined afterwards during full and over ripe stages of tomato ripening.

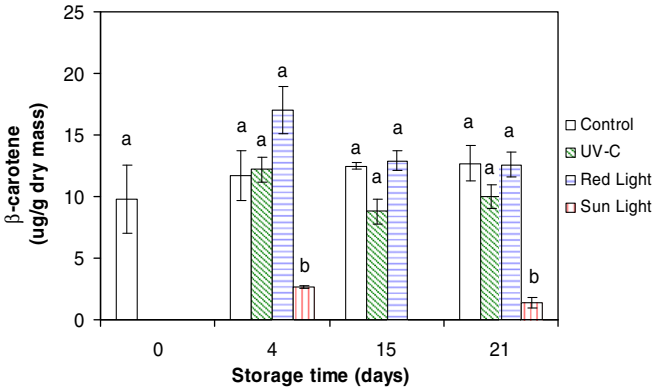

Fig. 3. Concentrations of β-carotene in untreated (Control), UV-C light-, red light- and sun light- treated tomatoes after 0, 4, 15 and 21 days of treatment and storage. The error bars represent standard errors of the means ($n = 3$).

3.5. The effects of light treatments on the physical properties of tomatoes during storage

3.5.1. Colour

The surface colour of the tomatoes, treated with UV-C, red light or sun light, was evaluated, and compared to that of the untreated tomatoes (Fig. 4). As storage progressed, the lightness factor, L^*, of untreated and treated tomatoes decreased during the initial 4 days of storage and then remained constant. There was no difference between untreated and light- treated tomatoes during the 21 days of treatment and storage.

Initially all fruits were mature-green at breaker colour. As expected, the a^* values of untreated and treated tomatoes increased dramatically during the initial 4 days of storage, followed by a slight increase from 4 to 21 days (Fig. 5). The a^* values of untreated, red light- and sun light- treated tomatoes were almost identical ($p > 0.05$), while those of UV-C treated tomatoes were significantly lower ($p < 0.05$) throughout storage. Maharaj's and co-workers (1999) reported that tomatoes treated with UV-C irradiation (3.7 kJ/m^2) had significantly retarded senescence, resulting in delayed colour development and fruit softening. They reported that the colour changes in tomatoes, as measured by the Hunter tristimulus a^* values, were significantly affected by UV-C irradiation and storage time.

The b^* values (Fig. 6) of untreated and treated tomatoes appeared to be greatest after 4 days of treatment, followed by a decrease during further treatment and storage. At day 4 the b^* values of tomatoes treated with red light or sun light were slightly greater than those of tomatoes that were untreated or treated with

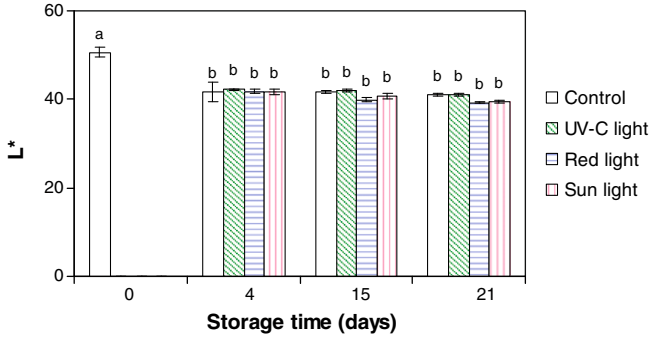

Fig. 4. Hunter L^* values of untreated (Control), UV-C light-, red light- and sun light-treated tomatoes after 0, 4, 15 and 21 days of treatment and storage. The error bars represent standard errors of the means ($n = 81$).

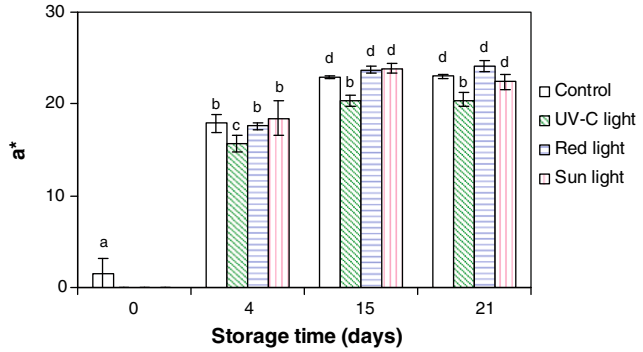

Fig. 5. Hunter a^* values of untreated (Control), UV-C light-, red light- and sun light-treated tomatoes after 0, 4, 15 and 21 days of treatment and storage. The error bars represent standard errors of the means ($n = 81$).

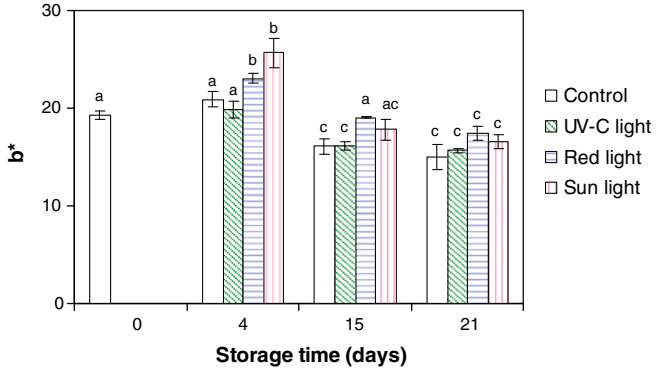

Fig. 6. Hunter b^* values of untreated (Control), UV-C light-, red light- and sun light-treated tomatoes after 0, 4, 15 and 21 days of treatment and storage. The error bars represent standard errors of the means ($n = 81$).

UV-C light ($p < 0.05$). At day 15, the b^* values of tomatoes treated with red light were slightly elevated ($p < 0.05$) when compared to untreated and UV-C treated tomatoes, while, at day 21, there was no significant difference ($P > 0.05$) between untreated and treated tomatoes.

The Hunter a^*/b^* ratio of tomato surface colour has been used as a reference parameter for red colour development in tomatoes (Arias, Lee, Logendra, & Janes, 2000). Fig. 7 shows the Hunter a^*/b^* ratios of untreated and treated tomatoes during 21 days of storage. During the initial 4 days of storage, the a^*/b^* ratio increased dramatically, about 10-fold from day 0 to day 4. Between days 4 and 15, the a^*/b^* ratios increased significantly ($p < 0.05$). However, there was no significant change observed thereafter (between days 15 and 21; $p > 0.05$). The a^*/b^* ratios of all light- treated samples were slightly lower than those of the untreated samples, but this differences was insignificant ($p > 0.05$).

3.5.2. Relationships between lycopene contents and colours of untreated and treated tomatoes

Changes in the surface a^*/b^* ratios were poorly related ($R^2 = 0.7219$) to changes in the lycopene contents of all tomatoes (untreated, red light, UV–C light and sun light) during 21 days of storage (Fig. 8). D'Souza, Singha, and Ingle (1992) and Arias et al. (2000) reported that Hunter L^*, a^* and b^* values of the tomato surface correlate with tomato lycopene concentration, and that the a^*/b^* ratios could be used to predict the lycopene content of various tomato cultivars at different ripening stages. Our observations were different from those reported by others, possibly due to light treatments influencing the lycopene content of tomatoes, without influencing tomato skin colour, for example Hunter a^*/b^* ratios. Our observations therefore suggest that there are limitations in using a^*/b^* ratios to predict the lycopene content of tomatoes.

3.5.3. Hardness and total soluble refractive solids

Hardness (as measured by penetrometer penetration force) of untreated and treated tomatoes gradually decreased during the 21 days of storage (Fig. 9). The hardness of tomatoes was not significantly affected by light treatments ($p > 0.05$), being almost identical to that of untreated tomatoes throughout the 15 days of storage. However, the hardness of UV-C and sun light- treated tomatoes was significantly decreased ($p < 0.05$) at 21 days of storage, compared to that of the untreated or red light- treated toma-

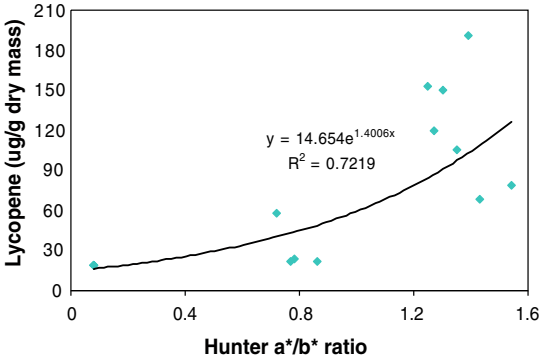

Fig. 8. Plot of Hunter a^*/b^* ratios versus tomato lycopene contents.

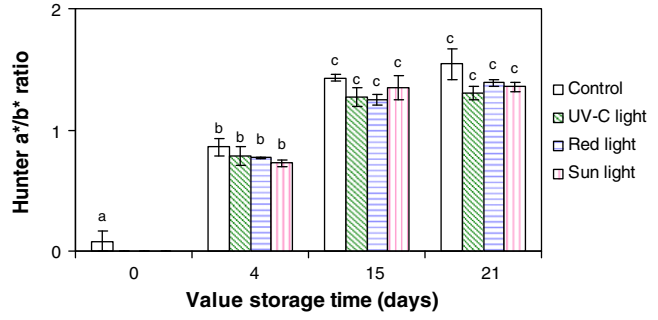

Fig. 7. Hunter's a^*/b^* values of untreated (Control), UV-C light-, red light- and sun light- treated tomatoes after 0, 4, 15 and 21 days of treatment and storage. The error bars represent standard errors of the means ($n = 81$).

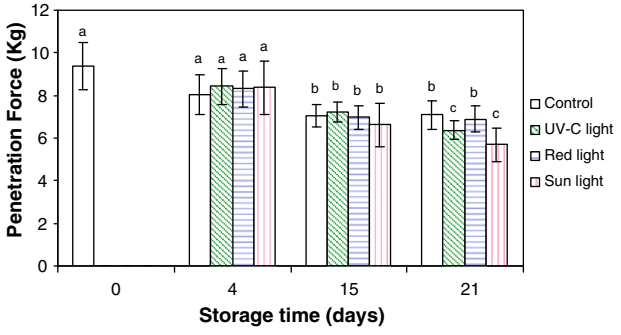

Fig. 9. Hardness (penetration force in Kg) of untreated (Control), UV-C light-, red light- and sun light- tomatoes after 0, 4, 15 and 21 days of treatment and storage. The error bars represent standard errors of the means ($n = 18$).

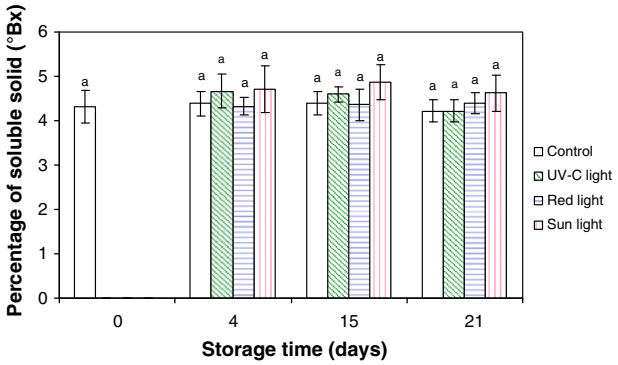

Fig. 10. Total soluble refractive solids (°Brix) of untreated (Control), UV-C light- and red light- treated tomatoes after 0, 4, 15 and 21 days of treatment and storage. The error bars represent standard errors of the means ($n = 18$).

toes. These results differ from studies examining the effect of UV-C treatment on tomato firmness (Barka et al., 2000; Stevens et al., 2004). These authors reported that tomato firmness was significantly increased by low-dose UV-C treatment, and that cell-wall degrading enzyme activities were also decreased. The difference in results may be due to the use of different equipment: we measured resistance to penetration with a probe, whereas tissue texture was measured with a texture analyser in the cited studies. However, our results are consistent with Alba's study (Alba et al., 2000), which showed that tomato pericarp softening was not influenced by red or red/far-red light treatments.

The total soluble refractive solids (°Brix) results are presented in Fig. 10. The °Brix values of all untreated and treated tomatoes remained constant during 21 days of treatment and storage and were not significantly influenced by any of the light treatments studied ($p > 0.05$). To the best of our knowledge, this is the first report showing that daily light treatment does not influence the total soluble refractive solids content of tomatoes.

4. Conclusion

It can be concluded from this study that daily light treatment of tomatoes enhances exocarp lycopene accumulation with minimal effects on the colour, hardness or °Brix during post-harvest storage. This indicates that light (especially at red and UV-C wavelengths) is a regulator of carotenoid synthesis and accumulation in tomatoes during post-harvest storage. It can also be concluded that tomato skin colour (as measured by the Hunter a^*/b^* ratio) is not always an accurate measurement of pericarp lycopene content. The findings of this study could be applied to increase the lycopene content of tomatoes without influencing β-carotene content or fruit texture.

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