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## Postharvest Biology and Technology



journal homepage: www.elsevier.com/locate/postharvbio

# Effects of postharvest light spectra on quality and health-related parameters in green *Asparagus officinalis* L.



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#### ARTICLE INFO

Article history: Received 11 May 2015 Received in revised form 1 October 2015 Accepted 19 October 2015 Available online xxx

Keywords: Green asparagus Light treatments Lignin Pigments Vitamin C

#### ABSTRACT

The monitoring of quality parameters in horticultural crops under artificial light during postharvest storage is important for controlling the shelf-life of the crops. In this work, white light, red light, blue light and dark conditions were used at various durations to evaluate the effects of different spectral properties of light on parameters related to physiological and biochemical processes in green asparagus, and on compounds related to human health. For this aim, the level of glucose, fructose and sucrose, as well as that of vitamin C and the levels of lignin, chlorophyll a and b, carotenoids and anthocyanins, were determined in apical and basal segments of the edible portion of green asparagus spears before and after light treatments. A dark control was stored at 4 °C. The irradiance levels of the light treatments were 100, 117 and  $116\,\mu\text{mol}\,\text{m}^{-2}\,\text{s}^{-1}$ , respectively for white, blue and red light. Before treatments, in the apical segments, the content of analysed components were higher than in the basal segments, except for soluble sugars and starch, of which the basal segment exhibited higher levels; these results exhibited different nutritional value of the two segments. After the light treatments, the analysed quality-related parameters were differently influenced in the apical and basal segments during postharvest storage. The increase in dry matter content in the apical segment after both white light and red light treatments was most likely attributable to the presence of physiological postharvest activity, rather than to increased transpiration. The results indicated that light with different spectral properties vs. dark-stored controls had small or no effects on the measured parameters. Both light and dark caused the starch levels to increase in both segments. A decrease in the sugar content in the basal part might be explained by translocation of hexoses from basal towards apical regions of the spear. White light primarily determined the lighin deposition in the apical part most likely due to the synergistic effect of red and blue light on lignin biosynthesis. The vitamin C, chlorophyll a and b and carotenoids decreased in light and dark treatments in both segments. Anthocyanins were induced by light in the basal part only, most so by blue light. © 2015 Elsevier B.V. All rights reserved.

#### 1. Introduction

In developed countries where a diet rich in protein and fat results in a sharp increase in obesity and related diseases, the consumption of plant foods with high fibre, vitamin and antioxidant content appears to be beneficial. For these reasons, the market demand for fresh vegetable products with high quality of nutrients and low calories has increased (Fuentes-Alventosa et al., 2009). Green asparagus (*Asparagus officinalis* L.) is one of the most widespread vegetables, due to its important dietary properties (Steinmetz and Potter, 1996). After asparagus harvest, the product is cut and subsequently tied in bundles; next, it is subjected to the canning process or, more frequently, used for fresh

http://dx.doi.org/10.1016/j.postharvbio.2015.10.010 0925-5214/© 2015 Elsevier B.V. All rights reserved. consumption. During postharvest, asparagus is subjected to minimal processing consisting of washing, cutting and packaging in plastic film. Therefore, the plant tissues of these minimally processed vegetables are physiologically and metabolically active (Heyes et al., 1998). This implies a quick perishability of the product and a short period of commercialization (shelf-life). Indeed, in white and green asparagus, several studies have sought to identify the best storage conditions and the reason for the short shelf-life (Siomos et al., 2000, 2001; Scheer et al., 2003; Albanese et al., 2007). The high respiratory rate that occurs after harvest (60 mg  $CO_2 Kg^{-1}h^{-1}$  at 5 °C) seems to be the primary cause of the perishability of fresh plant tissue (Papadopoulou et al., 2001). During storage, there is a loss of the bright green colour, soluble sugars (Lipton, 1990), vitamins, flavour, and aroma, and water loss causes the appearance of streaks along young stems (King et al., 1987; Zagory and Kader, 1988; Wills et al., 1999). During storage,

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other physical parameters such as O<sub>2</sub> and CO<sub>2</sub> partial pressure may influence the nutritional quality of the spears and their shelf-life (Everson et al., 1992; Siomos et al., 2000; Villanueva et al., 2005). In asparagus, changes in consistency, colour and nutrients occur especially in relation to minimal processing and storage. In addition, the increased lignification that occurs during storage is responsible for the progressive hardening of the shoots (Waldron and Selvendran, 1990; Rodríguez et al., 2004; Herppich and Huyskens-Keil, 2008). Previous studies have reported that in the first three days postharvest, an increase in the activity of the enzymes involved in the biosynthesis of lignin, namely, phenylalanine ammonia lyase, peroxidase together with cinnamyl alcohol dehydrogenase, and the augmentation of total phenols and lignin occurs in green asparagus (Liu and Jiang, 2006). Moreover, the persisting increase in lignin that mainly occurs in the tips of the shoots after five days of storage has led to the conclusion that this effect is due to postharvest physiological processes, rather than to responses induced by cut wounds (Liu and Jiang, 2006). The bright colour of green asparagus is due to chlorophyll, and this is the "conditio sine qua non" to marketing (Hutchings, 1999). The catabolism of chlorophyll, which occurs in plants during the phenomena of physiologically programmed or stress-induced senescence, is accelerated or decelerated by changes related to light or temperature (Lin et al., 2013). The healthy beneficial effects of asparagus may be also attributed to its higher contents of antioxidants, such as vitamin C (Tsushida et al., 1994). Asparagus has high content of vitamin C and its postharvest loss causes the decrease of its total antioxidant activity and its guality (Ournac, 1970; Rodkiewicz, 2008). Light influences the levels of ascorbate (AsA) (Siomos et al., 2000). In oat leaf segments, the intensity and quality of the light modifies the AsA content (Mastropasqua et al., 2012). Red and blue lights are reported to have the greatest impacts on plant growth and development because they are the major energy sources for photosynthesis, and regulate many responses in higher plants (Saebo et al., 1995; Wu et al., 2007; Xu et al., 2012; Lin et al., 2013). Red and blue lights are known to influence quality parameters of vegetables; in white asparagus, blue light preserves for a longer period the sensory quality, particularly colour and texture, than white lighting (Sanz et al., 2009). It has been reported that blue LED light was more effective for inducing carotenoid accumulation in the juice sacs of citrus fruit than red LED light (Zhang et al., 2012). However, little is known regarding the influence of lighting on the quality parameters of green asparagus, including the steps that occur postharvest until sale. In this study, in order to simulate the artificial fluorescent light in retail shops a particular white light, 76 NATURA<sup>®</sup> (Osram), was employed. Moreover, to evaluate the effects of different spectral properties of light on parameters related to physiological and biochemical processes, and on compounds related to human health, the effect of red light, blue light and darkness on the spears of green asparagus were also investigated. For this aim, the levels of soluble sugars (glucose, fructose and sucrose), vitamin C, lignin, chlorophyll a and b, carotenoids and anthocyanins, in the apical and basal segments of the edible part of green asparagus spears were determined.

#### 2. Materials and methods

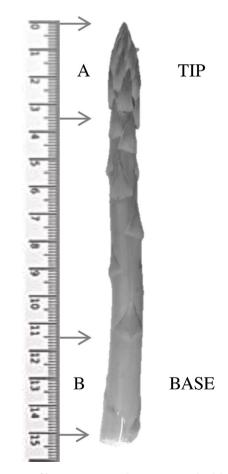
#### 2.1. Plant material

Fresh green asparagus spears (*A. officinalis* L. var. UC157) were obtained from commercial farm "Agritrade s.r.l." in Carapelle, Foggia province, Italy. The asparagus were grown on loam soil under commercial growing conditions. The spears were harvested in May 2014 (on days 2, 9, 16 and 23) between 7:30 and 9:00 AM, at  $17 \pm 2$  °C and 70% relative air humidity. The asparagus of 14 to

18 mm of diameter, cut at ground level, were washed with water and packaged in perforated polypropylene bags. The bags, each containing 12–15 spears (500 g for bag), were cooled at 4°C and transported at the local market within 2 h. Immediately, the raw material was purchased and selected based on the diameter  $(15 \pm 1 \text{ mm})$ , colour and no visible signs of injury. Spears were cut to 22 cm lengths. Before treatment, the material was washed with distilled water. The spears were placed in upright position in testtube racks, which were put in a water bath (vase) in such a way that only 2 cm of the basal end of the spear was submerged in the distilled water, and the water was replaced at time interval of 24 h. Three test-tube racks (24 spears per each rack) for a single treatment were used. After light and dark treatments, the test materials were obtained on days 3, 6 and 9. Segments of 3.5 cm in length of the apical and basal parts of the first 15 cm starting from tip of the spear, corresponding to the edible portion of asparagus, were used for the analyses (Fig. 1). Time zero samples were represented by the basal and apical segments of asparagus that were not subjected to the light or dark treatments.

#### 2.2. Light and dark treatments

The racks containing asparagus were incubated in a growth chamber at 16 °C with a relative air humidity of 78% under different light conditions with 8 h photoperiod or in continuous darkness at 4 °C (D). White light (WLN) was provided by a T8 NATURA<sup>®</sup> L36W/ 76 Osram 120 cm linear fluorescent tube, with a wavelenght range of 400–700 nm with three different peaks: blue at 440 nm, green at 550 nm and red at 630 nm. Blue light (BL) was provided by an



**Fig. 1.** Areas measured in green asparagus. Segments measuring 3.5 cm in length of the apical (A) and basal (B) segments of the first 15 cm starting from the tip of the spear, corresponding to the edible portion of asparagus, were utilized.

L36W/67 Osram 120 cm linear fluorescent tube, with a wavelength range of 400–520 nm and a peak at 440 nm. Red light (RL) was provided by an L36/60 Osram 120 cm linear fluorescent tube, with a wavelength range of 575-650 nm and a single peak at 610 nm. The full light intensities on the asparagus spear surface were 100, 117 and 116  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>, respectively for WLN, BL and RL. Irradiations were measured using a Delta OHM Photo-Radio meter, mod. HD2302.0 (Pordenone, Italy).

#### 2.3. Determination of dry matter content

Dry matter content of the apical and basal segments was measured at time zero and after 3, 6, and 9 days of various treatments. For the analysis, 5 g of fresh matter of each sample was dried at  $60 \,^{\circ}$ C in an oven until a constant weight was obtained. Dry matter content was calculated as a percentage of constant weight related to fresh matter.

#### 2.4. Vitamin C content

The fresh tissue was homogenized with two volumes of cold 5% (w/v) metaphosphoric acid in a porcelain mortar. The homogenate was centrifuged for 15 min at  $20,000 \times g$ , and the supernatant was collected for analysis of the ascorbate pool as described by Paciolla et al. (2008).

#### 2.5. Soluble carbohydrates and starch measurements

At various times (0, 3, 6 and 9 days), 1 g of apical and basal segments of fresh tissue from each treatment was cut up and analysed. After hot  $(80 \,^\circ\text{C})$  extraction with ethanol (95%) and centrifugation at  $4,500 \times g$  for 10 min, the levels of mono- and disaccharides (glucose, fructose and sucrose) were determined spectrophotometrically using a Megazyme kit (Sucrose/Fructose/ D-Glucose Assay Kit; K-SUFRG); the alcohol insoluble fraction (pellet) was used to determine the starch content using a Megazyme kit (Total Starch Assay Kit AA/AMG; K-TSTA).

#### 2.6. Determination of chlorophyll and carotenoid contents

The chlorophyll and carotenoid content was determined according to Lichtenthaler (1987), with some modifications; 500 mg of fresh material was homogenized with 1.5 mL of absolute acetone and 3 mg of NaCO<sub>3</sub> and centrifuged at  $20,200 \times g$  for 15 min. Absorbance of the supernatant was measured spectrophotometrically at 645 and 662 nm, respectively for chlorophyll and carotenoid content was calculated using Lichtenthaler's equations.

#### *2.7. Anthocyanin content*

Fresh tissue segments (700 mg) were cut up and incubated at 65 °C for 2 h with 4 mL of a solution containing 98% methanol and 0.24 M HCl. After centrifugation at  $4,500 \times g$  for 10 min, the anthocyanin content was measured spectrophotometrically at 530 nm and 657 nm. The formula  $A_{530} - 0.25 \times A_{657}$ , that corrects absorbance for chlorophyll degradation products, was utilized (Serafini-Fracassini et al., 2002).

#### 2.8. Lignin quantitative determination

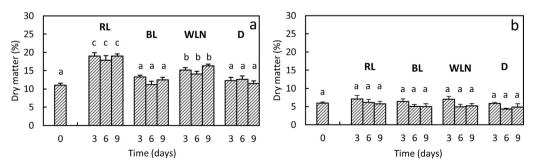
The lignin content was determined using the thioacidoglycolysis method, as described by Bruce and West (1989), with some modifications. At time zero and at various durations of the different treatments, 1 g of fresh tissue was homogenized with 95% ethanol and centrifuged at  $20,200 \times g$  for 15 min. The pellets were subsequently air-dried at 60°C and 10 mg of insoluble cell wall material was resuspended in 1.5 mL 2 M HCl and 250 µL thioglycolic acid. The mixture was heated for 4 h in boiling water and centrifuged at  $20,200 \times g$  for 30 min. The pellets were washed with bidistilled water, resuspended in 1.7 mL 0.5 M NaOH and left overnight at room temperature. After centrifugation, the supernatant containing thioglycolate lignin was precipitated by the addition of 400  $\mu L$  of 37% (W/V) HCl for 3 h at 4  $^\circ C.$  After centrifugation at  $20,000 \times g$  for 30 min, the pellets were resuspended in 1 mL 0.5 M NaOH. The absorbance was recorded at 280 nm, and the lignin content was expressed as  $mgg^{-1}$  fresh matter, using a linear calibration curve with commercial lignin alkali.

#### 2.9. Lignin qualitative analysis

Sections of  $20 \pm 2 \,\mu$ m thick of apical and basal segments were cut with a DSK-1000 vibratome (Dosaka,Kyoto, Japan) and stained with 70% (V/V) ethanol solubilized phloroglucin, and some drops of 37% (W/V) HCl was added to turn the contained lignin red. Sections were examined with a light microscope (DMLS, Leica, Wetzlar, Germany).

#### 2.10. Statistical analysis

The reported data are the average of at least three replications from four independent experiments. One-factor ANOVA was performed on the observed means of the compound content for each segment and the significance of the different treatments (red light, white light, blue light, dark) within each segment with respect to time zero was evaluated using Tukey's HSD test for multiple comparisons (P < 0.05).



**Fig. 2.** Dry matter content in the apical (a) and basal (b) segments at time zero and after 3, 6, and 9 d of the various treatments. Values represent the means of at least three replications from four independent experiments. Identical letters over the columns indicate non-significant differences between treatments within each segment (Tukey's HSD test, P < 0.05). Red light, RL; blue light, BL; white light, WLN; dark, D.

#### 3. Results

The spears appeared fresh-like for the entire duration of the experiment and no significant change in fresh matter weight, before and after light and dark treatments, was observed (data not shown).

#### 3.1. Effect of light spectral properties on dry matter content

At time zero, the apical and basal segments of the asparagus spears showed different dry matter content (DM); in the basal part, the DM was 40% lower than in the apical part (Fig. 2). Lighting caused a significant increase in DM in the apical segment (P < 0.05) after the red and white light treatments, whereas blue light and darkness did not have an effect (Fig. 2a). In the basal part, no significant changes occurred for any of treatments (Fig. 2b).

#### 3.2. Influence of light and dark on vitamin C

At time zero, the vitamin C (ASC plus DHA) was more than 2.5 times higher in the apical part than in the basal part (Fig. 3). When the apical and basal segments of asparagus were incubated in continuous darkness or in WLN, BL or RL, the vitamin C significantly (P < 0.05) decreased to a low and stable level over the different time treatments with respect to the time zero (Fig. 3). The DHA content was notably low in all treatments in both the apical and basal segments.

#### 3.3. Effects of light and dark on soluble sugars and starch

Fig. 4a and b shows the changes in D-fructose, D-glucose and sucrose content in both dark and the different light conditions. At time zero, sucrose is the main sugar that is present, followed by glucose and fructose; the basal portion contained a significantly (P < 0.05) higher total content of soluble sugars than the apical portion (more than six times higher). A significant (P < 0.05) increase in all sugars in the dark and light conditions in the apical part was observed. In the basal segment, a significant (P < 0.05) decrease in the sum of three sugars occurred during the light treatment with the lowest value obtained with BL. In darkness, after a significant decrease at three days, an increase at 6 and 9 days occurred.

The starch content was higher in the basal part than in the apical part (0.645 vs.  $0.135 \text{ mg g}^{-1}$ ) (Fig. 4c and d). During storage in both dark and light conditions, a significant (P < 0.05) increase in starch was observed. Blue light induced the highest increase at three days (more than 11 times compared to the time zero) in the apical portion. In the basal part, the starch content was increased, although a greater increase (in%) occurred in the apical part. In the basal part, the increase was higher with WLN than in BL and RL; in

the dark, the substantial increase at three days was transient because a decrease in starch occurred over the longer periods.

#### 3.4. Changes in chlorophylls after light and dark treatment

The total chlorophyll (chl a + chl b) at time zero was higher in the apical part of the spear (120 vs.  $68 \mu g g^{-1}$ ) (Fig. 5a). In both darkness and under various light conditions, a gradual and significant (P < 0.05) decrease in total chlorophyll content was observed compared to the time zero (Fig. 5b). On the third day, all treatments showed a high chl a/chl b ratio that was similar to the control, whereas at subsequent times, the chla/chl b ratio was decreased in both the apical and basal parts.

#### 3.5. Changes in carotenoids after light and dark treatment

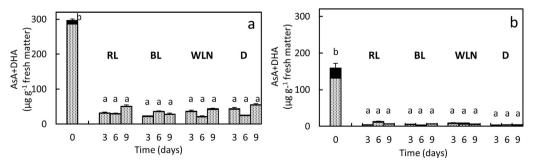
The levels of carotenoids at time zero were higher in the apical part than in the basal part (52 vs.  $33 \mu g g^{-1}$ ) (Fig. 6). During the time-course under different lighting conditions, a significant decrease in the content in the apical part occurred with lowest values after 9 days of treatment (Fig. 6a). In the basal part, no change was observed up to 6 days under both the light and dark conditions; thereafter, a significant (*P* < 0.05) decrease occurred (Fig. 6b).

#### 3.6. Changes in anthocyanins after light and dark treatment

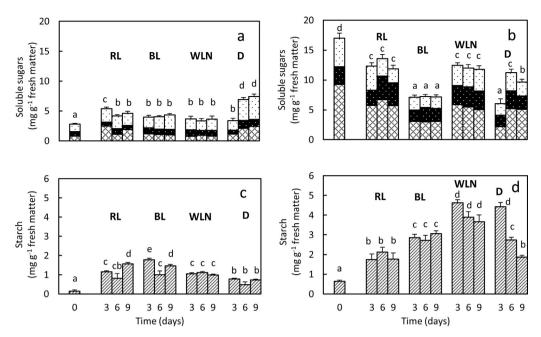
The content of anthocyanins at time zero was higher in the apical part than in the basal part (0.38 vs. 0.03 absorbance units  $g^{-1}$ ) (Fig. 7). During the time-course, in the apical part, the anthocyanin content was unchanged in both light and dark conditions (Fig. 7a). A significant (P < 0.05) increase in anthocyanin content was observed in the basal part in the light, with the highest increase occurring with BL, while no change with the darkness treatment occurred (Fig. 7b).

#### 3.7. Influence of light and dark on lignin content

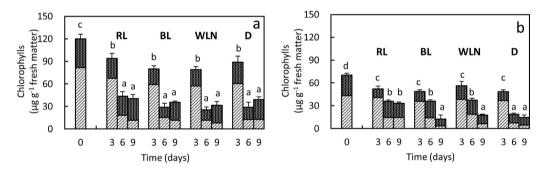
At time zero, lignin content was significant (P < 0.05) higher in the apical segment than in the basal segment (0.24 vs. 0.07 mg g<sup>-1</sup>) (Fig. 8). No change was observed at three days after the light and dark treatments in the apical part, whereas a significant (P < 0.05) gradual increase in lignin content on the 6th and 9th days occurred in relation to the control. The highest lignification occurred in WLN and the lowest in darkness (Fig. 8a). After the different lighting treatments, the basal part of the spears exhibited an increase in lignin level although this level was lower than in the apical part; compared to the control, there was no change in the lignin content after the darkness treatment (Fig. 8b).



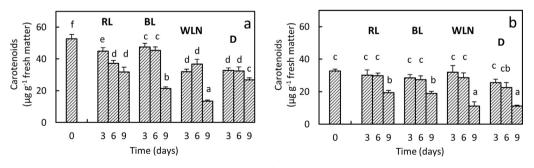
**Fig. 3.** Content of vitamin C–L-ascorbic acid (AsA, ) and L-dehydroascorbic acid (DHA, ) – in the apical (a) and basal (b) segments of green asparagus at time zero and after 3, 6, and 9 d of various treatments. Values represent the mean of at least three replications from four independent experiments. Identical letters over the columns indicate non-significant differences between treatments within each segment (Tukey's HSD test, *P*<0.05). Red light, RL; blue light, BL; white light, WLN; dark, D.



**Fig. 4.** Contents of soluble sugars – sucrose ( $\square$ ), p-fructose, ( $\square$ ), p-glucose ( $\square$ ) – and starch in apical (a and c) and basal segments (b and d) at time zero and after 3, 6, and 9 d of various treatments. Values represent the means of at least three replications from four independent experiments. Identical letters over the columns indicate non-significant differences between treatments within each segment (Tukey's HSD test, P < 0.05). Red light, RL; blue light, BL; white light, WLN; dark, D.



**Fig. 5.** Contents of chlorophylls a (2) and b (2) in the apical (a) and basal (b) segments at time zero and after 3, 6, and 9 d of various treatments. Values represent the means of at least three replications from four independent experiments. Identical letters over the columns indicate non-significant differences between treatments within each segment (Tukey's HSD test, *P* < 0.05). Red light, RL; blue light, BL; white light, WLN; dark, D.

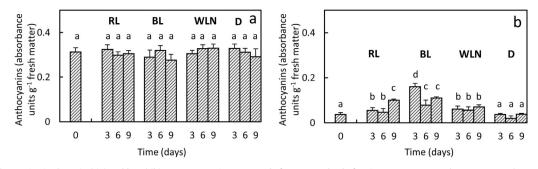


**Fig. 6.** Carotenoid content in the apical (a) and basal (b) segments at time zero and after 3, 6, and 9 d of various treatments. Values represent the means of at least three replications from four independent experiments. Identical letters over the columns indicate non-significant differences between treatments within each segment (Tukey's HSD test, P < 0.05). Red light, RL; blue light, BL; white light, WLN; dark, D.

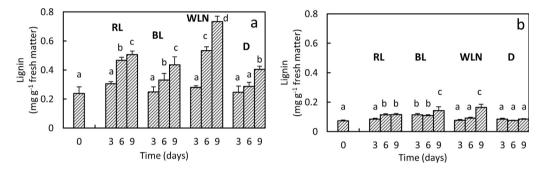
#### 3.8. Microscope analysis

In transversal sections of the stems at time zero (Fig. 9a), various vascular bundles with xylem characterized by large tracheas and lignified protoxylematic portions were observed.

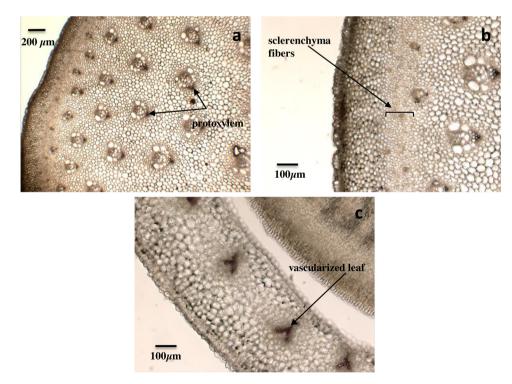
The sclerenchyma, made up of long fibres were a little lignified. In addition, in the basal segments, the sclerenchyma fibres were not completely lignified for all 15 cm of the segment length analysed (Fig. 9b). The microscope observations confirm the high lignin content in the apical part, characterized by the higher number of vascularized leaves (Fig. 9c).



**Fig. 7.** Level of anthocyanins in the apical (a) and basal (b) segments at time zero and after 3, 6, and 9 d of various treatments. Values represent the means of at least three replications from four independent experiments. Identical letters over the columns indicate non-significant differences between treatments within each segment (Tukey's HSD test, P < 0.05). Red light, RL; blue light, BL; white light, WLN; dark, D.



**Fig. 8.** Changes in lignin content in the apical (a) and basal (b) segments at time zero and after 3, 6, and 9 d of various treatments. Values represent the means of at least three replications from four independent experiments. Identical letters over the columns indicate non-significant differences between treatments within each segment (Tukey's HSD test, *P* < 0.05). Red light, RL; blue light, BL; white light, WLN; dark, D.



**Fig. 9.** (a) Transversal section of apical segment at time zero of *Asparagus officinalis*. Arrows indicate the protoxylem zone. (b) Transversal section of basal segment at time zero of *Asparagus officinalis*. Arrow indicates the presence of sclerenchyma fibers. (c) Transversal section of leaf bract in the apical segment of *Asparagus officinalis* time zero. Arrow indicates vascularized leaf containing lignin after reaction with phloroglucin.

#### 4. Discussion

The aim of this paper was to monitor the changes in important biochemical parameters in the apical and basal segments of the edible portions of green asparagus by simulating storage conditions (WLN, 16°C) that occur during the sale of this horticultural crop. In addition, the effects of WLN were compared with those of red and blue lights to identify the spectral ranges of light that could have an effect on physiological and biochemical alterations during accelerated senescence that occurs during storage after harvest. Indeed, it is well known that the environmental light spectrum properties strongly affect plant development and physiology. Red light is needed for the proper development of the photosynthetic apparatus and for starch accumulation in plants (Saebo et al., 1995). Blue light regulates many plant physiological events, such as stomatal opening, leaf expansion and biomass production (Xu et al., 2012; Lin et al., 2013). It has been reported that in spinach blue light in the presence of red light increases the photosynthetic capacity (Matsuda et al., 2007, 2008).

The physiological and compositional changes occurring during postharvest and storage reduce the spear quality: stiffening, loss of water and changes in carbohydrate, protein and amino acid patterns occur (Chang, 1987). The water represents the principal component of fresh matter in green asparagus. In order to prevent water loss and to eliminate water stress interference on the analysed parameters, the basal end of asparagus stalks was submerged in distilled water during the experiments both in light and dark conditions. No apparent changes in spears were detected during the entire period of the experiments (data not shown). The higher dry matter content at time zero in the apical segment (40% higher than the basal segment) is most likely due to high density of vascular apparatus present in the leaf bracts. Compared to the time zero and darkness, the significant increase in dry matter content observed in the apical part at various times in the presence of WLN and RL, might be due to longer opening time of stomatal aperture and consequently greater transpiration. In this respect, the leaves of Chinese kale and Roman lettuce under light maintained, respectively, 100% and 74% of open stoma contributing to the loss of water content (Noichinda et al., 2007; Martínez-Sánchez et al., 2011). However, because under our experimental conditions, water loss and dehydration were prevented by immersing the basal ends of the asparagus stalks in distilled water, the increase in dry matter content that occurred in the apical part after both the WLN and RL treatments might be due to persisting metabolic and physiological activity postharvest and "de novo" synthesis rather than to increased transpiration. Indeed, previous studies have reported the presence of active metabolism in harvested asparagus (Heyes et al., 1998) and photosynthesis and respiratory activity in lettuce leaves exposed to white fluorescent light (Martínez-Sánchez et al., 2011). Additionally, the apical segment of asparagus is subject to physiological changes more than the basal segment in terms of the presence of younger leaves and active meristems.

In plants, photosynthetic and non-photosynthetic tissues contain high levels of ascorbate, an important nutrient and antioxidant molecule. AsA is involved in many metabolic processes, such as growth and development, and in defence against abiotic and biotic stresses (Foyer et al., 1991; Córdoba and González-Reyes, 1994). Higher AsA content in the apical part compared to the basal part time at zero is most likely due to higher mitotic activity of the meristematic tissues of which the apical segment of asparagus is richer. A decrease in AsA has been reported in green asparagus postharvest during cold storage (Esteve et al., 1995). The properties and quantity of light influences biosynthesis; in particular, low irradiance induced an AsA increase in oat leaves (Mastropasqua et al., 2012). On the other hand, in Roman lettuce

leaves in darkness and under low light intensity, no AsA change occurred (Martínez-Sánchez et al., 2011). In contrast, under light and dark conditions, a significant AsA decrease in the apical and basal parts of the spear was observed. Most likely, the high irradiance, utilized in our experiment (see Section 2.2) was responsible for a lower level of AsA synthesis compared to AsA consumption.

Sugars are other nutrients of asparagus. The presence of soluble sugars in the plant tissue is equivalent to the available energy for the growth and development but also for molecular signals that are involved in important physiological and biochemical regulation processes. Glucose is involved in cell respiration, cell wall biosynthesis and photosynthesis; sucrose is involved in anthocyanin biosynthesis and storage, differentiation and antioxidative processes (Solfanelli et al., 2006; Nishizawa et al., 2008; Ritsema et al., 2009). The lower content of the soluble D-glucose, D-fructose and sucrose sugars at time zero in the apical part compared to the basal part is caused by their high consumption in cell growth processes. Indeed, a high activity of invertase in the apical segment of green asparagus spears has been reported (Benkeblia and Shiomi, 2009). The significant decrease in the total sugar content in the basal part under the dark and light conditions might be correlated with translocation of hexoses from the basal part towards the apical part, where a significant increase in the three sugars occurred. In addition, the higher sugar content occurring in the apical part of the spears under different types of lighting, may also be due to high photosynthesis level due to the presence of numerous leaf bracts. On the other hand, the highest levels of soluble sugars in the apical part after six days in the dark compared with the light treatments may be due to lower consumption. whereas in the basal segment, the recovery of the sugar levels after six days in the dark might be due, at least in part, to hydrolysis of starch, which is in effect decreased over longer periods. Low temperature and light might preserve the horticultural crop quality. In white asparagus stored in darkness, the consumption of soluble sugars at 20 °C was rapid, whereas at 5 and 10 °C, the consumption was low (Herppich and Huyskens-Keil, 2008). Moreover, the use of low-intensity light pulses is not sufficient for net synthesis of sugars in basil leaves (Costa et al., 2013). In the basal segments of green asparagus, the temperature of 16°C and 8h of continuous lighting used in our experiment, limited the decrease in the levels of soluble sugars, especially in the presence of red light and WLN.

At time zero, the starch content was higher in the basal segment of the spears probably due to a higher presence of parenchyma cells. After the light treatments, the different trends in the starch levels in the basal and apical segments, which represent evidence of the effects of red and blue light individually, are not predictive of their combined effects when they are simultaneously present, and the apical and basal segments are influenced in different ways. The highest and lowest levels, respectively, in the basal and apical segments under WLN (including both red and blue light) might be due to different interactive effects of both RL and BL and support the above statement. The starch levels were also augmented in dark. An increase in the dark was also reported in Chinese kale leaves (Noichinda et al., 2007). The higher starch increase in the apical part under different light spectra than in the dark can be correlated to photosynthesis that occurs during the 8-hour light cycle. In the basal part, the increase in starch content was correlated to a decrease in soluble sugars.

In green asparagus, the degradation of chlorophyll with consequent loss of its bright green colour and the increase in anthocyanins and carotenoids are related to altered sensorial aspects that occur in its short shelf-life (Chang, 1987). Indeed, in both apical and basal segments, a progressive loss of both a and b chlorophylls occurs in the light and the dark, which is already significant at three days. Interestingly, the decrease in the chl a/chl b ratio confirmed that the degradation process of chlorophyll requires the conversion of chl b to chl a (Tanaka et al., 1995; Scheumann et al., 1999). This trend was also reported in basil leaves treated with white low-intensity light pulses where the loss of chlorophylls, especially chl a, was observed after three days (Costa et al., 2013).

Light can also influence the synthesis and degradation of carotenoids, which are pigments that are components of the lightharvesting complex and play a protective role (Goodwin, 1980; Simkin et al., 2003). Their progressive decrease occurs over time and seems to occur early in the apical segment compared to the basal segment. Because the carotenoids protect the chlorophylls by photo-oxidation reactions, their low levels might promote those reactions.

The anthocyanins are pigments that are considered plant antioxidants and are important per human diet. The exposure of white asparagus to light accelerated the metabolic and physiological changes causing alterations in the apex colour and an increase in hardness (Sanz et al., 2009). However, due to anthocyanins, there is a colour change from white to red during the storage of white asparagus. This change is responsible for the spears having an unattractive appearance thus resulting in reduced appeal and purchasing by consumers (Sanz et al., 2009). In the basal part, the increase under BL, WLN and RL indicates that the anthocyanin synthesis is light-induced. Indeed, several plant species synthesize anthocyanins in the light, whereas others synthesize them in the dark, although their synthesis speed and total content is higher when exposed to light; higher synthesis of anthocyanins requires extended exposure to elevated irradiance in the spectral region between UV and far red (290–750 nm) with typical characteristics of photomorphogenetic processes (Mancinelli, 1985). In the basal part, the highest anthocyanin synthesis occurs in BL, and this is also reported in previous works in which BL is considered one of the most effective for regulating anthocyanin biosynthesis, which strongly induces their accumulation in arabidopsis seedlings (Chen et al., 2006), in apple fruit (Saure, 1990) and in postharvest strawberry fruit (Xu et al., 2014). On the contrary, no significant increase in anthocyanins was observed in the apical part exposed to different light spectral properties. The lack of the increase in anthocyanins in the apical segments could be due to the achievement of the maximum biosynthetic capability of these pigments already at the time zero. Additionally, the persistence of anthocyanins in both the apical and basal parts in darkness is most likely due to light exposure of asparagus during the various commercial and manipulation steps preceding the experimental dark incubation. On the other hand, it is confirmed that in white asparagus, three hours of light is sufficient to promote the anthocyanin synthesis (Siomos et al., 2001, 1995).

Mechanical properties are responsible for texture of the spears and are correlated with different components of the cell wall (Rodríguez et al., 2004), such as lignin. Lignin is an important component of cell walls. The phenylalanine ammonia lyase, cinnamyl alcohol dehydrogenase and peroxidase enzymes are involved in the lignification process in plants; in this respect, their involvement in the lignification of harvested asparagus has been demonstrated (Hennion et al., 1992). In green asparagus spears, microscopic observations at time zero revealed the presence of large xylematic tracheas in both apical and basal segments together with the existence of lignified protoxylematic portions and fibres of the sclerenchyma that are responsible for its hardening; additionally, in the apical segment, more lignin was observed due to the presence of leaf bracts that showed vascularization. After three days, the increase in lignin in the light was most likely due to higher lignification of the leaf bracts. In particular, when individually applied, red and blue lights stimulated the lignin deposition whereas when simultaneously present, such as in WLN, their synergistic additive effect on lignin biosynthesis was induced.

#### 5. Conclusions

Our results show that illumination with light of different spectral ranges induced small or no changes in most of the measured parameters in basal and apical parts of asparagus spears. compared with dark-stored controls. Treatment with white light at room temperature, which simulates the conditions of storage occurring during the sale of this horticultural crop, and with RL induced an increase in dry matter in the apical part probably as a consequence of persistent and efficient photosynthesis. However, this increase in dry matter (from about 110 to  $190 \text{ mg g}^{-1}$  fresh matter) cannot be explained only by the increases in sugars, starch and lignin; other components such as, proteins, cell wall polysaccharides, lipids and mineral salts may contribute to the observed increase. In the basal part of the spears, the dry matter was unchanged after light treatment; the starch increase can be in part due to the decrease in soluble sugars and the increase in lignin was very small. White, red and blue light stimulated deposition of anthocyanins in the basal part and lignin predominantly in the apical part. White light induced the highest level of lignin, showing a probable synergistic effect of red and blue light in the apical part. However, white, red or blue light did not preserve the contents of chlorophylls, carotenoids and vitamin C. Instead, these compounds dropped to low levels during storage similarly to the dark-stored control samples. On this basis, we can state that during postharvest storage, exposure to light can differently influence various qualityrelated parameters, and the changes can differ between apical and basal parts.

Moreover, our data highlight that in green asparagus, the apical segment of the edible part showed higher levels of ascorbate, chlorophyll a and b, carotenoids, anthocyanins and lignin. Conversely, the basal segment had higher contents of soluble sugars and starch. For this reason, both the apical and basal segments are important for human dietary reasons.

#### Acknowledgment

This work was supported by Research Funds CUP H95E10000710005 of the University of Bari "Aldo Moro", Italy.

#### References

- Albanese, D., Russo, L., Cinquanta, L., Brasiello, A., Di Matteo, M., 2007. Physical and chemical changes in minimally processed green asparagus during cold storage. Food Chem. 101, 274–280.
- Benkeblia, N., Shiomi, N., 2009. Variation of fructooligosaccharide contents, invertase, 1-SST, 1-FFT, activities in green asparagus spears stored under different temperature regimes. Fresh Produce 3, 44–50.
- Bruce, R., West, C., 1989. Elicitation of lignin biosynthesis and isoperoxidase activity by pectic fragments in suspension cultures of castor bean. Plant Physiol. 91, 889–897.
- Chang, D.N., 1987. Asparagus. In: Weichmann, J. (Ed.), Postharvest Physiology of Vegetables. Marcel Dekker, New York, pp. 523–525.
- Chen, Z., Zhang, H., Jablonowski, D., Zhou, X., Ren, X., Hong, X., Schaffrath, R., Zhu, J. K., Gong, Z., 2006. Mutations in ABO1/ELO2, a subunit of holo-Elongator, increase abscisic acid sensitivity and drought tolerance in *Arabidopsis thaliana*. Mol. Cell. Biol. 26, 6902–6912.
- Córdoba, F., González-Reyes, J.A., 1994. Ascorbate and plant cell growth. J. Bioenergy Biomembr. 26 (4), 399–405.
- Costa, L., Montanoa, Y.M., Carrióna, C., Rolnya, N., Guiameta, J.J., 2013. Application of low intensity light pulses to delay postharvest senescence of *Ocimum basilicum* leaves. Postharvest Biol. Technol. 86, 181–191.
- Esteve, M.J., Farre, R., Frigola, A., Clemente, G., 1995. Changes in ascorbic acid content of green asparagus during the harvesting period and storage. J. Agric. Food Chem. 43 (8), 2058–2061.
- Everson, H.P., Waldron, K.W., Geeson, J.D., Browne, K.M., 1992. Effect of modified atmospheres on textural and cell wall changes of asparagus during shelf-life. Int. J. Food Sci. Technol. 27, 187–199.

- Foyer, C.H., Lelandais, M., Edwards, E.A., Mullineaux, P.M., 1991. Active Oxygen/ Oxidative Stress and Plant Metabolism. In: Pell, E., Steffen, K. (Eds.), American Society of Plant Physiologists, Rockville, pp. 131–144.
- Fuentes-Alventosa, J.M., Rodríguez-Gutiérrez, G., Jaramillo-Carmona, S., Espejo-Calvo, J.A., Rodríguez-Arcos, R., Fernández-Bolaños, J., Guillén-Bejarano, R., Jiménez-Araujo, A., 2009. Effect of extraction method on chemical composition and functional characteristics of high dietary fibre powders obtained from asparagus by-products. Food Chem. 113, 665–671.
- Goodwin, T.W., 1980. 2nd ed. The Biochemistry of the Carotenoids, vol. 1. Chapman & Hall, London.
- Hennion, S., Little, C.H.A., Hartmann, C., 1992. Activities of enzymes involved in lignifications during the postharvest storage of etiolated asparagus spears. Physiol. Plant. 86, 474–478.
- Herppich, W.B., Huyskens-Keil, S., 2008. Cell wall biochemistry and biomechanics of harvested white asparagus shoots as affected by temperature. Ann. Appl. Biol. 152, 377–388.
- Heyes, J.A., Burton, V.M., de Vré, L.A., 1998. Cellular physiology of textural changes in harvested asparagus. Acta Hortic. 464, 455–460.
- Hutchings, J.B., 1999. Food Colour and Appearance. Aspen Publishers, Gaithersburg, MD, USA, pp. 610.
- King, G.A., Henderson, K.G., Lill, R.E., 1987. Sensory analysis of stored asparagus. Sci. Hortic. 31, 11–16.
- Lichtenthaler, H.K., 1987. Chlorophylls and carotenoids: pigments of photosynthetic biomembranes. Methods Enzymol. 148, 350–382.
- Lin, K.H., Huang, M.Y., Huang, W.D., Hsu, M.H., Yang, Z.W., Yang, C.M., 2013. The effects of red, blue, and white light-emitting diodes on the growth, development, and edible quality of hydroponically grown lettuce (*Lactuca sativa* L. var. capitata). Sci. Hortic. 150, 86–91.
- Lipton, W.J., 1990. Postharvest biology of fresh asparagus. Hort. Rev. 12, 69–115.
- Liu, Z.Y., Jiang, W.B., 2006. Lignin deposition and effect of postharvest treatment on lignification of green asparagus (*Asparagus officinalisL*.). Plant Growth Regul. 48, 187–193.
- Mancinelli, A.L., 1985. Light dependent anthocyanin synthesis: A model system for the study of plant photomorphogenesis. Bot. Rev. 51 (1), 107–157.
- Martínez-Sánchez, A., Tudela, J.A., Luna, C., Allende, A., Gil, M.I., 2011. Low oxygen levels and light exposure affect quality of fresh-cut Romaine lettuce. Postharvest Biol. Technol. 59, 34–42.

Mastropasqua, L., Borraccino, G., Bianco, L., Paciolla, C., 2012. Light qualities and dose influence ascorbate pool size in detached oat leaves. Plant Sci. 183, 57–64.

- Matsuda, R., Ohashi-Kaneko, K., Fujiwara, K., Kurata, K., 2007. Analysis of the relationship between blue-light photon flux density and the photosynthetic properties of spinach (*Spinacia oleracea* L.) leaves with regard to the acclimation of photosynthesis to growth irradiance. Soil Sci. Plant Nutr. 53, 459–465.
- Matsuda, R., Ohashi-Kaneko, K., Fujiwara, K., Kurata, K., 2008. Effects of blue light deficiency on acclimation of light energy partitioning in PSII and CO<sub>2</sub> assimilation capacity to high irradiance in spinach leaves. Plant Cell Physiol. 49, 664–670.
- Nishizawa, A., Yukinori, Y., Shigeoka, S., 2008. Galactinol and raffinose as a novel function to protect plants from oxidative damage. Plant Physiol, 147, 1251–1263.
- Noichinda, S., Bodhipadma, K., Mahamontri, C., Narongruk, T., Ketsa, S., 2007. Light during storage prevents loss of ascorbic acid, and increases glucose and fructose levels in Chinese kale (*Brassica oleracea* var. alboglabra). Postharvest Biol. Technol. 44, 312–315.
- Ournac, A., 1970. Vitamin C, thiamin and riboflavin in asparagus. Changes during growth:storage and cooking. Bull. Soc. Sci. Hyg. Aliment. 58, 164–172.
- Paciolla, C., Ippolito, M.P., Logrieco, A., Mulé, G., Dipierro, N., Dipierro, S., 2008. A different trend of antioxidant defence responses makes tomato plants less susceptible to beauvericin than to T-2 mycotoxin phytotoxicity. Physiol. Mol. Plant P 72, 3–9.
- Paradopoulou, P.P., Siomos, A.S., Dogras, C.C., 2001. Metabolism of etiolated and green asparagus before and after harvest. J. Hortic. Sci. Biotechnol. 76, 497–500. Rodkiewicz, T., 2008. Vitamin C changes and total antioxidant activity of fresh and
- stored green Asparagus spears. Acta Hortic. (ISHS) 776, 235–238.
- Ritsema, T., Brodmann, D., Diks, S.H., Bos, C.L., Nagaraj, V., Pieterse, C.M.J., Boller, T., Wiemken, A., Peppelenbosch, M.P., 2009. Are small GTPases signal hubs in sugar-mediated induction of fructan biosynthesis? PLoS One 4 (8), e6605.

- Rodríguez, R., Jaramillo, S., Heredia, A., Guillén, R., Jiménez, A., Fernández-Bolaños, J., 2004. Mechanical properties of white and green asparagus: changes related to modifications of cell wall components. J. Sci. Food Agric. 84, 1478–1486.
- Saebo, A., Krekling, T., Appelgren, M., 1995. Light quality affects photosynthesis and leaf anatomy of birch plantlets in vitro. Plant Cell Tissue Org. Cult. 41, 177–185.
- Sanz, S., Olarte, C., Ayala, F., Ech'avarri, J.F., 2009. Evolution of quality characteristics of minimally processed asparagus during storage in different lighting conditions. J. Food Sci. 74 (6), 296–302.
- Saure, M.C., 1990. External control of anthocyanin formation in apple. Sci. Hortic. 42, 181–218.
- Scheer, C.H., Schonhof, I., Bruckner, B., Schreiner, M., 2003. Influence of a shorttermstorage on quality determining product characteristics of white prepared asparagus. Acta Hortic. 604, 437–441.
- Scheumann, V., Schoch, S., Rüdiger, W., 1999. Chlorophyll b reduction during senescence of barley seedlings. Planta 209, 364–370.
- Serafini-Fracassini, D., Del Duca, S., Monti, F., Poli, F., Sacchetti, G., Bregoli, A.M., Biondi, S., Della Mea, M., 2002. Transglutaminase activity during senescence and programmed cell death in the corolla of tobacco (*Nicotiana tabacum*) flowers. Cell Death Differ. 9, 309–321.
- Simkin, A.J., Zhu, C., Kuntz, M., Sandmann, G., 2003. Light-dark regulation of carotenoid biosynthesis in pepper (*Capsicum annuum*) leaves. J. Plant Physiol. 160, 439–443.
- Siomos, A., Sfakiotakis, E.M., Dogras, C.C., 2000. Modified atmosphere packaging of white asparagus spears: composition, color and textural quality responses to temperature and light. Sci. Hortic. 84, 1–13.
- Siomos, A.S., Dogras, C., Sfakiotakis, E., 1995. Effect of temperature and light during storage on the composition and color of white asparagus spears. Acta Hortic. 379, 359–366.
- Siomos, A.S., Dogras, C.C., Sfakiotakis, E.M., 2001. Color development in harvested white asparagus spears in relation to carbon dioxide and oxygen concentration. Postharvest Biol. Technol. 23, 209–214.
- Solfanelli, C., Poggi, A., Loreti, E., Alpi, A., Perata, P., 2006. Sucrose-specific induction of the anthocyanin biosynthetic pathway in Arabidopsis. Plant Physiol. 140, 637–646.
- Steinmetz, K.A., Potter, J.D., 1996. Vegetables, fruit, and cancer prevention: A review. J. Am. Diet. Assoc. 10, 1027–1039.
- Tanaka, A., Tanaka, Y., Takabe, T., Tsuji, H., 1995. Calcium induced accumulation of apoproteins of the light-harvesting chlorophyll a/b protein complex in cucumber cotyledons in the dark. Plant Sci. 105, 189–194.
- Tsushida, T., Suzuki, M., Kurogi, M., 1994. Evaluation of antioxidant activity of vegetable extracts and determination of some active compounds. J. Jpn. Soc. Food Sci. Technol. 41 (9), 611–618.
- Villanueva, M.J., Tenorio, M.D., Sagardoy, M., Redondo, A., Saco, M.D., 2005. Physical, chemical, histological and microbiological changes in fresh green asparagus (Asparagus officinalis, L.) stored in modified atmosphere packaging. Food Chem. 91, 609–619.
- Waldron, K.W., Selvendran, R.R., 1990. Effect of maturation and storage on asparagus (Asparagus officinalis) cell wall composition. Physiol. Plant 80, 576–583.
- Wills, R., McGlasson, B., Graham, D., Joyce, D., 1999. Introduccion alla fisiology manipulacion postcosecha de frutas, hortalizas y plantas ornamentales. Zaragoza, España: Edit. Acribia 240.
- Wu, M.C., Hou, C.Y., Jiang, C.M., Wang, Y.T., Wang, C.Y., Chen, H.H., Chang, H.M., 2007. A novel approach of LED light radiation improves the antioxidant activity of pea seedlings. Food Chem. 101, 1753–1758.
- Xu, H.L., Xu, Q.C., Li, F.L., Feng, Y.Z., Qin, F.F., Fang, W., 2012. Applications of xerophytophysiology in plant production—LED blue light as a stimulus improved the tomato crop. Sci. Hortic. 148, 190–196.
- Xu, F., Cao, S., Shi, L., Chen, W., Su, X., Yang, Z., 2014. Blue light irradiation affects anthocyanin content and enzyme activities involved in postharvest strawberry fruit. J. Agric. Food Chem. 62, 4778–4783.
- Zagory, D., Kader, A.A., 1988. Modified atmosphere packaging of fresh produce. Food Technol. 9, 70–77.
- Zhang, L.C., Ma, G., Kato, M., Yamawaki, K., Takagi, T., Kiriiwa, Y., Ikoma, Y., Matsumoto, H., Yoshioka, T., Nesumi, H., 2012. Regulation of carotenoid accumulationand the expression of carotenoid metabolic genes in citrus juice sacs in vitro. J. Exp. Bot. 63, 871–886.