



Effects of alginate edible coating on preserving fruit quality in four plum cultivars during postharvest storage

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ABSTRACT

Four plum (*Prunus salicina* Lindl.) cultivars (“Blackamber”, “Larry Ann”, “Golden Globe” and “Songold”), were treated with 1 or 3% alginate as an edible coating before storage. Analytical determinations were made after 7, 14, 21, 28 and 35 days at 2 °C and after a 3 day period at 20 °C (shelf-life). Both treatments were effective in inhibiting ethylene production for all cultivars, especially when 3% alginate was used. The changes in fruit quality parameters related to plum postharvest ripening, such as weight and acidity losses, softening and colour changes, were significantly delayed by the use of both edible coatings. The delay of the ripening process was also related to lower anthocyanin and carotenoid accumulation. Overall results suggest that these treatments could increase the plum storage period with optimum quality, 2 weeks for “Larry Ann” and “Songold” and 3 weeks for “Blackamber” and “Golden Globe” more than controls.

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1. Introduction

Plums, in general, are very appreciated by consumers, the degree of acceptance depending on organoleptic properties such as colour, texture, flavour and aroma, which vary among cultivars and production areas and from season to season (Crisosto et al., 2004; Díaz-Mula et al., 2008). In addition, plum consumption has beneficial health effects due to their antioxidant compounds such as vitamin C, carotenoids, polyphenols and anthocyanins (Cevallos-Casals et al., 2006; Vizzotto et al., 2007; Díaz-Mula et al., 2008). Given the perishable nature of plum fruit, the use of cold storage is necessary to delay changes related to ripening, such as ethylene production, respiration rate, softening, pigment changes, weight and decrease in acidity (Guerra and Casquero, 2008; Díaz-Mula et al., 2009). However, cold storage is not enough to preserve plum quality at optimum levels during transportation and marketing, often leading to the incidence of severe chilling injury symptoms, evident as mealiness, translucency, and flesh reddening. Therefore, appropriate postharvest technologies combined with cold storage are needed. In this sense, several treatments prior to cold storage, such as calcium, heat, polyamines or 1-methylcyclopropene (Valero and Serrano, 2010), as well as the use of modified atmosphere packaging (Díaz-Mula et al., 2011a), have been reported

to maintain plum quality for longer periods than low temperature storage alone.

On the other hand, edible coatings are also effective as postharvest treatments to preserve fruit quality, with the additional benefit of reducing the volume of non-biodegradable packaging materials (Olivas et al., 2008; Campos et al., 2011). Thus, maintenance of fruit quality has been achieved by using some edible coatings, such as chitosan in peach (Ruoyi et al., 2005), methylcellulose in apricot (Ayranci and Tunc, 2004), and hydroxypropylmethylcellulose (Navarro-Tarazaga et al., 2008), versasheen (Eum et al., 2009) and whey protein, in plum (Reinoso et al., 2008). Such edible coatings act as physical barriers on the fruit surface and decrease its permeability to O₂, CO₂ and water vapour, leading to reductions in respiration rate and transpiration and to retardation of the natural physiological ripening process.

Alginate is a natural polysaccharide extracted from brown sea algae (Phaeophyceae), and it is composed of two uronic acids: β-D-mannuronic acid and α-L-guluronic acid. Alginate is known as a hydrophilic biopolymer that has a coating function because of its well-studied unique colloidal properties, which include its use for thickening, suspension forming, gel forming and emulsion stabilising (Acevedo et al., 2012). Alginate-based edible coatings have been effective in maintaining postharvest quality of tomato (Zapata et al., 2008), peach (Maftoonazad et al., 2008) and sweet cherry (Díaz-Mula et al., 2012). However, no information is available on the use of alginate as an edible coating to preserve plum fruit quality, which is the main objective of this paper, using four plum cultivars.

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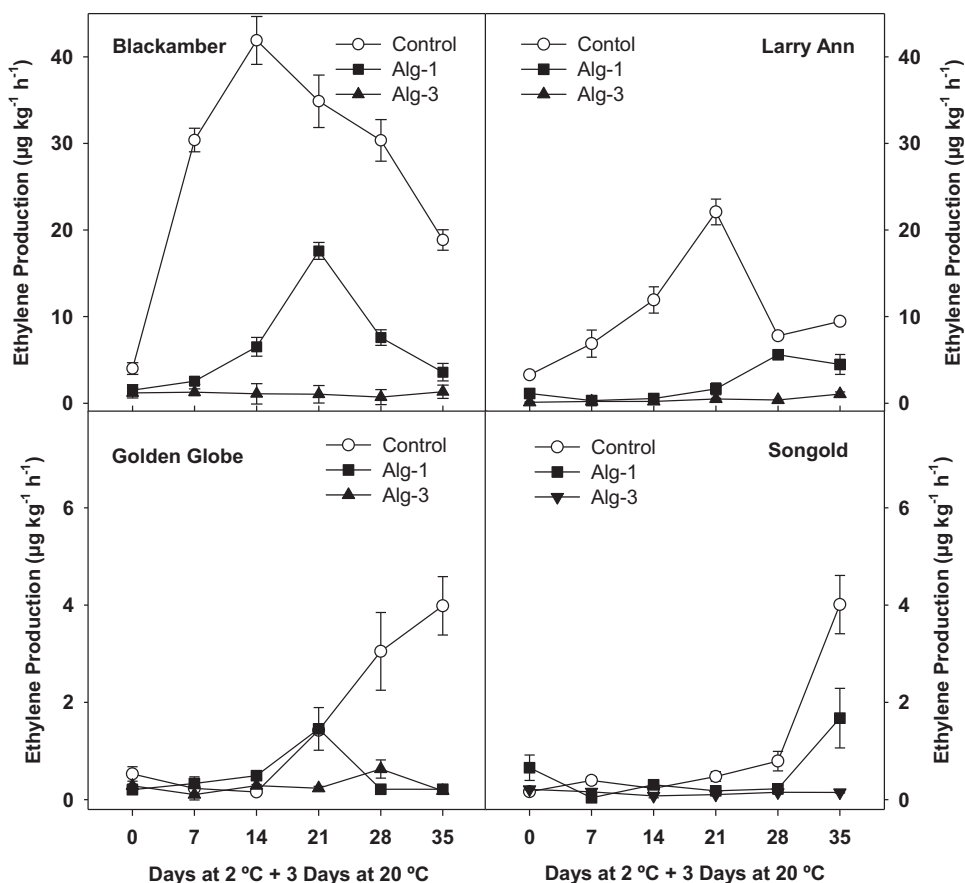


Fig. 1. Ethylene production after 0, 7, 14, 21, 28 and 35 days of storage at 2 °C + 3 days at 20 °C of control and coated plums with alginate at 1 (Alg-1) and 3% (Alg-3). Data are the mean \pm SE. LSD = 1.41, 0.73, 0.27, and 0.22 for “Blackamber”, “Larry Ann”, “Golden Globe” and “Songold”, respectively.

2. Materials and methods

2.1. Fruit material and experimental design

“Blackamber”, “Larry Ann”, “Golden Globe” and “Songold” plum (*Prunus salicina* Lindl.) fruit were harvested in 2010 from a commercial plot (Finca Los Frutales, Villena, Alicante, Spain) at the commercial ripening stage, and transported immediately to the laboratory. Then, 102 homogeneous lots (based on colour and size) of ten fruit each were assembled at random for each cultivar. Three lots were used to determine the fruit properties at harvest (day 0) and the 99 remaining were split into three groups for the following treatments in triplicate: 0% (control), 1% and 3% (w/v) alginate coating. Three replicates of 11 lots were used for each treatment. After treatments, fruit were dried for 30 min using an air-flow heater at 25 °C. After drying, the lots were weighed, and then one lot from each replicate and treatment was kept at 20 °C for 3 days, and the remaining lots were stored in a controlled chamber at 2 °C and relative humidity of 90%. After 7, 14, 21, 28 and 35 days of cold storage at 2 °C, two lots from each replicate and treatment were taken at random; one was analysed immediately, and the other analysed after 3 days at 20 °C with RH of 65% (shelf-life, SL). Alginate (alginic acid sodium salt from brown algae purchased from Sigma, Madrid, Spain) was prepared according to a previous paper (Zapata et al., 2008) at two concentrations, 1% (Alg-1) and 3% (Alg-3), w/v, dissolved in hot water (45 °C) with continuous shaking until the solution became clear. After cooling to 20 °C, glycerol at 20% (v/v) was added as a plasticiser, and treatments were performed by dipping the fruit twice in fresh coating solutions for 1 min to ensure the

uniformity of the coating of the whole surface. Control fruit were dipped in distilled water.

2.2. Analytical determinations

Weight loss of each lot was calculated as % with respect to the weight on day 0. Ethylene production was measured by placing each lot of ten fruit in a 3 L glass jar hermetically sealed with a rubber stopper for 30 min. 1 mL of the atmosphere was withdrawn with a gas syringe, and the ethylene was quantified using a Shimadzu™ GC-2010 gas chromatograph (Kyoto, Japan), equipped with a flame ionisation detector (FID) and a 3 m stainless steel column with an inner diameter of 3.5 mm containing activated alumina of 80/100 mesh. The carrier gas was helium, column temperature was 90 °C, and injector and detector temperatures were 150 °C. Results were the mean \pm SE of determinations for three replicates of ten fruit and expressed as $\mu\text{g kg}^{-1} \text{h}^{-1}$.

Colour parameters (L^* , a^* and b^*) were determined individually in the 10 fruit of each replicate, using the CIE Lab System in a Minolta colorimeter CR200 model (Minolta Camera Co., Japan). Two determinations were performed in opposite side of each fruit, the chroma index ($\text{Chroma} = (a^2 + b^2)^{1/2}$) was calculated and results were the mean \pm SE. Fruit firmness was measured on the fruit shoulder using a flat steel plate coupled with a texture analyser (TX-XT2i, Stable Microsystems, UK) interfaced to a personal computer. A bevelled holder prevented bruising of the opposite side. For each fruit, the diameter was measured and then a force that achieved a 3% deformation of the fruit diameter was applied. Results were expressed as the force-deformation (N mm^{-1}) and were the mean \pm SE. After

Table 1

Effects of alginate edible coatings, on weight loss (%) with respect to weight at day 0 of plum cultivars (BA = “Blackamber”, LA = “Larry Ann”, GG = “Golden Globe”, SG = “Songold”). Data are the mean \pm SE of three replicates.

Cultivar	35 days at 2 °C			35 days at 2 °C + 3 days at 20 °C		
	Control	Alg-1	Alg-3	Control	Alg-1	Alg-3
BA	10.8 \pm 0.5a	8.4 \pm 0.4b	5.8 \pm 0.6b	16.2 \pm 0.7a	13.4 \pm 0.5b	10.2 \pm 0.5c
LA	6.1 \pm 0.4a	5.2 \pm 0.3b	5.0 \pm 0.1b	8.7 \pm 0.3a	7.2 \pm 0.3b	6.5 \pm 0.5b
GG	5.5 \pm 0.4a	4.4 \pm 0.3b	3.9 \pm 0.5b	7.8 \pm 0.5a	6.9 \pm 0.2b	6.1 \pm 0.2c
SG	7.4 \pm 0.4a	6.7 \pm 0.3a	6.1 \pm 0.4b	10.2 \pm 0.5a	8.2 \pm 0.4b	7.4 \pm 0.3c

*Different letters show significant differences ($p < 0.05$) between control and alginate-treated plums (Alg-1 = alginate at 1% and Alg-3 = alginate at 3%).

firmness determination, the fruit from each replicate were manually peeled and the flesh tissue was cut in small pieces and used to determine total soluble solids concentration (TSS) and titratable acidity (TA) in duplicate. The peels were immediately frozen and ground in liquid N₂ and stored at -40°C until analysis of anthocyanins and total carotenoids were carried out in duplicate.

TSS were measured with a digital refractometer Atago PR-101 (Atago Co. Ltd., Tokyo, Japan) at 20°C and expressed as % ($^{\circ}\text{Brix}$). Total acidity (TA) was determined by automatic titration (785 DMP Titrino, Metrohm) with 0.1 N NaOH up to pH 8.1, using 1 mL of diluted juice in 25 mL of distilled H₂O, and results expressed as g malic acid equivalent per 100 g⁻¹ fresh weight. 1 g of skin tissue was homogenised in 5 mL of 50 mmol L⁻¹ phosphate buffer (pH 7.8) and 3 mL of ethyl acetate and then centrifuged at $10,000 \times g$ for 15 min at 4°C . The lipophilic upper fraction was used to estimate total carotenoids as previously described (Díaz-Mula et al., 2011b), by reading the absorbance at 450 nm in a UNICAM Helios α spectrophotometer (Cambridge, UK). Results were expressed as mg of β -carotene equivalent kg⁻¹ fresh weight, taking into account the $\epsilon_{\text{cm}}^{1\%} = 2560$ and were the mean \pm SE.

Total anthocyanins were extracted by homogenising 1 g of peel tissue with 10 mL of water/methanol (2:8) containing 2 mM NaF (to inactivate polyphenol oxidase activity and prevent phenolic degradation) and centrifuged at $10,000 \times g$ for 15 min at 4°C . 1 mL from the supernatant was filtered through a 0.45 μm Millipore filter and then injected into a Hewlett-Packard HPLC series 1100 equipped with a C18 Supelco column (Supelcogel C-610H, 30 cm \times 7.8 mm, Supelco Park, Bellefonte, USA) and detected by absorbance at 510 or 340 nm. The peaks were eluted by a gradient using the following mobile phases: 95% water + 5% methanol (A); 88% water + 12% MeOH (B); 20% water + 80% MeOH (C); and MeOH (D) at a rate of 1 mL min⁻¹. Peaks were identified using authentic standards by comparing the retention times and peak spectral analysis. The anthocyanin standards (cyaniding 3-glucoside and cyanidin 3-rutinoside) were provided by Dr. García-Viguera. Results were expressed as mg kg⁻¹ fresh weight, and were the mean \pm SE.

2.3. Statistical analysis

Experimental data were subjected to ANOVA analysis. Sources of variation were treatment and storage. The overall least significant differences (Fisher's LSD procedure, $p < 0.05$) were calculated and used to detect significant differences among treatments and storage time. All analyses were performed with SPSS software package v. 11.0 for Windows (SPSS, 2001).

3. Results and discussion

3.1. Ethylene production

Ethylene production rate at harvest was 0.25–0.50 $\mu\text{g kg}^{-1} \text{h}^{-1}$ for all plum cultivars and remained at these low levels during storage at 2°C , without significant differences between control and treated plums (data not shown). However, when plums were

transferred to 20°C , ethylene production increased, and the four plum cultivars showed the typical climacteric ripening pattern of plum fruit, although some plum cultivars have a suppressed-climacteric phenotype, such as “Shiro”, “Rubyred” (Abdi et al., 1997), “Golden Japan” (Zuzunaga et al., 2001), “Angeleno” (Candan et al., 2008), “Black Diamond” (Serrano et al., 2003), and “TC Sun” (Díaz-Mula et al., 2009). However, edible coatings significantly inhibited ethylene production for all plum cultivars, especially in Alg-3 treated plums, in which the climacteric peak of ethylene production was highly inhibited (Fig. 1). The barrier properties of the edible coatings also reduce the selective permeability to O₂ and CO₂ of the fruit surface leading to an increase in CO₂ concentration in the fruit tissues and a decrease in O₂ concentration (de Wild et al., 2005), which could be responsible for the reduced ethylene production rate in the alginate-coated plums. Accordingly, in tomato fruit the ethylene production was decreased by alginate and zein coatings, with a concomitant reduction in 1-aminocyclopropane-1-carboxylic acid (ACC) concentration (Zapata et al., 2008), due to the effect of elevated CO₂ concentration on inhibiting the conversion of S-adenosylmethionine (SAM) to ACC by ACC synthase (de Wild et al., 2005).

3.2. Weight loss and fruit quality parameters

Weight loss increased during cold storage for all plum cultivars, with final values after 35 days ranging from $\approx 11\%$ in “Blackamber” to 5.5% in “Golden Globe”, which reached higher values when fruit were transferred to 20°C (Table 1). However, alginate treatments significantly decreased weight loss for all plum cultivars, the effect being higher for Alg-3 treatment. Weight loss of fruit is due to the transpiration process which is determined by the gradient of water vapour pressure between the fruit and the surrounding

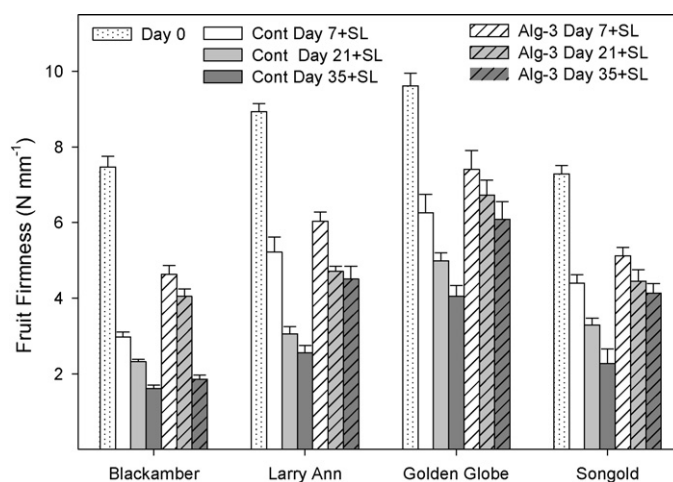


Fig. 2. Firmness values at harvest (day 0) and after 7, 21 and 35 days of storage at 2°C + 3 days at 20°C (SL) of control (Cont) and coated plums with alginate at 3% (Alg-3). Data are the mean \pm SE. LSD = 0.12.

air. Transpiration is usually reduced by both epidermal cell layer and cuticle. Thus, as fruit surface/volume ratio and epidermis and cuticle structure are different among plum cultivars, differences in weight loss were observed in control fruit depending on cultivar. In addition, edible coatings act as an extra layer which also coats the stomata leading to a decrease in transpiration and in turn, to a reduction in weight loss, this being the primary beneficial effect of edible coatings, as has been demonstrated in a wide range of fruit including apricot, pepper, peach, sweet cherry, and litch (Ayranç and Tunc, 2004; Dong et al., 2004; Maftoonzad et al., 2008; Díaz-Mula et al., 2012). Moreover, differences in the ability to reduce weight loss are attributed to the different water vapour permeability of the polysaccharides used in the formulation of the edible coating (Vargas et al., 2008). In this sense, the addition of glycerol as plasticiser to the coating gave good results in terms of reducing plum weight loss, according to previous reports on tomato (Zapata et al., 2008), apple (Moldão-Martins et al., 2003) and strawberry (García et al., 1998), in which the addition of 20% glycerol to 1% alginate or starch was sufficient to achieve the maximum reduction of moisture loss.

Fruit firmness at harvest was $\approx 7.5, 9.0, 9.6, 7.3 \text{ N mm}^{-1}$ and continuously decreased with the progress of storage at 2°C , reaching final values of $\approx 2.8, 4.0, 5.4,$ and 3.5 N mm^{-1} for “Blackamber”, “Larry Ann”, “Golden Globe” and “Songold”, respectively (data not shown). This softening process was faster when plums were transferred to 20°C after cold storage, since after 7 days of cold storage + SL, firmness levels in control plums were close to those found after 35 days of cold storage. However, alginate edible coatings slowed down the softening process for all plum cultivars, either during cold storage or subsequent SL, the effect being significantly

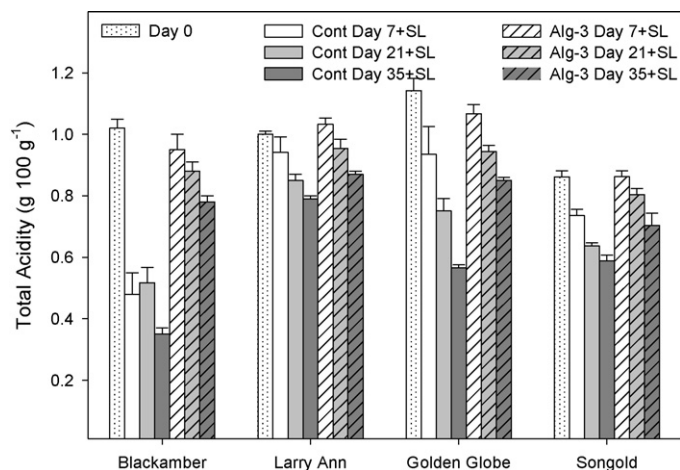


Fig. 3. Total acidity at harvest (day 0) and after 7, 21 and 35 days of storage at 2°C + 3 days at 20°C (SL) of control (Cont) and coated plums with alginate at 3% (Alg-3). Data are the mean \pm SE. LSD = 0.01.

higher for the Alg-3 than the Alg-1 treatment for most sampling dates (data not shown). In order to simplify the results presentation, only firmness data at harvest and after 7, 21 and 35 days of cold storage + SL of control and Alg-3 treated plums are provided (Fig. 2). The effect of alginate edible coating on delaying the softening process was also evident after the SL period for all plum cultivars, which showed firmness levels after 35 days at 2°C + SL similar to those found in control fruit after just 7 days at 2°C + SL. Changes in cell wall composition, especially cell wall mechanical

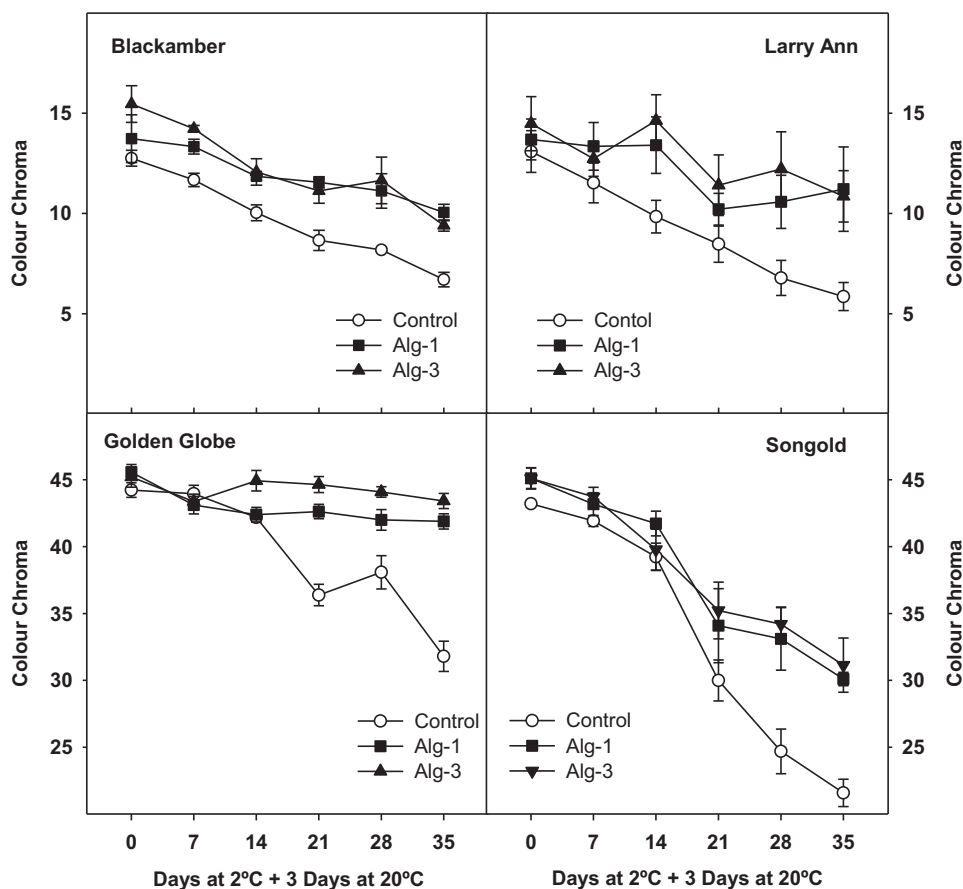


Fig. 4. Colour chroma after 0, 7, 14, 21, 28 and 35 days of storage at 2°C + 3 days at 20°C of control and coated plums with alginate at 1 (Alg-1) and 3% (alg-3). Data are the mean \pm SE. LSD = 0.58, 1.19, 0.68, and 1.38 for “Blackamber”, “Larry Ann”, “Golden Globe” and “Sungold”, respectively.

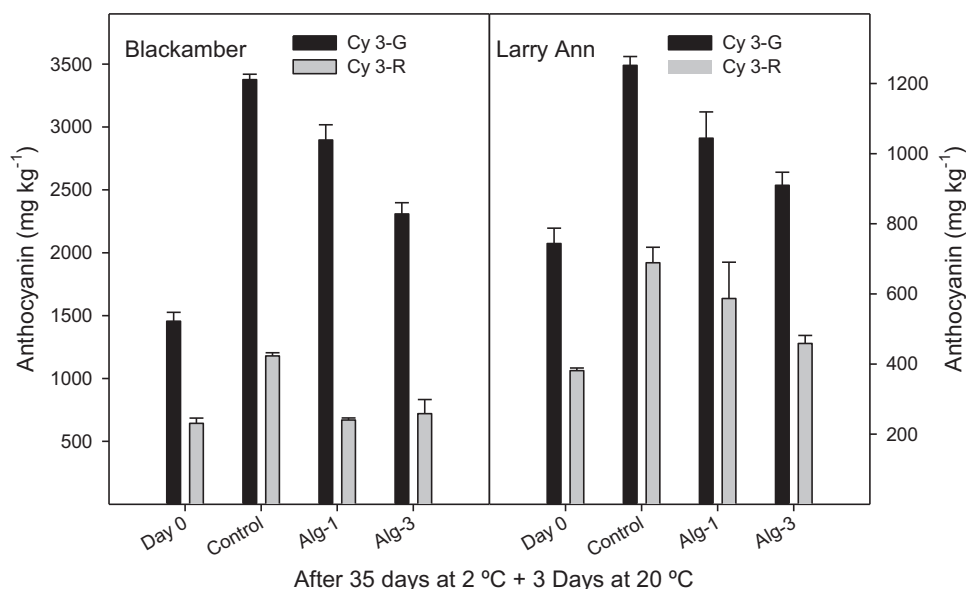


Fig. 5. Cyanidin 3-glucoside (Cy 3-G) and cyaniding 3-rutinoside (Cy 3-R) in the skin of “Blackamber” and “Larry Ann” plums, at harvest (day 0) and after 35 days at 2 °C + 3 days at 20 °C, in control and alginate-coated plums at 1 (Alg-1) and 3% (Alg-3). Data are the mean \pm SE. LSD = 72.97 and 51.87 for “Blackamber” and “Larry Ann”, respectively.

strength and cell-to-cell adhesion are the most important factors contributing to firmness losses during fruit on-tree ripening or after harvesting, the activity of cell wall hydrolysing enzymes being enhanced by ethylene in climacteric fruit (Valero and Serrano, 2010). In plums, the main cell wall-degrading enzymes are polygalacturonase, pectin methylesterase, 1,4- β -D-glucanase/glucosidase and β -galactosidase (Manganaris et al., 2008) Thus, the inhibition of ethylene production observed in alginate-coated plums could be responsible for their lower softening process with respect to control fruit.

Decrease in total acidity is also typical during postharvest storage of fleshy fruit, including plums, and has been attributed to the use of organic acids as substrates for the respiratory metabolism in detached fruit (Díaz-Mula et al., 2009; Valero and Serrano, 2010). However, acidity losses were different depending on plum cultivar, since after 7 days at 2 °C + SL losses in control fruit were \approx 53%

in “Blackamber”, \approx 15% in “Golden Globe” and “Songold” and just \approx 6% in “Larry Ann”. Alginate edible coating delayed acidity losses in all plum cultivars, with losses in Alg-3 treated plums \approx 25% in “Blackamber” and “Golden Globe”, \approx 20% in “Songold” and \approx 13% in “Larry Ann” after 35 days at 2 °C + SL (Fig. 3). With the Alg-1 edible coating, acidity losses were also delayed with respect to control plums (data not shown), and in general no significant differences were observed between Alg-1 and Alg-3 treatments. During cold storage, acidity losses were very low in control plums and no significant effect attributed to edible coating was observed (data not shown).

Skin colour also changed during storage in all plum cultivars, to dark purple in “Blackamber” and “Larry Ann” and to deep yellow in “Golden Globe” and “Songold”, as could be inferred from the changes in the Chroma index (Fig. 4). Colour changes were delayed by Alg-1 and Alg-3 edible coatings, without significant

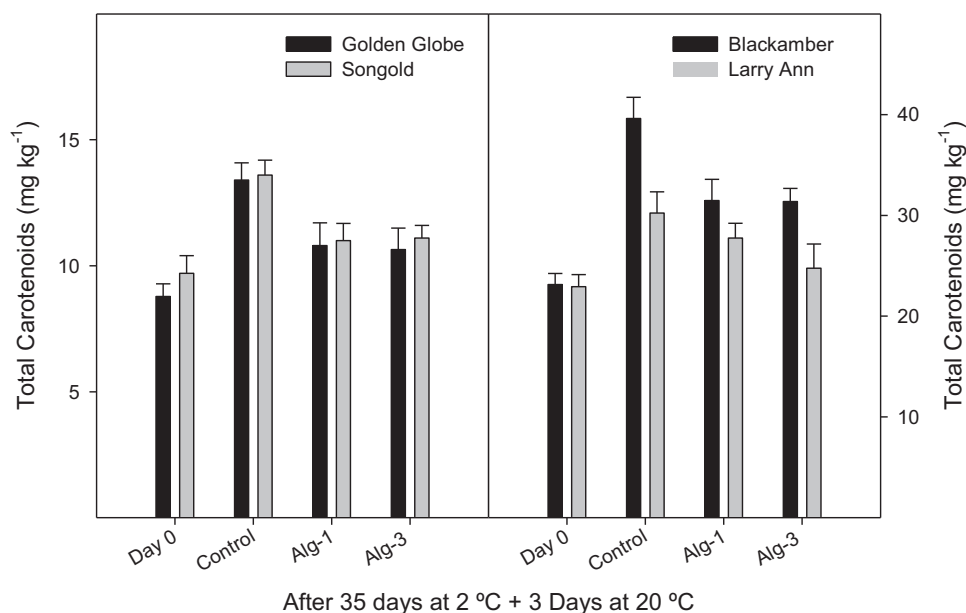


Fig. 6. Total carotenoids in the skin of yellow (“Golden Globe” and “Songold”) and purple (“Blackamber” and “Larry Ann”) plums, at harvest (day 0) and after 35 days at 2 °C + 3 days at 20 °C, in control and alginate-coated plums at 1 (Alg-1) and 3% (Alg-3). Data are the mean \pm SE. LSD = 0.67 and 1.74 for yellow (“Golden Globe” and “Songold”) and purple plums (“Blackamber” and “Larry Ann”), respectively.

differences between them, except for “Golden Globe” plum, where Alg-3 was the most effective. During cold storage, colour changes were lower than after the SL periods and the effect of alginate edible coating was less evident (data not shown). Skin colour in purple plums is due to anthocyanins, the main anthocyanin quantified in “Blackamber” and “Larry Ann” plums being cyanidin 3-glucoside followed by cyanidin 3-rutinoside, as previously reported for these and other purple plum cultivars (Tomás-Barberán et al., 2001; Wu and Prior, 2005; Díaz-Mula et al., 2011b). Both anthocyanins increased with the progress of cold storage, these increases being lower in alginate-coated plums than in control ones (Fig. 5). Thus, alginate treatment delayed colour change in purple plum cultivars by retarding the anthocyanin synthesis associated to the postharvest ripening process (Serrano et al., 2009; Díaz-Mula et al., 2012). Similarly, strawberry treated with alginate at 2% showed lower increases in total anthocyanin than controls (Fan et al., 2009). Total carotenoids also increased in plum skin during storage, in both purple and yellow cultivars, the increase was delayed by alginate treatments (Fig. 6). In the purple plum cultivars “Blackamber” and “Larry Ann”, skin colour is due to anthocyanin pigments, although carotenoids are also present and even at higher concentration than in the yellow-coloured cultivars “Golden Globe” and “Songold”. Thus, given the antioxidant properties of both pigment groups (Cevallos-Casals et al., 2006; Vizzotto et al., 2007; Díaz-Mula et al., 2008, 2011b), purple cultivars could have higher health beneficial effects than yellow ones.

4. Conclusions

Alginate treatment could be used as natural postharvest treatments in plum cultivars with the aim to delay the postharvest ripening process and to maintain fruit quality. Alginate treatment at 1% retarded the onset of the ethylene climacteric peak in coated plums, which was highly inhibited in those plums treated with alginate at 3%, both treatments being effective on delaying weight and acidity losses, softening and colour changes. In terms of storability, the treatment with alginate coatings could increase the plum storage period with optimal quality by 2 weeks for “Larry Ann” and “Songold” and 3 weeks for “Blackamber” and “Golden Globe” more than controls.

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