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Effect of oxalic acid treatment on maintaining pomegranate fruit quality and antioxidant potential

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Abstract. The aim of this work was to study the effect of pre-storage oxalic acid treatment at three concentrations (2, 4, and 6mM) on pomegranate CI and fruit quality after long-term storage at 2ºC. The CI symptoms were significantly reduced by oxalic acid treatment, especially for the 6 mM concentration. In addition, control pomegranates showed a significant reduction in the content of total phenolics and ascorbic acid as well as in total antioxidant activity (TAA), in both hydrophilic (H-TAA) and lipophilic (L-TAA) fractions after long term storage. However, the application of oxalic acid led to lower losses of total phenolics and significant increase in both ascorbic acid content and H-TAA, whereas L-TAA remained unaffected. Thus, oxalic acid could be a natural promising postharvest treatment to alleviate CI and increase antioxidant potential of pomegranate, by enhancing or maintaining the bioactive compounds.

Keywords. Fruit quality – Organic acids – Phenolic compounds – Antioxidant activity.

I – Introduction

During postharvest storage of pomegranate important quality loss occurs due to several physiological and enzymatic disorders, such as desiccation, browning symptoms in both peel and arils, and losses of firmness, aril color, vitamin C and acidity, leading to reduction of acceptability in terms of freshness, juiciness and taste (Kader, 2006). Refrigeration is effective to prolong the storability of pomegranate, but the fruit are susceptible to chilling injury (CI). Oxalic acid is a natural organic anion which is found ubiquitously in plant species and plays different roles in controlling fruit tissue browning, inducing systemic resistance, retarding fruit ripening and controlling decay. In addition, the endogenous concentration of oxalic acid in many plant foods has been considered as a natural antioxidant by suppressing in vitro lipid peroxidation (Kayashima and Katayama, 2002). Thus, the aim of this work was to study the effect of pre-storage oxalic acid treatment at 3 concentrations on pomegranate fruit quality and on the contents of ascorbic acid, total phenolic compounds, and anthocyanin profile, and total antioxidant activity after prolonged postharvest storage.

II – Materials and methods

Fully mature pomegranate fruits (*Punica granatum* L. cv. Mollar de Elche) were picked in a commercial orchard in Elche (Alicante), 75 homogeneous fruits in size and color were selected and sorted at random in lots of 5 fruits. Three lots were used to analyze fruit properties at harvest (day 0) and the remained lots were randomized and divided into 4 groups for the following treatments in triplicate (each replicate contained 5 individual fruits): control (distilled water at 25℃) and oxalic acid treatment at 3 concentrations (2, 4 and 6 mM). Oxalic acid was purchased from Sigma (Sigma Aldrich, Madrid, Spain 97% purity). Fruits were dipped in 20-L solution for 10 minutes. Following treatments, fruits were placed on Kraft paper and allowed to dry for 20 hours at 20°C. Then, they were transferred to a temperature-controlled chamber at 2°C, in permanent darkness and with relative humidity of 90%. After 84 days, fruits were sampled for the following determinations: electrolyte leakage (EL), total soluble solids (TSS), total acidity (TA), fruit firmness, weight loss, respiration rate, chilling injury index (CI), total antioxidant activity, in both hydrophilic (H-TAA) and lipophilic fractions (L-TAA), total phenolic, total anthocyanin and ascorbic acid, according to Mirdehghan *et al.* (2006; 2007a; b).

III – Results and discussion

Control pomegranates exhibited CI symptoms manifested as browning, pitting and dehydration of the husk surface, reaching a CI index value of 1.64±0.24, that is CI symptoms affected from 25-50% of the fruit surface. In addition, increases in respiration rate, weight loss and electrolyte leakage (EL) were observed after 84 days of cold storage. However, these changes were significantly ($P<0.05$) reduced in oxalic acid-treated fruits, the efficacy being higher for the 6 mM oxalic acid applied dose (data not shown). This effect could be attributed to the inhibition of polyphenoloxidase and peroxidase activites, and to its role as antisenescente agent. Along prolonged storage, control pomegranates showed significant reduction in the content of total phenolics, ascorbic acid and TAA, in both hydrophilic (H-TAA) and lipophilic (L-TAA) fractions (Table 1). The application of oxalic acid led to lower losses of total phenolics and to a significant increase in ascorbic acid and TAA after 84 days of cold storage, while L-TAA remained without significant changes. A significant increase in total anthocyanins was also observed during storage, which was higher in OA treated fruits, specialy at 6 mM dose (data not shown). The increase in anthocyanin concentration in control fruits is in agreement with previous reports (Artés *et al.*, 2000; Mirdehghan et al, 2006), which was associated with the advancement of the ripening process during postharvest storage. Although no information is available on the role of oxalic acid on anthocyanin biosynthesis, exogenous oxalic acid could act as elicitor of anthocyanin synthesis. Five individual anthocyanins were identified HPLC-DAD analysis: cyanidin-3-glucoside (Cy-3-Gluc), delphinidin-3-glucoside (Dp-3-Gluc), pelargonidin-3-glucoside (Pg-3-Gluc), cyanidin-3,5-diglucoside (Cy-3,5-Digluc), and pelargonidin-3,5-diglucoside (Pg-3,5-Digluc), the predominant being Cy-3-Gluc, according to previous rapports in other cultivars, such as ‘Assaria’ (Miguel *et al.*, 2004) and Primosole (D’Aquino *et al.*, 2010). Contrarily, in the sour cultivars, total anthocyanins decreased during cold storage and the predominant anthocyanin was Cy-3,5-Digluc followed by Dp-3,5-Digluc and Cy-3-Gluc (Alighourchi *et al.*, 2008).

Table 1. Bioactive compounds (total phenolics, total anthocyanins and ascorbic acid) and antioxidant activity (hydrophilic H-TAA and lipophilic L-TAA) in pomegranate arils at harvest (day 0) and after 84 days of storage in control and oxalic acid treated fruits. mg 100 g^{-1}

Parameter	Day 0	Day 84			
		Control	Oxalic 2 mM	Oxalic 4 mM	Oxalic 6 mM
Phenolics	252.4 a	167.6 b	192.8 c	212.5 d	182.1 c
Anthocyanins	59.0 a	109.5 b	107.1 b	122.5 c	135.9 d
Ascorbic acid	36.6 a	33.8 b	39.9 c	40.4 c	44.3 d
H-TAA	81.9 a	73.3 b	122.8 c	116.5 c	112.9 c
L-TAA	15.9 a	12.6 b	16.6 a	17.6 a	16.9 a

For each parameter means followed by different letter are significantly different at $P<0.05$ from day 0 to day 84 and among treatments.

During postharvest storage of fruit and vegetables, loss of health-beneficial compounds has been reported. However, the application of oxalic acid induced beneficial effects in terms of maintaining or increasing the pomegranate potential antioxidant activity during postharvest storage. The mechanism by which oxalic acid increased the bioactive compounds and antioxidant properties is not well known, although oxalic acid has been reported as a natural antioxidant by suppressing lipid peroxidation in vitro in a concentration-dependent manner and reducing the ascorbic acid oxidation (Kayashima and Katayama, 2002). Thus, oxalic acid treatment could be considered as a natural postharvest tool to reduce CI and maintain pomegranate fruit quality and its health beneficial effects.

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