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Research Article

Effects of thermal treatments on chilling injury and shelf life time of *Citrus reticulata* Blanco[1]

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ABSTRACT

The 'Arrayana' mandarin (*Citrus reticulata* Blanco) is considered the variety most widely grown in Colombia. Despite being a non-climacteric fruit, it has a short postharvest life. In order to evaluate the effects of thermal treatments on the chilling injury and shelf life time of this cultivar, ripe fruits were selected and submitted to the following treatments: non-treated fruit (control); hot water at 50 ºC for 5 min; hot water at 53 ºC for 3 min; intermittent warming in 8-day cycles at 2 ºC + 1 day at 18 ºC; and intermittent warming in 12-day cycles at 2 ºC + 1 day at 18 ºC. The fruits were stored at 2 ºC for 40 days and then left for one week at room temperature. The thermal treatments decreased the chilling injury in the mandarin fruits during the shelf life time, with the intermittent warming treatments being the most favourable ones (mainly in cycles of 12 days at 2 ºC + 1 day at 18 ºC), because, besides decreasing the chilling injuries and electrolytes leakage, they promoted a higher carotenoid biosynthesis and epidermis colouring, without negative effects on the fruit internal quality.

KEYWORDS: Mandarin, epidermis color, postharvest quality.

RESUMO

Efeitos de tratamentos térmicos na injúria por frio e no período de vida útil de *Citrus reticulata* Blanco

A tangerina 'Arrayana' (*Citrus reticulata* Blanco) é considerada a variedade mais cultivada na Colômbia. Apesar de ser uma fruta não climatérica, apresenta curta vida pós-colheita. Para avaliar os efeitos do tratamento térmico na injúria por frio e na vida útil dessa cultivar, frutos maduros foram selecionados e submetidos aos seguintes tratamentos: fruto não tratado (controle); água quente a 50 ºC por 5 min; água quente a 53 ºC por 3 min; aquecimento intermitente em ciclos de 8 dias a 2 ºC + 1 dia a 18 ºC; e aquecimento intermitente em ciclos de 12 dias a 2 ºC + 1 dia a 18 ºC. Os frutos foram armazenados a 2 ºC por 40 dias e deixados por uma semana à temperatura ambiente. Os tratamentos térmicos diminuíram o dano por frio durante o período de vida útil dos frutos, sendo os tratamentos de aquecimento intermitentes os mais favoráveis (principalmente em ciclos de 12 dias a 2 ºC + 1 dia a 18 ºC), pois, além de diminuírem as lesões por frio e a liberação de eletrólitos, promoveram maior biossíntese de carotenoides e coloração da epiderme, sem efeitos negativos na qualidade interna dos frutos.

PALAVRAS-CHAVE: Tangerina, cor da epiderme, qualidade pós-colheita.

INTRODUCTION

Among the citrus fruits, mandarin (*Citrus reticulate*) is the second most important crop in Colombia after orange (23.9 % and 49.7 % of the total production, respectively) (Aguilar et al. 2012). The 'Arrayana' mandarin is considered the variety most widely grown in Colombia (Ordúz-Rodríguez et al. 2012, Chaparro-Zambrano et al. 2017). However, in Colombia, citrus fruits show high postharvest losses - between 12 % and 25 % (Aguilar et al. 2012) - which are due, among other factors, to the lack of research and implementation of practices that increase their useful life. This is reflected in the weak competitiveness of the national production and the risk of producers to be displaced from the national trade.

Conservation under low temperatures is an efficient way of extending the postharvest life of fruits and vegetables, as well as reducing commercial losses during storage (Lado et al. 2015). Recommended safe minimum temperatures for mandarin postharvest storage are between 5 ºC and 8 ºC (Kader & Arpaia 2002). However, fruit species of different cultivars and *Citrus* species are sensible to chilling injury when stored at temperatures below 10 ºC (Lafuente & Zacarías 2006).

The main symptoms of chilling injury include stem-end rind breakdown (Porat et al. 2004), changes in the respiratory rate, poor color development, reduced

tissue softening, internal and external browning and higher susceptibility to diseases (Hobson 1987, Cheng & Shewfelt 1988, Lafuente & Zacarías 2006). Primary responses to chilling injury at the cellular level include changes in the fruit membranes and structures, which affect the membrane permeability. Secondary responses include symptoms such as electrolyte release, decrease in metabolic energy, oxidative damage and cellular lysis (Raison & Orr 1990, Shadmani et al. 2015).

The first method to avoid chilling injury is storage at a temperature above the threshold for the development of this alteration. Sometimes, this storage limits the product's commercial life, because the temperature is too high to slow down physiological processes. Thus, finding a temperature that lengthens the postharvest life of mandarin fruits without the appearance of chilling injury is a very difficult task. Chilling injury can be reduced by increasing the product tolerance or delaying the symptom development. In general, there are not efficient strategies to avoid the chilling injury (Wang 2000), but a viable solution is the application of thermal treatments that decrease the sensitivity of mandarin fruits to chilling injury. Several postharvest treatments that reduce the symptoms of chilling injury have been reported, including intermittent warming, vapour and hot water treatments (Sala & Lafuente 2000, Fallik 2004, Liu et al. 2015).

Intermittent warming represents an effective physical technique to decrease the chilling injury in many fruits (Liu et al. 2015). This treatment consists of storing the fruit at low temperatures, and then, periodically, relocating it to a different environment or increasing the temperature for a few hours or days, in order to make the incipient chilling injury processes reversible at a physiological level that it does not show symptoms. Afterwards, the temperature is lowered again to benefit from refrigeration (Artés & Artés-Hernández 2003). The intermittent warming with cycles of one week at 2 ºC + one day at 20 ºC during seven weeks of storage for the 'Navelate' orange, followed by one week of shelf life at 20 ºC, reduced the chilling injuries to zero, if compared to 34.5 % of injury in the control (Artés et al. 1998).

Other thermal treatments, such as the immersion of fruits in hot water, can induce the tolerance to cold through the modulation of anti-oxidant systems, which can prevent the accumulation of reactive oxygen species (Sala & Lafuente 2000, Ma et al. 2014). For 'Satsuma' mandarins stored for 8 weeks at 2 ºC, Ghasemnezhad et al. (2008) reported a lower incidence of chilling injury when mandarins were treated with water at 47.5 ºC for 2 min and 5 min and at 50 ºC for 2 min. Temperatures higher than 50 ºC increased the heat damage in the epidermis, indicating the importance of the temperature and immersion time in hot water as determining factors of the efficiency of the thermal treatment. According to Sevillano et al. (2009) and Holland et al. (2012), prolonged times and higher temperatures of thermal treatment with water may produce negative effects on the fruit quality. However, both the treatment time and temperature depend on the species, size and maturity stage, among other factors (Ma et al. 2014). Another feature of thermal treatments is a delay in fruit ripening (Shadmani et al. 2015).

The shelf life time was the focus of this research, because it is determinant for the commercialisation of mandarin fruits, and also because symptoms of chilling injury are often visible in products that have been removed from cold storage and exposed to higher temperatures (Schirra & Cohen 1999). Thus, this study aimed to evaluate the effects of thermal treatments on chilling injury and shelf life time of 'Arrayana' mandarin fruits (*Citrus reticulata* Blanco) after being stored at 2 ºC.

MATERIAL AND METHODS

Ripe 'Arrayana' mandarin fruits were selected from commercial crops in Apulo (Cundinamarca, Colombia) with the following chromatic coordinates: L* = 45.38; a* = 6.72; and b* = 34.29. The experiment was carried out at the Universidad El Bosque (Bogotá, Colombia), between 5 Nov. 2016 and 22 Oct. 2017.

Using a completely randomized design, four thermal treatments were evaluated as it follows: T1 - fruit with no treatment (control); T2 - hot water at 50 ºC for 5 min; T3 - hot water at 53 ºC for 3 min; T4 - intermittent warming in cycles of 8 days at 2 ºC + 1 day at 18 ºC; and T5 - intermittent warming in cycles of 12 days at 2 ºC + 1 day at 18 ºC. Each treatment had four replicates, and each one of the 20 experimental units was composed of approximately 2 kg of fruits. After the treatments, the fruits were stored at 2 ± 0.5 ºC for 40 days and then left for a week at room temperature (18 ± 2 ºC) to evaluate the shelf life behaviour.

During the shelf life time (zero, third and seventh day), the following variables were measured: respiration rate (mg CO_2 kg^{-1} h^{-1}), determined following the methodology used by Balaguera-López et al. (2017) through a system of infrared CO_2 sensors; and weight loss (%), calculated with 100 g of fruits by relating the weight loss to the initial fruit weight, using a 0.001 g precision scale.

At the end of shelf life time, the total carotenoids were measured according to Balaguera-López & Palacios (2018). Approximately 0.5 g of flavedo was weighted, mixed with 5 mL of acetone in a vortex for 1 min and then placed in a centrifuge for 10 min at 4,000 rpm. The supernatant was placed into a 20 mL volumetric flask, and acetone was added to the pellet again, repeating the process until the pellet was colourless. Acetone was added to the supernatant to a volume of 20 mL, absorbance was measured with a spectrophotometer at 450 nm, and a calibration curve was constructed to express the carotenoids as µg of β-carotene g^{-1} of fresh weight.

The chilling injury was evaluated using the chilling injury index (CII). Fruits were classified into different classes using a visual scale and the following values: 0 = no injury; 1 = slight injury with up to 10 % of damaged surface; 2 = medium injury with 10-50 % of the surface with spots; and 3 = severe injury with more than 50 % of the surface with larger damages. The CII was calculated using the following formula (Herrera 2007): CII = [∑(number of fruits in each class * class value)]/(total number of examined fruits).

The electrolyte leakage was measured using 10 discs with a diameter of 2.5 mm of the fruit epidermis. These discs were placed in falcon flasks with 4 mL of deionised water at room temperature. The electric conductivity was measured at 22 h using a digital conductivity meter. After the measurement, the sample was heated at 90 ºC for 15 min, and the electric conductivity was measured using this value as maximum electrolyte leakage. Electric conductivity values were expressed as percentage of the highest value (Rodríguez et al. 2005).

For the epidermis color, the L*, a* and b* parameters of the CIELab system were determined for three points at equal distances in the equatorial plane of the fruit using a Minolta CR-20 colorimeter, and, from these parameters, the difference or change in color (ΔE) was calculated according to Mendoza et al. (2006) by the following formula: $\Delta E^*_{ab} = [(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2]^{1/2}$

The titratable acidity (% of citric acid) was measured using NaOH acid-base titration in a digital burette. Soluble solids were obtained with a Hanna digital refractometer, 0-85 % Brix range (0.1 Brix precision). Soluble sugars (sucrose, fructose and glucose; g L^{-1}) and organic acids (citric, malic, oxalic and ascorbic; g L^{-1}) were quantified through high performance liquid chromatography (HPLC) using a Ultimate 3000 Dionex HPLC System, a Hypersil Gold Amino 250 µm × 4.6 µm × 5 µm, DAD 4-channel detector (210 nm, 240 nm, 272 nm and 280 nm), refraction index (35 ºC) and acetonitrile-water as the mobile phase 75:25 1.5 mL min^{-1}.

The resultant data were tested for normality (Shapiro-Wilk test) and variance homogeneity (Levene's test) and an analysis of variance was performed, followed by the Tukey test ($p < 0.05$). The Statistical Package for the Social Sciences (SPSS) version 19 was used.

RESULTS AND DISCUSSION

There was a linear increase in the weight loss with statistical differences only at the end of the shelf life time. The highest weight loss was obtained with fruit immersion for 5 min in water at 50 ºC, and the weight loss in the control fruit was lower (Table 1). Treatments may negatively affect the fruit epidermis and cause heat injury because of the high temperature, and even though there were no visible symptoms of this damage, it is possible that, in the fruit treated with water at 50 ºC for 5 min, the epidermis was slightly affected, and thus the water (and consequently the weight) loss by transpiration increased. Thermal treatments, according to Bassal & El-Hamahmy (2011), can increase or decrease the fruit weight loss, depending on the treatment and the product, mainly because of the particular response of each fruit to the heat treatment. For instance, in *Citrus*, the hot water treatment increased the weight loss in 'Fortune' mandarin (Schirra & D'hallewin 1997) and 'Navel' orange (Bassal & El-Hamahmy 2011; water at 41 ºC for 20 min) and decreased it in 'Valencia' orange (Mohamed et al. 2002), whereas, in 'Satsuma' mandarin, there was no change (Hong et al. 2007).

At the beginning of the shelf life time, the highest respiratory rate was obtained in the fruit treated with intermittent warming, and the lowest

Table 1. Effect of thermal treatments on weight loss and respiration rate of 'Arrayana' mandarin during the shelf life time, after storage for 40 days at 2 ºC.

Thermal treatments	Weight loss (%)			Regression
	Shelf life (days)			
	0	4	7	
Control	0	7.34 ± 0.42 a	10.30 ± 0.56 b	Y = 1.5612x; R^2 = 0.97
5 min at 50 ºC	0	8.97 ± 0.50 a	12.26 ± 0.65 a	Y = 1.8725x; R^2 = 0.96
3 min at 53 ºC	0	6.74 ± 1.10 a	10.38 ± 1.15 b	Y = 1.5327x; R^2 = 0.99
IW/8d	0	8.05 ± 0.32 a	10.57 ± 0.43 b	Y = 1.6335x; R^2 = 0.94
IW/12d	0	7.31 ± 1.44 a	11.26 ± 1.62 ab	Y = 1.6623x; R^2 = 0.99
Significance	-	ns	*	
Thermal treatments	Respiration rate (mg CO_2 kg^{-1} h^{-1})			Regression
	Shelf life (days)			
	0	4	7	
Control	2.83 ± 1.31 b	8.92 ± 3.41 a	15.22 ± 2.91 a	Y = 2.0664x + 0.7239; R^2 = 0.99
5 min at 50 ºC	5.93 ± 2.00 b	7.18 ± 2.96 a	7.59 ± 1.57 d	Y = 0.2411x + 6.0159; R^2 = 0.96
3 min at 53 ºC	5.47 ± 0.51 b	11.61 ± 1.50 a	12.58 ± 2.26 b	Y = 1.1842x + 5.1487; R^2 = 0.85
IW/8d	15.76 ± 2.63 a	12.19 ± 1.60 a	11.08 ± 1.51 c	Y = -0.7813x + 16.137; R^2 = 0.92
IW/12d	17.65 ± 1.22 a	8.67 ± 2.68 a	10.66 ± 2.86 c	Y = 0.6095x² - 6.041x + 23.082; R^2 = 1
Significance	*	ns	*	

ns: no statistical differences; * statistical differences at 5 %, according to the analysis of variance. Means followed by different letters in each column show statistical differences according to the Tukey test (p < 0.05). ± standard error. 5-min and 3-min treatments involved immersion in hot water. IW/8d: intermittent warming in 8-day cycles at 2 ºC + 1 day at 18 ºC; IW/12d: intermittent warming in 12-day cycles at 2 ºC + 1 day at 18 ºC.

was in the control fruit and immersion in hot water treatments. Up to four days of shelf life there were no significant differences, whereas, at the end of the period, the control fruit showed a significantly higher respiration. The opposite happened with the fruit treated with water immersion for 5 min at 50 ºC. The respiratory rate showed linear trends, except for the fruit with intermittent warming every 12 days (quadratic trend) (Table 1).

The higher respiratory rate at the beginning of the shelf life in the fruit treated with intermittent warming happens because, in these treatments, the storage refrigeration is periodically interrupted, and thus the fruit tends to increase its respiratory rate and metabolic activity and then stabilise. At the end, the thermal treatments significantly decreased the respiratory rate, if compared to the control, possibly because they suffered less chilling injury, given that this disorder results in an increase in the respiratory rate. Ghasemnezhad et al. (2008) report that both the respiration and ethylene production are correlated to the chilling injury in mandarin skin.

The hot water treatments slightly increased the respiratory rate, but decreased the chilling injury. It is possible that this increase in the respiratory rate was also related to a tolerance response to chilling injury, as well as to the sudden temperature change in the shelf life time. However, the observed increase

was significantly lower than that of the control fruit. Similar results were found by Ghasemnezhad et al. (2008) in 'Satsuma' mandarin, with hot water at 50 ºC or 55 ºC for 2 min, in which both the respiratory rate and ethylene production decreased.

Statistical differences (p < 0.01) among the treatments were found for the chilling injury index. The highest value was obtained for the control fruit, whereas the fruits subjected to intermittent warming showed the lowest indices, with values below 1 (Figure 1a). These results show that the 'Arrayana' mandarin is sensitive to chilling injury when stored at 2 ºC for an extended period. Furthermore, it is important to mention that, at the beginning of the shelf life time, the fruit did not show symptoms of chilling injury, what confirms the fact that these symptoms are visible in products that have been removed from refrigerated storage and subjected to higher temperatures (Schirra & Cohen 1999). All thermal treatments were effective in reducing the chilling injury, even though they did not prevent them altogether, what is coincident with the results for 'Satsuma' mandarin (Ghasemnezhad et al. 2008).

Chilling injury symptoms are the result of oxidative stress at the tissue level (Lukatkin 2002); thus, it is possible that the thermal treatments produced chilling tolerance in the mandarin fruit through the modulation of the antioxidant systems,
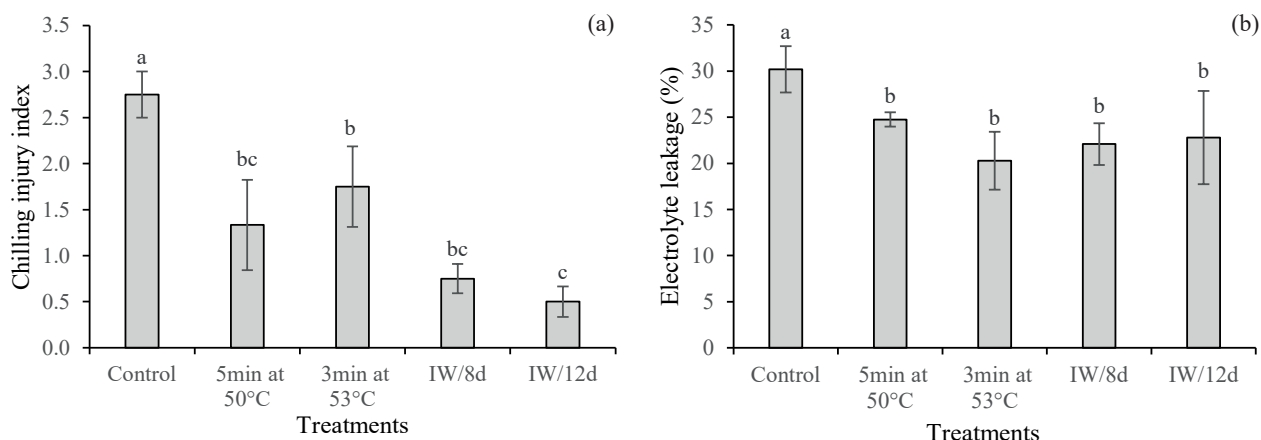
Figure 1. Effect of thermal treatments on the chilling injury index (a) and electrolyte leakage (b), in 'Arrayana' mandarin fruits at the end of the shelf life period, after storage for 40 days at 2 ºC. Means followed by different letters show statistical differences according to the Tukey test ($p < 0.05$). Vertical bars on each mean indicate the standard error (n = 4). 5-min and 3-min treatments involved immersion in hot water. IW/8d: intermittent warming in 8-day cycles at 2 ºC + 1 day at 18 ºC; IW/12d: intermittent warming in 12-day cycles at 2 ºC + 1 day at 18 ºC.

which can prevent the accumulation of reactive oxygen species, as reported by several authors (Sala & Lafuente 2000, Ma et al. 2014, Maul et al. 2011).

The intermittent warming was the most effective treatment in reducing the chilling injury, as reported for other fruits (Liu et al. 2015). Furthermore, a 12-day warming cycle is effective for mandarins, with intermediate results for the 8-day heating cycle and 50 ºC hot water pre-treatment. Crisosto et al. (2008) reported that an incorrect intermittent warming prevents the fruit from regaining its metabolic functions, producing an accelerated senescence. In accordance with these results, for 'Navelate' orange, the intermittent warming for 1 day at 20 ºC per week for seven weeks at 2 ºC reduced the chilling injury (Artés et al. 1998). Biswas et al. (2012) found that three intermittent heating cycles were enough to reduce the chilling injury in tomato fruits. Ghasemnezhad et al. (2008) found that the treatment in water at 47.5 ºC for 2 min and 5 min and at 50 ºC for 2 min decreased the incidence of chilling injury in 'Satsuma' mandarin.

The electrolyte leakage showed significant statistical differences among the treatments ($p < 0.05$). A higher electrolyte leakage was found in the control fruit (30.18 ± 2.51 %). In all the thermal treatments there was a lower leakage, and there were no significant statistical differences among them (Figure 1b).

The fruit subjected to chilling injury showed changes in the integrity of the cell membrane, which led to a higher electrolyte release (Raison & Orr 1990, Shadmani et al. 2015). Damages to the cell membranes during the cold storage are caused by lipid peroxidation. Unsaturated fatty acids are affected by oxygen-reactive species (Liu et al. 2015), and it could be that this happened in the mandarin fruit. However, the electrolyte leakage was lower with the thermal treatments, possibly because these treatments lower the production of oxygen-reactive species (Sala & Lafuente 2000, Ma et al. 2014). In this respect, an increase in the peroxidase enzyme with the hot water treatments has been found in 'Navel' and 'Valencia' orange fruits (Bassal & El-Hamahmy 2011), as well as an increase in the catalase enzyme in 'Satsuma' (Ghasemnezhad et al. 2008) and 'Fortune' (Sala & Lafuente 2000, with hot water at 53 ºC for 3 min) mandarins. Likewise, unsaturated fatty acids increased in chili pepper fruits with intermittent warming treatments and were associated with minor damages to the membrane (Liu et al. 2015).

A lower electrolyte leakage in mandarin fruit treated with hot water could be because this kind of treatment can increase the production and activation of thermal shock proteins (Rozenzvieg et al. 2004). Some of these proteins may act as chaperones, involved in the folding, assemblage and transport of proteins, many of which are present and have biological roles in the cell membrane, thus avoiding a further damage, even under low temperature conditions in this important cell organelle (Rozenzvieg et al. 2004).

There was a higher change in color in the fruits subjected to intermittent heating for both 8 and 12 days (p < 0.05), whereas the other treatments showed the lowest change (Figure 2a). As for total carotenoids, there were statistical differences between the treatments (p < 0.05). The intermittent warming every 12 days induced the highest concentrations of total carotenoids, and an opposite response was observed in the fruit subjected to hot water for 3 min at 53 ºC (Figure 2b). The other treatments presented intermediate results.

It is possible that the temperature changes in the intermittent warming treatments be necessary for the synthesis and activity of enzymes involved in the carotenoid biosynthesis and chlorophyll degradation, what could explain the higher carotenoid accumulation in such treatments, as well as the increased change in the epidermis color. In accordance with this, Le Roux (2006) indicates the importance of temperature fluctuation on the development of color in *Citrus* fruits. Mandarins grown in the tropics rarely

develop an appropriate color, and that is the reason why, in many cases, they are treated with ethylene to make the green color disappear (Rodrigo et al. 2013). Accordingly, it is important to state that intermittent warming treatments become a favourable option as a postharvest technique for 'Arrayana' mandarins, because it decreases the chilling injury and could produce a more desirable fruit coloration, which is regarded as a favourable condition for commercial purposes.

For the soluble sugars, fructose and glucose did not show statistical differences among the treatments, whereas, for sucrose, significantly higher values were obtained in the mandarin fruit subjected to thermal treatment with hot water for 3 min at 53 ºC. An opposite response was found in the control fruit and that subjected to intermittent warming every 12 days. The other treatments showed intermediate results. Furthermore, sucrose was found to be the predominant sugar (Table 2), as also happened for 'Satsuma', 'Okitsuwase' and 'Dobashi Beni'
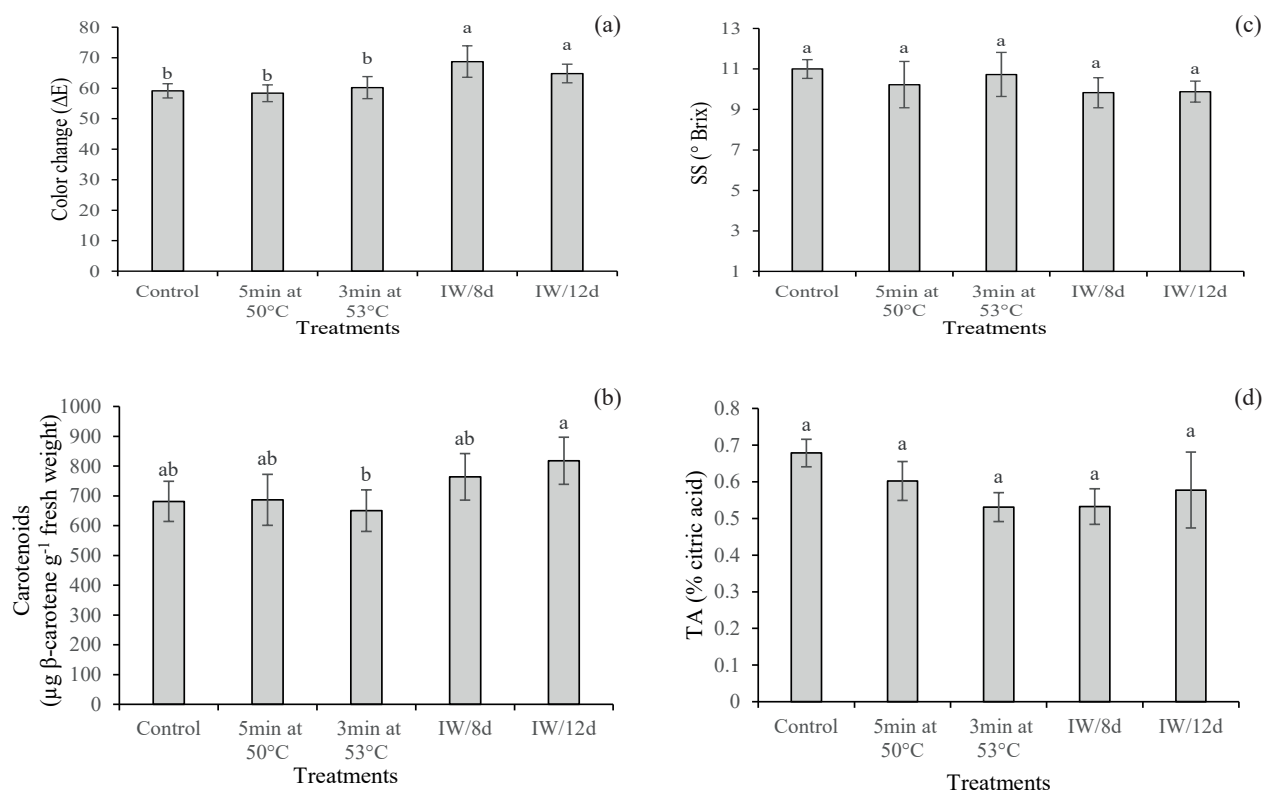

Figure 2. Effect of thermal treatments on color change (ΔE) (a), total carotenoids (b), soluble solids (c) and titratable acidity (d) on 'Arrayana' mandarin fruits at the end of the shelf life time, after storage for 40 days at 2 ºC. Means followed by different letters show statistical differences according to the Tukey test (p < 0.05). Vertical bars on each mean indicate the standard error (n = 4). 5-min and 3-min treatments involved immersion in hot water. IW/8d: intermittent warming in 8-day cycles at 2 ºC + 1 day at 18 ºC; IW/12d: intermittent warming in 12-day cycles at 2 ºC + 1 day at 18 ºC.

Table 2. Effect of thermal treatments on soluble sugars (g L⁻¹) and organic acids (g L⁻¹) for 'Arrayana' mandarin fruits at the end of the shelf life time, after storage for 40 days at 2 ºC.

Thermal treatments	Sucrose	Fructose	Glucose	Citric acid	Malic acid	Oxalic acid	Ascorbic acid
Control	26.17 ± 5.95 b	15.70 ± 0.88 a	19.40 ± 2.36 a	15.74 ± 3.83 a	5.17 ± 0.75 a	3.76 ± 0.51 a	0.22 ± 0.07 a
5 min at 50 ºC	35.35 ± 1.64 ab	20.15 ± 1.33 a	23.86 ± 2.72 a	15.36 ± 1.60 a	5.42 ± 0.88 a	3.57 ± 0.62 a	0.50 ± 0.23 a
3 min at 53 ºC	48.81 ± 4.76 a	16.14 ± 4.19 a	19.91 ± 2.28 a	15.22 ± 3.24 a	3.95 ± 0.57 a	4.05 ± 0.22 a	0.83 ± 0.22 a
IW/8d	39.16 ± 3.38 ab	18.14 ± 1.74 a	19.60 ± 3.10 a	17.36 ± 4.93 a	5.26 ± 0.30 a	3.75 ± 0.63 a	0.66 ± 0.32 a
IW/12d	30.57 ± 3.77 b	15.32 ± 1.78 a	20.12 ± 2.86 a	12.00 ± 1.97 a	3.61 ± 0.98 a	3.67 ± 0.49 a	0.65 ± 0.22 a
Significance	*	ns	ns	ns	ns	ns	ns

ns: no statistical differences; * statistical difference at 5 %, according to the analysis of variance. Means followed by different letters in each column show statistical differences according to the Tukey test ($p < 0.05$). ± standard error. 5-min and 3-min treatments involved immersion in hot water. IW/8d: intermittent warming in 8-day cycles at 2 ºC + 1 day at 18 ºC; IW/12d: intermittent warming in 12-day cycles at 2 ºC + 1 day at 18 ºC.

mandarins (Kafkas et al. 2011). As for soluble solids, there were no statistical differences (Figure 2c).

Mandarins are non-climacteric fruits that are also characterised by showing little change in internal ripening after the harvest, and, as such, changes in sugars and acids are not very noticeable. However, the increase in sucrose observed in the mandarin fruits treated with hot water and with intermittent warming every 8 days could be an indication that this treatment may be producing a higher biosynthesis of this sugar from glucose, fructose and organic acids, whose levels were lower in this thermal treatment. For 'Satsuma' mandarin fruits, the sugar levels were higher when treated at 50 ºC for 2 min (Ghasemnezhad et al. 2008). During refrigerated storage, there was a decrease in organic acids, what could be a sign of energetic metabolic changes, pH stability and defence compounds that prevent, or are involved in, the repairing of injuries caused by low temperatures (Maldonado et al. 2004). However, there were no statistical differences in the total titratable acidity and organic acids among the thermal treatments for the mandarin fruit (Figure 2d and Table 2).

It could be that there were different metabolic rates of organic acids, although the final levels were similar in all the fruits. For instance, in thermal treatments, the acids could have been directed to prevent chilling injury, whereas, in the control fruit, they could have been used mostly as respiratory substrates. It is remarkable that ascorbic acid levels were also similar in all the fruits, and thus the thermal treatments did not negatively affect this compound of high interest for human health. Ghasemnezhad et al. (2008) did not observe differences in acidity in mandarin fruits treated with hot water. Bassal & El-Hamahmy (2011) reported an increase in ascorbic

acid in 'Valencia' orange treated with hot water at 50 ºC, whereas Schirra et al. (2004) observed that the thermal treatment with hot water did not affect the soluble solids, titratable acidity or ascorbic acid.

CONCLUSIONS

1. Thermal treatments decrease the chilling injury in 'Arrayana' mandarin fruits stored at 2 ºC for 40 days, followed by a shelf life time of one week. These treatments also do not affect the acidity, organic acids or soluble solids;
2. Postharvest intermittent warming, mainly in cycles of 12 days at 2 ºC + 1 day at 18 ºC, is more favourable for mandarin fruits, because it reduces the chilling injury and electrolyte leakage, produces a high carotenoid biosynthesis, a better coloration and do not affect the internal quality of the fruit.

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