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Effect of preharvest foliar sprays of calcium nitrate on melon fruit quality

Efecto de la aspersión foliar con nitrato de calcio en precosecha sobre la calidad de melón

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Abstract. The aim of this work was to study the effect of preharvest foliar spraying with calcium nitrate solutions on the fruit melon quality. Treatments included a combination of five solutions with $Ca(NO_3)_2$ (0.0; 1.3; 2.6; 5.2 and 10.5 g/L) and two postharvest storage environments [(24.0 ± 1.5 ˚C and 6.4 ± 0,8 mbar of vapor pressure deficit (VPD), and 10.0 ± 0.5 ˚C and 3.0 ± 0.5 mbar (VPD)]. Fruits stored at 24 ˚C for 11 days had the lowest water loss with treatments 1.3 and 2.6 g/L Ca $(NO_3)_2$. It was observed that at both 24 ˚C and 10 ˚C, fruits that had a greater firmness at the 2.6 and 5.2 g/L of $Ca(NO_3)_2$ treatments, respectively. No differences (P>0.05) were observed in total soluble solids (TSS) between treatments and storage environments. However, spotting of the epidermis of the fruits increased as the $Ca(NO_3)_2$ concentration also increased.

Keywords: $Ca(NO_3)_2$ solutions; Fruit storage; Firmness; Total soluble solids.

Abbreviations: FW, fresh weight; DM, dry matter; TSS, total soluble solids; WL, water loss; Ca(%), calcium concentration in tissues.

Resumen. El objetivo de este trabajo fue estudiar el efecto de la pulverización foliar en un cultivo de melón en precosecha con soluciones de nitrato de calcio sobre la calidad de los frutos. Se utilizó una combinación de cinco soluciones con $Ca(NO_3)_2$ (0,0; 1,3; 2,6; 5,2 y 10,5 g/L) y dos ambientes de almacenamiento durante la poscosecha [(24,0 ± 1,5 ˚C y 6,4 ± 0,8 mbar de déficit de presión de vapor (VPD), y 10,0 ± 0,5 ˚C y 3,0 ± 0,5 mbar (VPD)]. Los frutos almacenados a 24 ˚C durante 11 días tuvieron la menor pérdida de agua con los tratamientos 1,3 y 2,6 g/L $Ca(NO_3)_2$. Se observó que tanto en las cámaras a 24 ˚C y 10 ˚C, los frutos presentaron una mayor firmeza con 2,6 y 5,2 g/L de tratamientos con $Ca(NO_3)_2$. No se observaron diferencias (P>0,05) en los sólidos solubles totales (TSS) entre tratamientos y almacenamiento, observándose la ocurrencia de daños por manchado de la epidermis de los frutos, incrementándose la gravedad de estos daños con el aumento de la concentración de $Ca(NO_3)_2$.

Palabras clave: Soluciones de $Ca(NO_3)_2$; Almacenamiento de frutos; Firmeza; Sólidos solubles totales.

Abreviaturas: FW, peso fresco; DM, materia seca; TSS, sólidos solubles totales; WL, pérdida de agua; Ca(%), concentración de calcio en los tejidos.

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INTRODUCTION

A frequent problem in melon cultivation is the unfavorable conditions for adequate calcium (Ca) absorption (Alarcón et al., 1999). Plants absorb calcium through the root, which is then distributed via the xylem sap flux by physiological and molecular mechanisms involved in the ion absorption and transport in the plant (Singh et al., 2015). Because of its dependence on the transpiration flux, the calcium concentration is much lower in the fruits than in the leaves (Saure, 2014). Calcium is important in melon because it participates in the regulation of fruit softening (Madani & Forney, 2015), consequently affecting the product quality and its potential shelf life.

In horticulture, foliar nutrition is a technique used to improve fruit quality and its productivity (Peyvast et al., 2009; Santos, 2013). Experiences have been done using fertilization by means of foliar sprays because it is simple, and used occasionally to increase the calcium level of the fruits (Gastol & Domagala-Swiatkiewicz, 2006). Foliar spraying with calcium solutions would allow to delay fruit ripening, to decrease fruit decay during postharvest, and to extend the shelf life (Lara, 2013).

However, some studies have shown that the efficiency of foliar sprays on the fruits is limited, if calcium is in the form of nitrate and chloride (Madani & Forney, 2015). In a previous work performed in different environments and with other calcium solutions, it was observed that the effectiveness of the treatments to keep both fruit firmness and quality of melon was different (Bouzo & Cortez, 2012). The calcium nitrate solution gave better results than other treatments. Even more, the use of increasing concentrations of calcium nitrate might have greater efficiency in decreasing postharvest melon deterioration. An issue that should be especially considered, in the light of previous works, is the risk of the occurrence of spotting on the fruits with the increase in calcium concentration (Lester & Grusak, 2001; Pereira et al., 2002; Lara, 2013). The aim of this work was to evaluate the effects of preharvest foliar sprays with calcium nitrate solutions on melon quality.

MATERIALS AND METHODS

The experiment was performed in the "Campo Experimental de Cultivos Intensivos y Forestales (CECIF)" (Intensive Farming and Agroforestry Experimental Field), Universidad Nacional del Litoral, UNL, (31°27´S; 60°56´W). The climate classification according to Köppen-Geiger is Cfa. Melon seeds (*Cucumis melo* L.) cv. "Fila" (yellow) were sown in multicell trays in a greenhouse. Transplanting was done at the beginning of the spring of 2015, when the first leaf was totally expanded. The experiment was carried out using a plantation frame (1.5 m x 0.6 m; 1.10 plants/m^2) on a typical Argiudoll soil. The chemical analysis of soil samples taken from 0-25 cm soil depth determined: pH 6.59 (soil: water ratio 1.0:2.5); electrical conductivity 0.08 dS/m; total N (Kjeldahl) 0.147%, P (Bray & Kurtz nº 1) 10 ppm; Ca 12.15 meq/100 g; Na 0.31 meq/100 g, and K 0.77 meq/100 g. All the exchangeable cations were determined by extraction with ammonium acetate, and subsequent ammonium determination by Kjeldahl using the standard method.

The experiment consisted of a combination of five treatments with different calcium nitrate concentrations [$Ca(NO_3)_2$] and two postharvest storage environment. The calcium nitrate concentrations used were: 0.0 (control); 1.3; 2.6; 5.2 and 10.5 g/L $Ca(NO_3)_2$, and they were supplied by foliar spraying with five applications, starting when the first fruits were established, followed by a weekly frequency. The sprinkling was allowed during the morning with a volume equivalent to 0.025 L/m^2 per application. The plants in the control treatment (0 g/L) were sprinkled with distilled water. The fertilization was performed by fertigation with N, P and K and whose doses were calculated according to the method of Bouzo et al. (2004). The harvest began when randomly selected fruits reached 10 °Brix. Then, postharvest storage environments were carried out using two independent chambers with temperature and humidity of: (1) 24.0 (± 1.5) °C and 6.4 (± 0.8) mbar of vapor pressure deficit (VPD), and (2) 10.0 (± 0.5) °C and 3.0 (± 0.5) mbar.

The fruit allocation to each environment was performed in a completely randomized manner, completing a total of 150 fruits in each storage environment, consisting of 30 fruits by each calcium nitrate treatment. Measurements were made at 11 and 22 days after harvest. Measurements on each fruit included: water loss (mg H_2O/kg FW/h/mbar), firmness (kg) and total soluble solids, TSS (°Brix). Water loss was estimated from the difference between the individual weight of each fruit at the beginning and end of the experiment, using a precision balance (± 0.01 g) (Ohaus Corporation, Parsippany, NJ, USA). Fruit firmness was measured with an Effegi penetrometer using a plunger tip with a 7.9 mm diameter (Novanna LDT, St Edmunds, UK). The TSS were measured using a manual refractometer with automatic temperature compensation (Atago Co. LTD, Tokyo, Japan). The analytic determination of calcium percentage in dry matter (DM %) of cortex and pulp was performed according to Horwitz & Latimer (2010).

Experimental data were analyzed with a two-way variance analysis (ANOVA). The Tukey´s test was used to separate mean values with a significance level of P≤0.05. The analysis was performed using the statistical software Statgraphics® on a Windows platform.

RESULTS AND DISCUSSION

Statistical differences of the water loss of the fruit depending on the treatments were observed (Fig. 1). For the fruit stored at 25 °C during 11 days, the lowest water loss was obtained for the

treatments with 1.3 and 2.6 g/L $Ca(NO_3)_2$, with no significant difference from the control treatment. When the fruit were stored at 10 ˚C for 22 days, the treatments with 1.3, 2.6 and 5.2 g/L gave the smallest water loss (Fig. 1).

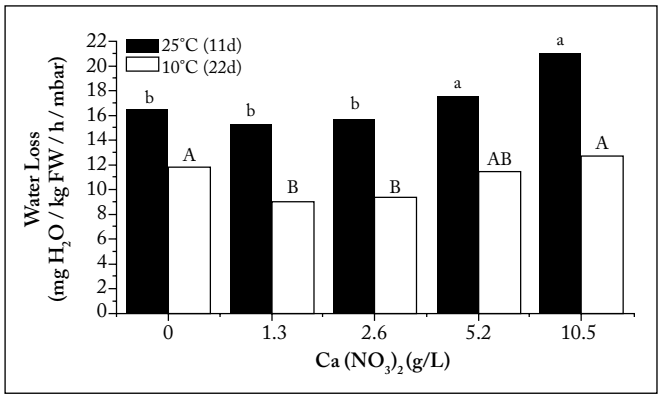

Fig. 1. Effect of the foliar application of $Ca(NO_3)_2$ on water loss of melon fruit. Different lowercase letters indicate significant differences among $Ca(NO_3)_2$ treatments stored at 25 ˚C during 11 days (11 d) (P≤0.05, Tukey's test). Different uppercase letters indicate significant differences among $Ca(NO_3)_2$ treatments stored at 10 ˚C during 22 days (22 d) (P≤0.05, Tukey's test).

Fig. 1. Efecto de la aplicación foliar de $Ca(NO_3)_2$ sobre la pérdida de agua en frutos de melón. Letras minúsculas diferentes indican diferencias significativas entre los tratamientos con $Ca(NO_3)_2$ almacenados a 25 °C durante 11 días (11 d) (P≤0,05; prueba de Tukey) Letras mayúsculas diferentes indican diferencias significativas entre los tratamientos con $Ca(NO_3)_2$ almacenados a 10 °C durante 22 días (22 d) (P≤0,05; prueba de Tukey).

From the measured firmness at the end of the storage period for each storage environment, fruits treated with 2.6 and 5.2 g/L of $Ca(NO_3)_2$ kept the higher firmness at 25 ˚C as well as at 10 ˚C (Fig. 2). These results indicate a favorable effect on the postharvest firmness for these calcium concentrations. A favorable effect on fruit firmness was also reported in *Psidium guajava* by a direct application of $Ca(NO_3)_2$ on the tree at a concentration of 27 g per plant (Castellano et al., 2004). In fresh processed melon, a lower firmness loss was obtained by using calcium chloride ($CaCl_2$) (García Méndez, 2008). A decrease of firmness with this calcium solution at a concentration of 0.7 g/L was also obtained in strawberry (García Méndez & Praderas Cárdenas, 2010). In pepper, application of calcium to leaves increased fruit firmness to harvest. This was assigned to an increase of the water insoluble pectin content, and of the cell wall of the pericarp (Toivonen & Bowen, 1999). Other possible causes might be the stabilization of the cell wall and membranes as a result of the interaction with the peptic acid (Díaz et al., 2007), as well as the inhibition of the poligaracturonase enzyme (Uhm et al., 2002).

Treatments did not affect (P≤0.05) the concentration of total soluble solids (Fig. 3). Similar findings were reported on melon, where a foliar application of chelated calcium did not show an increase of the TSS concentration (Lester & Grusak, 2001;

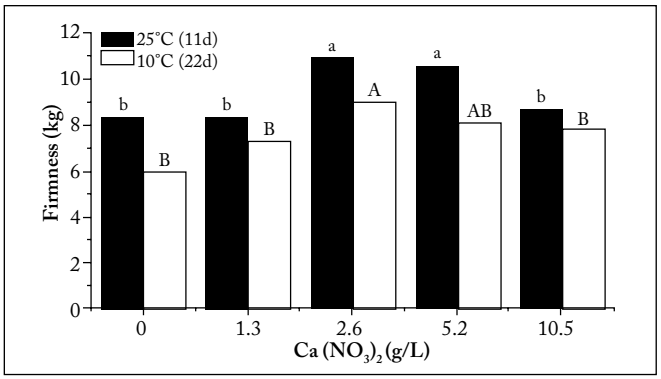

Fig. 2. Effect of the foliar application of $Ca(NO_3)_2$ on the firmness of melon fruit. Different lowercase letters indicate significant differences among $Ca(NO_3)_2$ treatments stored at 25 ˚C during 11 days (11 d) (P≤0.05, Tukey's test). Different uppercase letters denote significant differences among $Ca(NO_3)_2$ treatments stored at 10 ˚C during 22 days (22 d) (P≤0.05, Tukey's test).

Fig. 2. Efecto de la aplicación foliar de $Ca(NO_3)_2$ sobre la firmeza de frutos de melón. Letras minúsculas diferentes indican diferencias significativas entre los tratamientos con $Ca(NO_3)_2$ almacenados a 25 °C durante 11 días (11 d) (P≤0,05; prueba de Tukey) Letras mayúsculas diferentes indican diferencias significativas entre los tratamientos con $Ca(NO_3)_2$ almacenados a 10 °C durante 22 días (22 d) (P≤0,05; prueba de Tukey).

2004). In tomato, preharvest foliar applications of $Ca(NO_3)_2$ at concentrations of 4, 6 and 8 mmol/L did not increase TSS concentrations (Peyvast et al., 2009). However, the concentrations used on these investigations were lower than those used in this work (considering that 8 mmol/L is a concentration similar to the lowest concentration used in this work: 1.3 g/L).

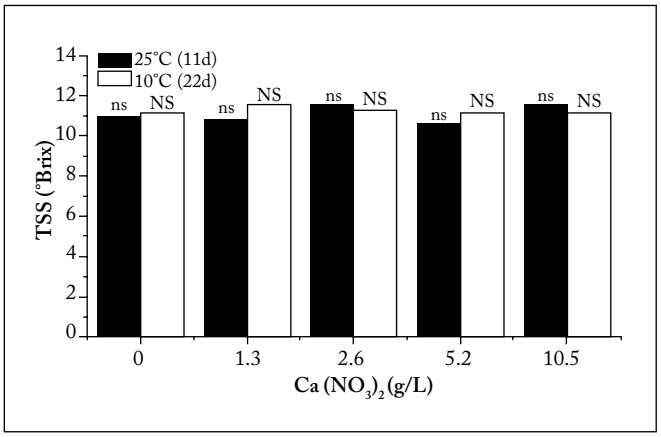

Fig. 3. Effect of the foliar application of $Ca(NO_3)_2$ on the concentration of total soluble solids in melon fruit. ns and NS indicate non-significant differences among $Ca(NO_3)_2$ treatments stored at 25 ˚C during 11 days (11 d), and at 10 ˚C during 22 days (22 d) (P≤0.05, Tukey's test), respectively.

Fig. 3. Efecto de la aplicación foliar de $Ca(NO_3)_2$ sobre la concentración de sólidos solubles totales en frutos de melón. ns y NS indican diferencias no significativas entre los tratamientos con $Ca(NO_3)_2$ almacenados a 25 °C durante 11 días (11 d), y a 10 °C durante 22 días (22 d) (P≤0,05; prueba de Tukey), respectivamente.

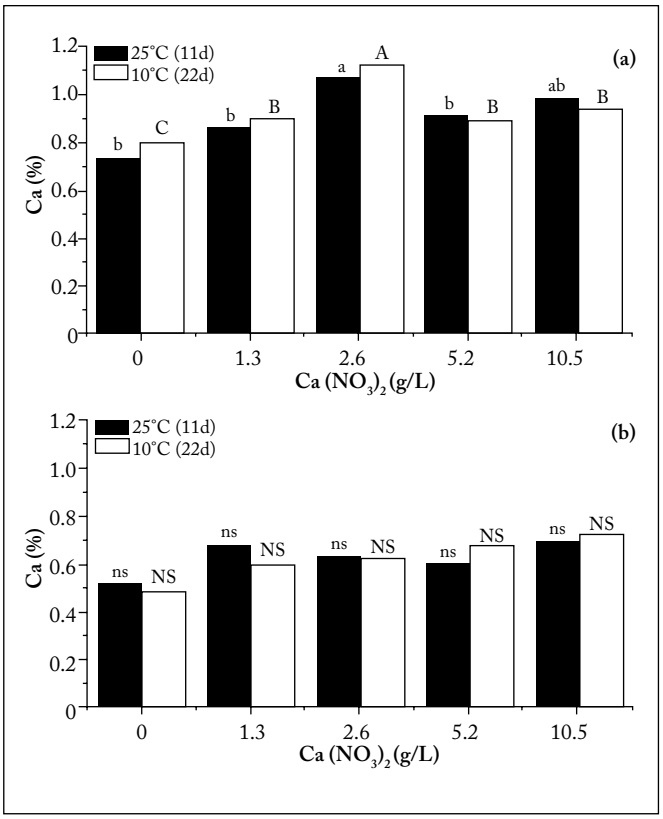

Fig. 4. Effect of the foliar application of $Ca(NO_3)_2$ on the calcium concentration in the epidermis (a), and the mesocarp (b) of melon fruits exposed to different storage environments. Different lowercase letters above the histograms indicate significant differences among the $Ca(NO_3)_2$ treatments when fruits were stored at 25 °C during 11 days (11 d) (P≤0.05, Tukey's test). Different uppercase letters indicate significant differences among $Ca(NO_3)_2$ treatments when fruits were stored at 10 °C during 22 days (22 d) (P≤0.05, Tukey's test). ns and NS indicate no significant differences (P>0.05).

Fig. 4. Efecto de la aplicación foliar de $Ca(NO_3)_2$ sobre la concentración de calcio en epidermis (a) y el mesocarpo (b) de frutos de melón expuestos a diferentes ambientes de almacenaje. Letras minúsculas diferentes sobre los histogramas indican diferencias significativas entre los tratamientos de $Ca(NO_3)_2$ cuando los frutos fueron almacenados a 25 °C durante 11 días (11 d). Letras mayúsculas diferentes indican diferencias significativas entre los tratamientos de $Ca(NO_3)_2$ cuando los frutos fueron almacenados a 10 °C durante 22 días (22 d) (P≤0,05; prueba de Tukey). ns y NS indican diferencias no significativas (P>0,05).

The Ca concentration in the fruit epidermis was highest (P≤0.05) when 2.6 g/L $Ca(NO_3)_2$ were applied in the two storage environments (Fig. 4a). However, the various treatments did not affect (P>0.05) the calcium concentrations of the mesocarp at any storage environment (Fig. 4b).

The measured increase of the calcium concentration in the fruit epidermis for the 2.6 g/L $Ca(NO_3)_2$ treatment (Fig. 4a) relates well with the higher firmness measured at this treatment (Fig. 2). There is some evidence that calcium can inhibit the activity of the water channel or aquaporins, whose complex regulation has not been yet clearly understood (Lee & Zwiazek, 2015). However, the results obtained here support the assumption that the measured increase of calcium percentage in the fruit epidermis for the 2.6 g/L $Ca(NO_3)_2$ treatment was related with a lower water loss in the fruit, especially on fruits stored at 10 °C (Fig. 1). On the other hand, for concentrations higher than 2.6 g/L $Ca(NO_3)_2$, no increases of Ca were observed in the fruit epidermis (Fig. 4b). This indicates that the calcium supplied by foliar spray was not fully absorbed by the fruit tissues. It is possible that the $Ca(NO_3)_2$ salts concentrated by drop formation, and the subsequent evaporation of these drops, led to a decrease of the surface osmotic potential causing a water release from the epidermal cells. This hypothesis is reinforced when damage is considered, as shown by the brown stains, caused by the increase in the $Ca(NO_3)_2$ concentrations (Fig. 5). A similar damage was observed by Sharples & Johnson (1976) in the epidermis of apples treated with $CaCl_2$, which led to a higher saline stress at increasing concentrations. In this work, this defect was observed on fruits stored for 22 days at 10 °C (Fig. 5). No spots were detected in the epidermis of the control treatment (Fig. 5a), while they increased as $Ca(NO_3)_2$ concentrations also increased (Table 1; Fig. 5 b-e), seriously damaging the appearance and external quality of the fruit. This is, the concentration of calcium salts was the major cause of the appearance of spots on the fruits.

Even in the 1.3 g/L treatment, dispersed stains were observed, localized in the fruit distal extreme (Fig. 5b). However, this defect was not observed on fruits stored for 11 days at 25 °C. Lester & Grusak (2001) did not observe stains at

Table 1. Effect of the $Ca(NO_3)_2$ treatments on the frequency of stains on fruits stored during 22 days at 10 °C, measured according an ad-hoc scale (*).

Tabla 1. Efecto de los tratamientos con $Ca(NO_3)_2$ sobre la frecuencia de manchas en frutos almacenados durante 22 días a 10 °C, medida de acuerdo a una escala ad-hoc (*).

$Ca(NO_3)_2$ (g/L)	Frequency of stains (*)
0.0	1.2 d
1.3	1.5 c
2.6	3.4 b
5.2	4.0 ab
10.5	4.7 a

*Estimated values from the random observation of 10 fruits per treatment, considering an ad-hoc scale of discrete values: 1. No stains; 2. Scarce stains <5 mm; 3. Abundant stains >5 mm; 4. Many stains >10 mm; 5. Stains in all fruits >10 mm. Different letters indicate significant differences among treatments (P≤0.05, Tukey's test).
*Valores estimados a partir de las mediciones al azar de 10 frutos por tratamiento, considerando una escala ad-hoc de valores discretos: 1. Sin manchas; 2. Manchas escasas <5 mm; 3. Manchas abundantes >5 mm; 4. Muchas manchas >10 mm; 5. Manchas en todos los frutos >10 mm. Letras diferentes indican diferencias significativas entre tratamientos (P≤0,05; prueba de Tukey).

Fig. 5. Appearance of the fruit under the spray treatments with $Ca(NO_3)_2$ and stored for 22 days in a cold chamber at 10 °C. (a) control; (b) 1.3; (c) 2.6; (d) 5.2; (e) 10.5 g/L $Ca(NO_3)_2$; (f) zoom image (70x) of a stain (bar = 1 mm).
Fig. 5. Aspecto que presentaron los frutos luego del rociado de $Ca(NO_3)_2$ y almacenamiento durante 22 días en una cámara frigorífica a 10 °C. (a) control; (b) 1,3; (c) 2,6; (d) 5,2; (e) 10,5 g/L $Ca(NO_3)_2$; (f) imagen ampliada (70x) de una mancha (barra = 1 mm).

day 14, using chelated calcium and $CaCl_2$ by immersion on postharvest fruits stored at 10 °C but they did after 22 days of storage. Even though the stains were brown and with the same characteristics as those observed in this work, the size was smaller (mean size = 1 mm). The occurrence of stains also increased with increases in the concentrations of the calcium solutions (Lester & Grusak, 2001).The occurrence of brown stains on the fruits was also observed for peach with applications of $CaCl_2$ at concentrations of 2 and 3% (Ali et al., 2014).

These results, when compared with the control treatment, would suggest that the Ca application is a cause, at least partially, of the stains, with an expanding stain proliferation at increasing concentrations of the sprinkled solution. The images of the stains analyzed with the binocular magnifying glass (70x) showed an alteration of the epidermis and hypodermis of the fruit (Fig. 5f). Trentham et al. (2008) studied the effect of the application of calcium in postharvest apples, using $CaCl_2$, and determined similar defects to those mentioned here. These authors analyzed the stains and concluded that they were caused by alterations of the cuticle, the epidermis and the hypodermis of the fruit. In other works where $CaCl_2$ was used, when compared with the control treatment, it was observed a defect on the lenticels at the epidermis of the fruits caused by the calcium application (Peryea & Neilsen, 2006). There are reported cases of phytoxicity after calcium application in kiwi (Cooper et al., 2007) and apple (Mayr & Schröder, 2002).

CONCLUSIONS

For the foliar sprays with solutions at concentrations of 2.6 g/L and 5.2 g/L of $Ca(NO_3)_2$, the lowest water loss and the highest hardness were obtained for fruits stored during 11 days at 25 °C, and 22 days at 10 °C. However, no changes in the concentration of total soluble solids were observed for any treatment. The highest calcium concentration in the fruit epidermis was obtained by the use of the solution at a concentration of 2.6 g/L of $Ca(NO_3)_2$. The treatments showed no effect in the calcium concentration in the mesocarp. Increasing the $Ca(NO_3)_2$ concentrations produced an increase of the epidermal damage on the fruits stored at 10 °C during 22 days. The damage appeared as brown stains that diminished the external quality of the product.

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