

Reducing Postharvest Softening of Papaya (*Carica papaya* cv. Maradol) by using an aqueous 1-Methylcyclopropene Application

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Abstract— The shelf life of ‘Maradol’ papaya fruit (*Carica papaya*) harvested at one-quarter stage maturity (yellow-orange color in 25% of fruit surface) and treated with aqueous 1-methylcyclopropene (1-MCP) was evaluated. The effects of immersion time (30 or 60 seconds) and dose response (50 or 300 ng·kg⁻¹) on the quality of papaya fruit stored for 12 days at 20 ± 2 °C and 85 ± 5% RH were studied. After 3 days, the control and fruit treated with 50 ng·kg⁻¹ 1-MCP for 30 seconds lost 82% and 72% of the initial firmness, respectively. A 1-MCP concentration of 300 ng·kg⁻¹ for 30 or 60 seconds inhibited softening and slowed skin color change for 12 days. The pH, titratable acidity, and soluble solids concentration ranged from 5.3-5.7, 0.06-0.12 % and 10-12 %, respectively. The shelf life of papaya was extended up to 3 days in fruit treated with 50 ng·kg⁻¹ 1-MCP for either 30 or 60 seconds of immersion.

Keywords— 1-MCP dipping treatment, color change, *Carica papaya*, ripening.

I. INTRODUCTION

Papaya fruit has a high rate of softening “off” the tree, and therefore, a short shelf life because it is highly susceptible to bruises, mechanical injury and postharvest diseases during postharvest handling. These deterioration problems contribute to substantial losses during handling, storage, transportation, marketing, and at households (Manenoi et al., 2007). ‘Maradol’ papaya produced in Mexico is usually recognized as a Mexican papaya in USA, where most of the production is exported in spite of postharvest limitations as high sensitivity to mechanical damage and diseases. Postharvest handling, storage, transportation and marketing conditions affect quality and, hence, consumer satisfaction. No estimates of wholesale and retail losses of Mexican papaya are reported, however, previous assessment of postharvest losses in Hawaiian papaya due to diseases, physiological disorders, mechanical damage and overripe fruit, have been estimated up to 75% (Paull et al., 1997). Mexican ‘Maradol’ papayas also have been observed to have high losses, but there is not an accurate accountability of wholesale and retail losses.

Papayas are climacteric fruits, thus, ethylene triggers ripening processes that include softening, flavor development, texture and color changes. Conversely, inhibition of ethylene production or action delays ripening changes (Sañudo-Barajas et al., 2009).

The ethylene inhibitor 1-MCP is a widely explored alternative as a strategy to delay papaya ethylene production, climacteric respiration, skin color development and softening without affecting total soluble solids and fruit weight loss (Manenoi et al., 2007; Shiga et al., 2009; Sañudo-Barajas et al., 2009). In Mexico, ‘Maradol’ papayas are harvested at the breaker maturity stage (yellow-reddish color around one-quarter of the fruit surface), while the fruit are still firm, to minimize mechanical damage during postharvest handling. Previous studies using 50 to 300 nL L⁻¹ 1-MCP applied as a gas formulation (SmartFresh™) promoted incomplete softening and undesirable rubbery texture. Furthermore, the lack of ripening was not reversed by later ethylene application (Manenoi et al., 2007; Manenoi and Paull, 2007; Sañudo-Barajas et al., 2009). As SmartFresh™ application required an extra step (12 to 24 hours) in the papaya handling, the short aqueous application (30 to 60 s) of 1-MCP application as a spray or immersion could be utilized for current papaya operations when papayas are immersed in water during standard packaging operations (Manganaris et al., 2008).

This short dipping application approach has been tested using aminoethoxyvinylglycine (AVG) (Garner and Crisosto, 2001) and 1-MCP (Blankenship and Dole, 2003) in other commodities. 1-MCP aqueous dipping treatments yielded the same results as the gas formulation in delaying fruit ripening and extending the shelf life of apples (Argenta et al., 2007), tomatoes and avocados (Tay-Choi et al., 2008), and plums (Manganaris et al., 2008). These studies also pointed out that the efficacy of aqueous 1-MCP depends on commodity, maturity stage, active ingredient concentration, and exposure time. To our knowledge, the use of an aqueous 1-MCP formulation to control ripening in papaya has not been studied. The objective of this study was to develop a practical and reliable 1-MCP dipping treatment to reduce the softening rate and extend the shelf life of 'Maradol' papaya.

II. MATERIALS AND METHODS

2.1 Plant material

A lot of 250 'Maradol' papaya fruit was harvested at one quarter maturity stage (20-25 % of yellowing on skin) from a commercial farm located in Nayarit, México. Fruit samples were classified as size 8 (1200 ± 100 g) according to the scale reported by Báez et al. (1999). Sound fruit without visible mechanical or pathogen wounds were washed and disinfected with $1 \text{ g}\cdot\text{L}^{-1}$ 2-(4-Thiazolyl) benzimidazole (Tecto[®] 60, Syngenta). The fruits were dried at room temperature and divided into sub-samples of fifty fruits for treatments.

2.2 Aqueous 1-MCP treatments

An aqueous solution of 50 or 300 $\text{ng}\cdot\text{kg}^{-1}$ of 1-MCP (AFxRD-3.8% 1-methylcyclopropene: provided by Agrofresh Inc. Rohm and Haas, Spring House, PA, USA) was prepared for dipping treatments. Fruit were dipped in 50 or 300 $\text{ng}\cdot\text{kg}^{-1}$ of 1-MCP solution for 30 or 60 s following Agrofresh's representative's instructions. Fruit dipped in water for 60 s were used as a control. After dipping, fruit were dried for 30 min at room temperature then stored at marketing conditions for 12 d at 20 ± 2 °C and $85 \pm 2\%$ RH for further postharvest evaluations.

2.3 Postharvest quality

Fruit quality was determined at harvest (before treatments) and after 3, 6, 9 and 12 days after treatment. On each evaluation time, firmness, flesh color, pH, titratable acidity (TA), and soluble solids concentration (SSC) were measured on five fruit, while accumulation of weight loss and change on skin color was evaluated on ten fruit through the storage time. Fifteen fruit were left in case of repetition or decays. Firmness and flesh color were measured on four previously peeled regions of each fruit (two at the equator, one at the apical and one at the basal area of the papaya fruit), and reported as the fruit average. Fruit firmness was measured using a Chatillon penetrometer (DFGS-100, Ametek, Largo, FL, USA) with an 8-mm diameter flat-tipped cylindrical probe and a TCD 200 motorized base. Maximum force required to penetrate 5 mm at a speed of $5.3 \text{ mm}\cdot\text{s}^{-1}$ was expressed in Newtons (N) (Bourne, 1980). Afterward, the fruit were cut in half lengthwise and the flesh color was determined (on the exposed cut area) using a Konica CM-2600d spectrophotometer (Konica-Minolta Inc, Ramsey, NJ, USA). Lightness (L), hue angle (°Hue), and chroma (color purity or saturation) values were calculated using the Minolta OnColor QC v5 software. Fruit pH, TA and SSC were evaluated in triplicate according to AOAC (1998), with slight modifications. A sample of 10 g of flesh tissue per fruit was blended with 50 ml of neutralized distilled water and filtered through cheesecloth. An aliquot of 50 ml of the filtrate was used to evaluate pH and the TA was determined with a Mettler automatic titrator (DL50, Mettler-Toledo Inc. Columbus, OH, USA) using NaOH 0.1N and expressed as percentage of malic acid. Another aliquot was used to measure the SSC with a Mettler refractometer (RE40D, Mettler-Toledo Inc. Columbus, OH, USA), and the results were expressed as SSC (%).

Ten papaya fruit were weighed individually initially and every three days throughout the storage period, and weight loss was expressed as the percentage of the cumulative difference between the initial weight and that recorded each day of evaluation. On the same fruit, the skin color was measured on four previously selected surface regions (as described above) using a Konica spectrophotometer (CM-2600d, Konica-Minolta Inc. Ramsey, NJ, USA) and the variables L, hue angle and chroma were reported.

2.4 Statistical analysis.

The experiment was arranged into a randomized design and data were subjected to analysis of variance (ANOVA) and Tukey's means separation test at a significance level of $P < 0.05$. Statistical analysis was performed using the statistical software Minitab 14.0 (Minitab Inc. State College, PA, USA).

III. RESULTS AND DISCUSSION

3.1 Firmness

At the beginning of the experiment, the 'Maradol' papaya fruit had a firmness of 141 ± 10 N, corresponding to fruit at the preclimacteric ripening stage $\frac{1}{4}$ (20-25 % of yellowing on skin) (Sanudo-Barajas et al., 2009). After 3 d storage, control fruit and those treated with $50 \text{ ng} \cdot \text{kg}^{-1}$ 1-MCP for 30 s of immersion time lost 82 and 72% firmness, respectively, with values of approximately 30-40 N with excellent commercial acceptability, according Santamaría et al. (2009), they found firmness values of 5-8 N to Maradol fully ripe fruits with commercialization quality. Meanwhile, fruit treated with the same 1-MCP dose, but with the 60 s immersion time showed a firmness loss of 47%. However, the use of $300 \text{ ng} \cdot \text{kg}^{-1}$ 1-MCP caused a strong response of softening inhibition during the 12 days of the storage period, resulting in firmer fruit flesh compared to the control and other treatments ($P \leq 0.05$) as shown in Fig. 1. The observed maintenance of papaya fruit flesh firmness has also been reported in 'Solo' papaya, and it was associated with 1-MCP application at early ripening stages of the fruit (Manenoi et al., 2007). At the end of the experiment, the control and $50 \text{ ng} \cdot \text{kg}^{-1}$ 1-MCP-treated fruit had lost about 95% firmness of the initial firmness (Fig. 1). Similar results have been reported in 'Solo' (Hofman et al., 2001), 'Maradol' (Sañudo-Barajas et al., 2009) and 'Sunset' (Thumdee et al., 2010) papaya fruits treated with 1-MCP gas, in all of them the fruit softening was due to a response induced either by ethylene or a dose of 1-MCP gas above $100 \text{ nL} \cdot \text{L}^{-1}$. This effect can cause an irreversible inhibition of softening by suppressing the xylanase enzyme activity.

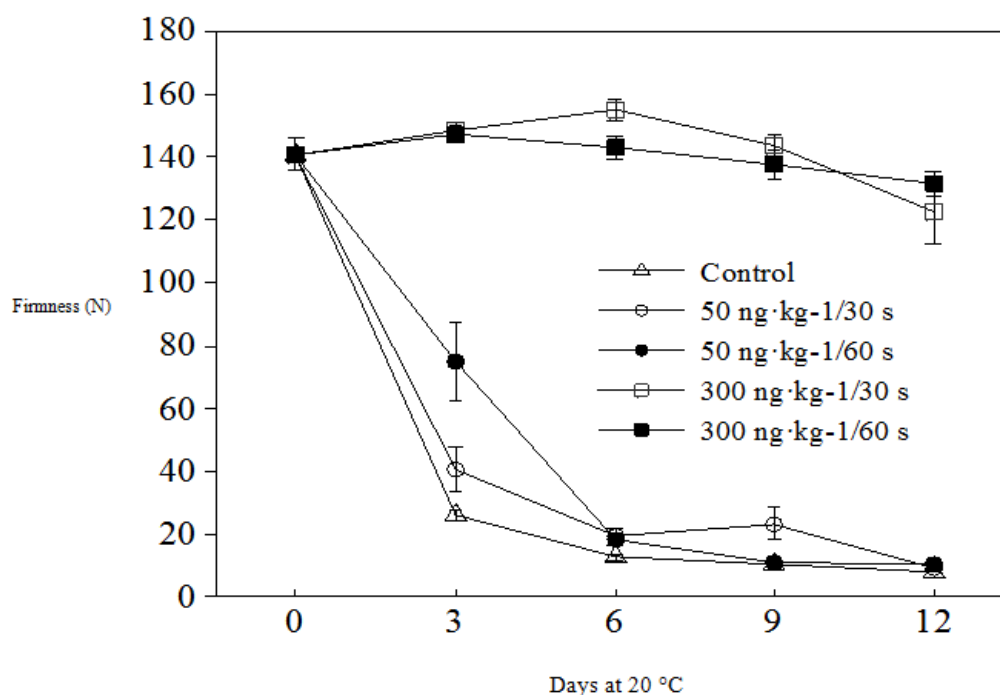


FIG. 1. Firmness of 'Maradol' papayas treated with 50 or 300 ng·kg⁻¹ of an aqueous 1-MCP formulation, applied under immersion conditions for 30 or 60 s. Bars represent standard error (n = 5). 9.8 N = 1 kg-force

One of the main advantages of using 1-MCP is the delay of fruit softening in various fruit through the modification of a diverse set of biochemical reactions (Blankenship and Dole, 2003). In 'Maradol' (Sañudo-Barajas et al., 2009) and 'Sunset' (Thumdee et al., 2010) papayas, 1-MCP inhibited the action of polygalacturonase, xylanase and galactanase, all enzymes related to fruit firmness. Sañudo-Barajas et al. (2009) showed that combinations of gaseous 1-MCP and ethephon have the potential to manipulate the ripening of 'Maradol' papaya. Most studies about the utilization of gaseous 1-MCP as an

alternative for extending shelf life of papaya fruits have found a challenge in the use of appropriate doses, exposition times and ripening stage of the fruit at treatment since adverse effects had been documented on the development of 'hardening' or 'rubbery' fruit texture. Because of this, the use of aqueous 1-MCP formulation can be an effective and alternative strategy to retain quality attributes without adversely affecting the ripening process of fruit.

3.2 Fruit external color

The external fruit color showed significant differences between all studied treatments on all sampling times as shown in Fig. 2A-C. Early in the experimental period, hue angle values averaged from 99 to 105° in all treated papaya fruit, and a marked decrease was observed by day 6 resulting in 77 °Hue for control fruit and near to 85 °Hue for treatments at 300 ng·kg⁻¹ 1-MCP as shown in Fig. 2A. After 9 d shelf life, all 1-MCP treated fruit exhibited delayed development of orange color in their peel or pericarp as compared to the control fruit, which could be associated with the ripening delay effect of the ethylene-blocking action of 1-MCP. Thumdee et al. (2010) observed a delay in the development of yellow color in 'Sunset' papaya treated with 1-MCP. The color results appear to be related to the trend observed in the firmness results. Sañudo-Barajas et al. (2009) and Santamaría et al. (2009) concluded that Hue angle changes were associated with changes in firmness in 'Maradol' papaya fruit, assuming that the external color of papayas can be considered a good reference for ripening.

The chroma values of papaya pericarp are shown in Fig. 2B, in which the control fruit showed the highest values. During storage, all treated fruit showed similar increasing chroma values, peaking with an average range of 50 to 52. The high chroma values are related to the higher purity of color, such as that found in the control fruit.

During the storage of the papaya fruit, the lightness (L) values showed similar patterns to those observed for the chroma values as shown in Fig. 2C. The control fruit showed the highest L values, despite a slight reduction by day 12, perhaps due to fruit spoilage that affected color. The increase in L and *b** values, not the loss of green color, has been used to describe the change in the external appearance of 'Maradol' papaya during the first stage of fruit maturity (Santamaría et al., 2009). Initial L values of between 43 and 47 were observed in all treated fruit, in which a dose 300 ng·kg⁻¹ 1-MCP applied for 60 s produced the lowest L values during the first 9 days of storage compared to the values observed in the control fruit. By day 12, the L values observed were in the range of 53-56 in for all of the papaya fruit (control and treated). The delay in orange color development in papaya fruit immersed in 1-MCP suggests that carotenoid biosynthesis and chlorophyll degradation are metabolic events regulated by ethylene (Lelièvre et al., 1997).

3.3 Fruit internal color

At the beginning, papaya fruit with internal color values of hue 57°, chroma 23, and L 60 were used as shown in Fig. 2D-F. After 3 d of storage, the hue angle showed an average decrease of 40-60% in most of the treated fruit, a condition related to the development of light orange color in the pulp. However, fruit treated with 300 ng·kg⁻¹ 1-MCP with either 30 or 60 s immersion times showed a minor change in hue angle, indicating a delay in the development of orange color in pulp as shown in Fig. 2D, although such change is unpredictable by consumers.

Internal color chroma values showed a slight decrease from the initial day to day 6 of storage, but at day 9 all treated samples showed a significant increase, indicative of color purity or saturation mostly observed in the control and 50 ng·kg⁻¹ 1-MCP treated fruit as shown in Fig. 2E. Fruit treated with 300 ng·kg⁻¹ 1-MCP for either 30 or 60 s showed a delay in the development of color purity, indicating less ripening of these samples.

Changes from yellow to orange pulp color were observed in all treated papaya fruit and were associated with the decrease in lightness as shown in Fig. 2F. A reduced decline in lightness was observed in the delayed ripening fruit tissue obtained from the 300 ng·kg⁻¹ 1-MCP treated samples, which was related to a lighter yellow-orange color. Paull et al. (1997) studied the color development of papaya fruit and observed an orange-reddish color synthesized from the inner to the outer tissue layer, from the mesocarp (pulp) to the pericarp (peel).

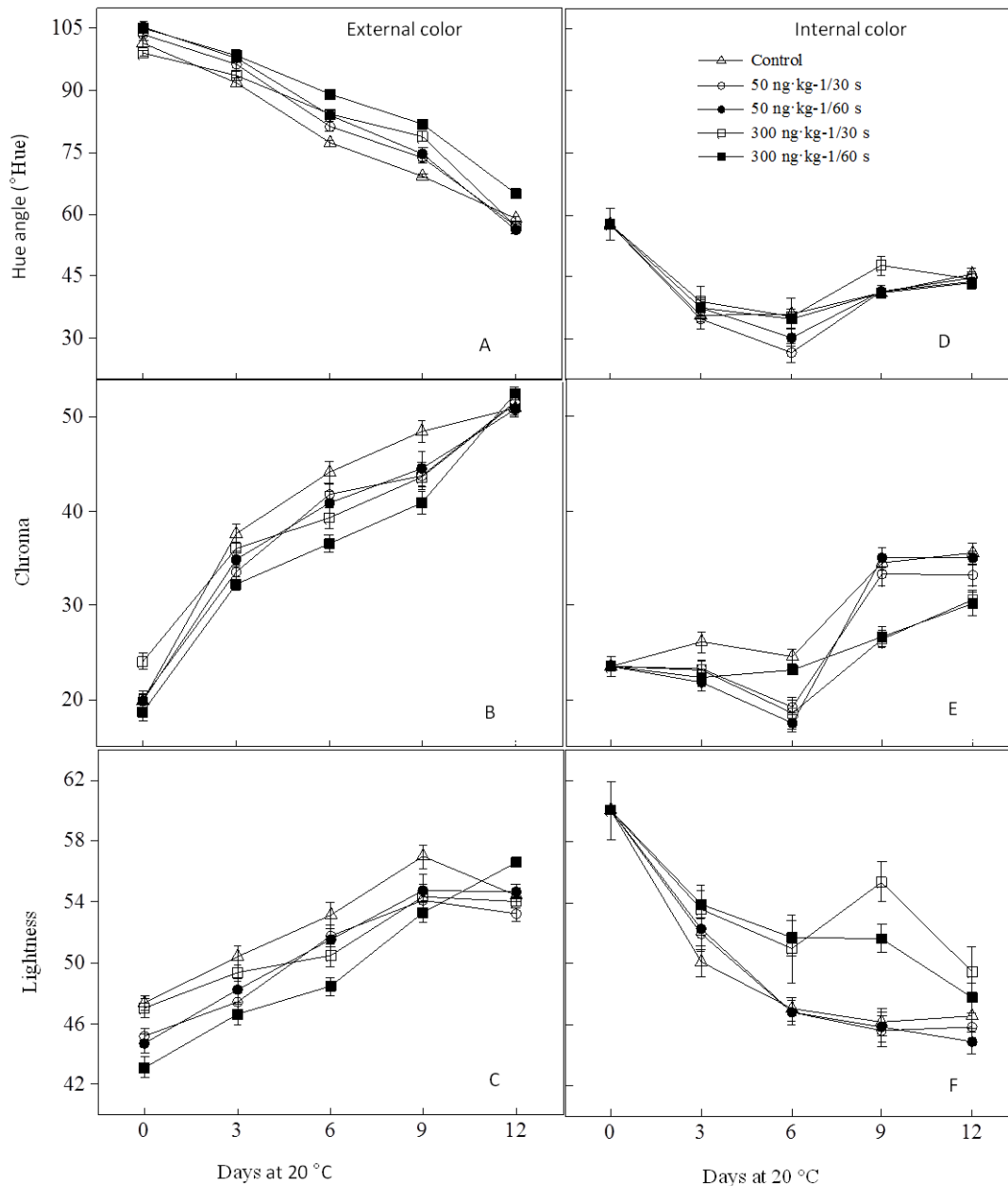


FIG. 2. External (left side) and internal (right side) color reported as Hue angle (A, D), chroma (B, E) and lightness (C, F) of 'Maradol' papayas treated with 50 or 300 ng·kg⁻¹ of an aqueous 1-MCP formulation, applied by immersion for 30 or 60 s. Bars represent standard error (n = 10 and 5, respectively).

3.4 Weight loss

Papaya fruit is considered a highly perishable produce due to the major weight loss that occurs during its postharvest storage (McGregor, 1987). In this study, all treated fruit showed an average weight loss of 5.5-7% by 9 d, which increased almost twice as much after 12 d of shelf life as shown in Fig. 3. Although no evident skin shrinkage was observed, fungi from the genera *Colletotrichum* (softness on rounded spot areas), *Penicillium* (greenish decay), *Fusarium* (pink-orange decay), and *Volutella* (black decay) associated with papaya diseases were identified (data not shown). Excessive weight loss can have undesirable effects on the overall quality of the fruit, such as a loss of firmness or color. Weight loss is usually related to the loss of water in papaya tissues and reaches the highest values during fruit marketing. Most of the water on papaya fruit is lost through the pedicel tissue ($\sim 3480 \text{ mg}\cdot\text{cm}^{-2}\cdot\text{day}^{-1}$) and, to a lesser degree, through the cuticle tissue ($16.4 \text{ mg}\cdot\text{cm}^{-2}\cdot\text{day}^{-1}$), despite the large surface area of the latter (Paull and Chen, 1989). Manganaris et al. (2008) applied $100 \text{ ng}\cdot\text{kg}^{-1}$ aqueous 1-MCP to plums and observed a reduction in weight loss, which was related to metabolic changes in cuticle waxes and also affected water vapor movement from the fruit outward. In our study, despite the observation of a delay in fruit ripening from

the 300 ng·kg⁻¹ aqueous 1-MCP treated fruit, papaya fruit showed a weight loss of 10-12% by day 12 of storage, which may have been related to the presence of fungi and further tissue deterioration. Fruits treated for less time of immersion in 1-MCP solution lost more weight and were statistically different that control ones. Fungi attack was similar in treated and untreated fruit at end of the experiment.

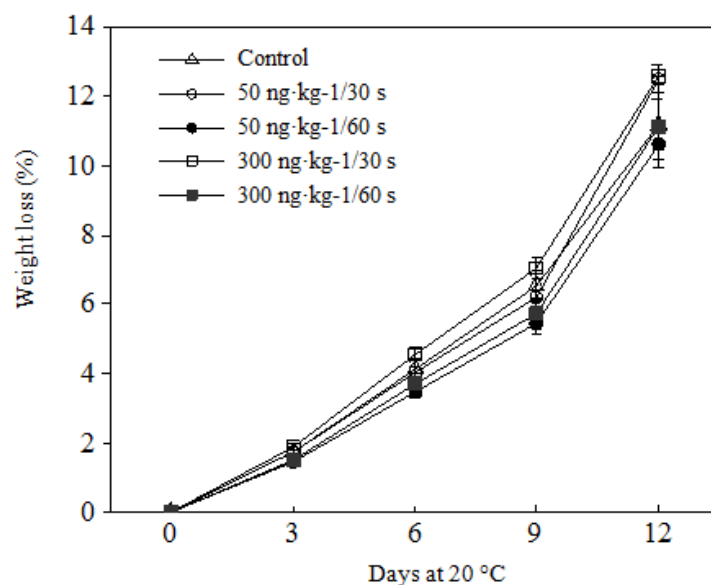


FIG. 3. Cumulative weight loss (%) of 'Maradol' papayas treated with 50 or 300 ng·kg⁻¹ of an aqueous 1-MCP formulation, applied under immersion conditions for 30 or 60 s. Bars represent standard error (n = 10).

3.5 pH, titratable acidity, and soluble solids concentration

The pH levels of all treated papaya fruit averaged 5.3-5.8 during storage and increased slightly by day 6, particularly in fruit treated with 300 ng·kg⁻¹ 1-MCP for 30 s as shown in Fig. 4A. After day 6, pH values were reduced in all treated fruit, which were more evident in the control and 50 ng·kg⁻¹ 1-MCP treated fruit. All treated fruit did not show significant changes in titratable acidity (TA) during storage, exhibiting 0.06-0.12% of malic acid equivalents as shown in Fig. 4B. Similar results have been published for 'Maradol' (Sañudo-Barajas et al., 2009) and 'Solo' papaya during fruit ripening (Cano et al., 1994).

Papaya fruit treated with 300 ng·kg⁻¹ 1-MCP for 30 s showed a slight reduction in TA from 0.09% to 0.06% at 6 d of storage. Additionally, the highest TA values were observed in the control and 50 ng·kg⁻¹ 1-MCP -treated fruit, contrary to the values observed at the lowest pH levels and similar to the results of Sañudo-Barajas et al. (2009) in stored papaya fruit as shown in Fig. 4A and B). 300 ng·kg⁻¹ 1-MCP treated papaya fruit showed a reduction in titratable acidity, assuming they were used as metabolic energy sources in cells, which corresponded to an increase in pH during the first 6 d of storage. In contrast, the control and 50 ng·kg⁻¹ 1-MCP treated fruit did not show significant changes in either TA or pH values, suggesting that these treated fruit used other substrates for cellular metabolic energy processes apart from the organic acids from the papaya fruit.

The initial SSC of the studied fruit was 10 % as shown in Fig. 4C. By storage day 3, fruit treated with 300 ng·kg⁻¹ 1-MCP for 30 s showed an increase in SSC up to 12.8 %. However, during the following 6 days of storage, the SSC values showed a reduction down to the 10 % initial level. The control fruit showed an increase from 10 to 12 % on day 3 and maintained that SSC level throughout storage. Similar results were observed by Santamaría et al. (2009) in 'Maradol' papaya, showing a good correlation between SC and firmness. It was observed that the longer immersion times for both 1-MCP doses coincidentally had lower SSC throughout the storage period, while the shorter 1-MCP immersion times and control fruit had the highest SSC, mostly observed during the first few days of storage. Our results, suggest that a 1-MCP immersion time of 30 s for either dose allowed papaya fruit to maintain an almost unaffected normal ripening process due to the synthesis of soluble sugars (mono and disaccharides). Sañudo-Barajas et al. (2009) showed an increase in SSC in 1-MCP treated papayas after 9 d of storage, similar to our findings with either one of the 1-MCP doses applied for 60 s. Other research has shown an increase from 38.6 to 48.6 mg·g⁻¹ fw of glucose, fructose and sucrose during the ripening of 'Solo' papaya, which is associated with a large synthesis of sucrose followed by an increase in the enzyme activities of sucrose synthase and sucrose phosphate synthase (Gómez et al., 2002). It should be noted that due to the extremely low starch content (0.06%) as the main

pool for sucrose production, there must be an additional source, which could be galactose, that is perhaps produced from plant cell walls.

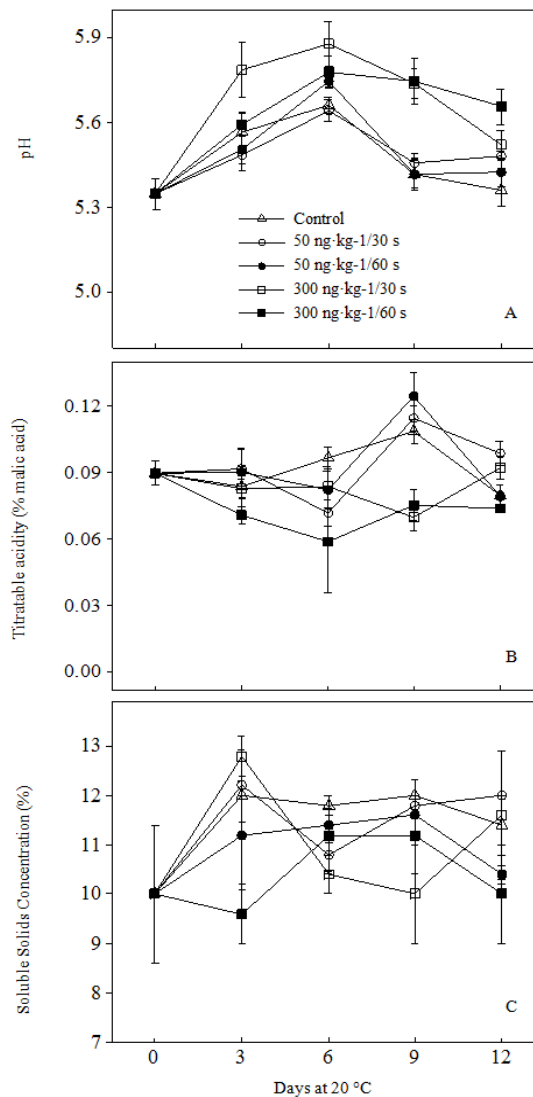


FIG. 4. pH (A), titratable acidity (B) and soluble solids concentration (C) of 'Maradol' papayas treated with 50 or 300 ng·kg⁻¹ of an aqueous 1-MCP formulation, applied under immersion conditions for 30 or 60 s. Bars represent standard error (n = 5).

IV. CONCLUSION

The use of an aqueous 300 ng·kg⁻¹ 1-MCP dose applied for 30 or 60 s delayed the postharvest ripening of 'Maradol' papaya by inhibiting fruit softening. Additionally, these same treated fruit showed the highest lightness values in their pulp (internal color), a condition related to less ripen fruit. The dose of 50 ng·kg⁻¹ 1-MCP applied for 60 s also delayed fruit softening, but only during the first 3 d of storage. As a first trial, the application of aqueous 1-MCP at a dose of 50 ng·kg⁻¹ showed promising results as a good alternative technology to extend the postharvest shelf life of 'Maradol' papayas by up to 3 days, according to flesh firmness value above 20 N, which is considered the threshold to fruit manipulation during marketing conditions. Dose of 300 ng·kg⁻¹ 1-MCP inhibited pulp softening. However, future studies should be done to establish a dose-response curve for 1-MCP, which would require the evaluation of at least five to ten concentrations and a similar number of exposure times.

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