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Research / Araştırma

EFFECT OF SOME POSTHARVEST TREATMENTS ON PHYSICAL AND BIOCHEMICAL PROPERTIES OF ANAMUR BANANAS (*Musa acuminata* Colla (AAA GROUP) DURING SHELF-LIFE PERIOD

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Abstract

Effects of modified atmosphere packaging (MAP) and 1-methylcyclopropene (1-MCP) treatments on physical and biochemical properties of Anamur bananas during shelf life period were investigated. The fruits treated with 1-MCP sustained better brightness and green color, measured with L^* and a^* values, respectively, than did the MAP treated and non-treated fruits. Exposure to 1-MCP delayed changes in skin color and flesh softening of bananas. CO_2 production steadily increased over time, with the lowest rate in 1-MCP treated samples. The lowest ethylene production was observed in the fruits treated with 1-MCP. The PPO activity in 1-MCP and MAP treated fruit flesh was erratic.

Key Words: Anamur banana, shelf-life, MAP, 1-MCP, postharvest changes

BAZI HASAT SONRASI UYGULAMALARININ RAF ÖMRÜ BOYUNCA ANAMUR MUZUNUN (*Musa acuminata Colla* (AAA GRUP) FİZİKSEL VE KİMYASAL ÖZELLİKLERİ ÜZERİNE ETKİSİ

Özet

Bu çalışmada raf ömrü boyunca Anamur muzlarının kimyasal ve biyokimyasal özellikleri üzerine modifiye atmosfer paketlemesinin (MAP) ve 1-metilsiklopropen uygulamasının (1-MCP) etkileri araştırılmıştır. 1-MCP uygulanmış meyvelerin, MAP uygulanmış ve hiçbir uygulama yapılmamış meyvelere göre parlak ve yeşil rengini koruduğu L^* ve a^* değerleri ile ölçülmüştür. 1-MCP uygulaması, muzların kabuk rengi değişimini ve meyve eti yumuşamasını geciktirmiştir. CO_2 üretimi tüm örneklerde düzenli bir şekilde artmıştır ve en düşük artış oranı 1-MCP uygulanmış örneklerde olmuştur. En düşük etilen üretimi 1-MCP uygulanmış meyvelerde gözlemlenmiştir. 1-MCP ve MAP uygulanmış meyve etlerinin PPO aktivitesi ise değişkenlik göstermiştir.

Anahtar kelimeler: Anamur muzu, raf ömrü, MAP, 1-MCP, hasat sonrası değişimler

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INTRODUCTION

Bananas are not only among the most important fruit in world trade but also among the most consumed fruits in the world. Global annual banana production amounts to 102 million metric tons. Bananas are usually harvested in the green-ripe stage. Once ripening begins, it is irreversible and is accompanied by a change in peel color from green to yellow, conversion of starch to sugars, flesh softening and aroma development. Brown spots appear on the yellow color at the end of ripening. The ripe fruit contains many of the elements that are essential for a balanced diet. Banana contains fat, natural sugars, protein, potassium and vitamins A, B complex and C (1-6).

Banana is a climacteric fruit which has a short shelf-life at ambient temperature. It was reported that consumers in Australia would purchase more bananas at one shopping occasion if the fruits had a longer shelf life. The short shelf life of bananas is attributed to a rapid senescence process leading to deterioration in visual appearances of the fruit peel. Therefore, an increase in the banana shelf life is expected to encourage consumers to purchase higher quantities at one shopping occasion (7). In order to extend postharvest shelf life fast cooling and temperature control (at ca. 13 °C) are commonly used, which are, however, costly and an alternative low-cost method that delays ripening is required for small operators and in developing countries (5, 8). Banana is extremely susceptible to chilling injury, which leads to pitting, peel discoloration, abnormal ripening, hardening of the central placenta, complete loss of flavor, flow of clear latex, subepidermal brown streaking, appearance of water-soaked areas and abnormal high susceptibility to mechanical damage and decay (9).

Modified atmosphere packaging (MAP) is a technique used for prolonging the shelf-life period of fresh or minimally processed foods. In this preservation technique the air surrounding the food in the package is changed to another composition. (10). Recently, a novel gaseous anti-ethylene compound 1-methylcyclopropene (1-MCP) has been reported to have inhibitory effects on ethylene action. Thus, 1-MCP has potential for the commercial control of ripening and senescence of harvested fruits and vegetables. 1-MCP treatment extended the green life and/or inhibited ripening of tomato, banana and plum fruit. 1-MCP is an inhibitor of ethylene action. 1-MCP delays postharvest ripening of several fruits, including banana (8, 11, 12).

According to Turkish Statistical Institute, annual banana production in Turkey was about 200 thousand tons in 2012, with Anamur banana (*Musa acuminata Colla* (AAA Group)) being the most common cultivar grown in Turkey, which is dwarf variety (13, 14). The present study was undertaken to investigate effects of 1-MCP and MAP treatment on physical and biochemical properties of Anamur banana during shelf life period.

MATERIAL AND METHODS

Plant material

Anamur banana fruits were harvested at green stage from the plantation of Alata Horticultural Research Station Directorate, Mersin, Turkey. The experiment was a completely randomized design (CRD) with 3 replications. For each replication 10 banana fingers were used.

Pretreatments

The hands of green bananas were dipped into fungicide for (0.2% imazalil) and air-dried. Uniform, undamaged fruits were selected, separated, and randomized for use in experiments. 10 fruits in each recurrence of each application were placed in open plastic containers, which were then taken to ripening room where they were exposed to 33.3 ppm/m^3 ethylene gas at 20 °C. The fruits were ripened to the ripening stage 3 (more green than yellow). In study, smartfresh commercial formulation was used in tablet form with an activator. Each tablet used in the study, providing a volume of 1 m^3 312.5 ppb concentrations. The fruit were treated with for the area 312.5 ppb/m^3 1-MCP in a gas tight room, and then kept at 20 °C for 24 h. For modified atmosphere packaging (MAP) Xtend® plastic bags were used. These packages have been developed specifically for bananas, are packages with macro and micro-holes, modified atmosphere and modified humidity technology. The fruit in plastic were placed in Xtend® bags and then sealed. The 1-MCP and MAP treated fruits and control group were placed in a shelf at 20 °C. The samples were taken at two days intervals starting from the day zero.

Color

The skin color of banana samples was determined using a Hunter Lab Colorimeter, (LabScan Minolta CR-300 model, Minolta Ramsey, NJ, USA). Instrument was calibrated with a black and white standard tiles. The results were expressed in terms of L* values and hue angle. L* values represents lightness. Hue angle, is a color circle, red-purple color between 0°-360° angle, yellow 90° angle, bluish-green color between 180° -270° angle (15).

Firmness

Fruit peel and flesh firmness were determined in three selected fruits in each replicate using a penetrometer (Fruit Pressure Tester, Model FT 327, Italy) fitted with an 6 mm diameter probe. Results were expressed as N/cm^2.

Titratable acidiy (TA) and total soluble solids (TSS)

50 grams of flesh tissue was homogenized in 250 mL deionised water and then filtered. TA was determined by titrating filtrate (10 mL) with 0.1 N NaOH using phenolphthalein as indicator and expressed as percent malic acid. 50 g of samples in 250 mL of distilled water were homogenized and then filtered. Brix was measured with a hand refractometer (Atago N-20 Brix 0-20 %, Japan,). Results were expressed as % total soluble solids (16).

Carbon dioxide and ethylene

Carbon dioxide and ethylene production were measured by enclosing fruit samples in an airtight a volume of 2.5 liters container for 1 h at room temperature. CO_2 production within jars was monitored by measuring the CO_2 using dual Gas Analyzer (United Kingdom). The results were expressed as (mL CO_2 / kg.h) by the following formula) (17).

$$(mL\ CO_2/kg.h) = \frac{\text{Gas Concentration Convertion x Vessel's Volume (mL)}}{\text{Fruit Mass (kg) x Time (h)}}$$

Ethylene production within jars was monitored by measuring the ethylene using ethylene analyzer (ICA56, United Kingdom). The results were expressed as (µL/kg.h) by the following formula) (17).

$$(\mu L/kg.h) = \frac{\text{Gas Concentration Convertion (ppm) x Vessel's Volume (L)}}{\text{Fruit Mass (kg) x Time (h)}}$$

Total Phenolics

The total phenolic concentration in the samples was determined spectrophotometrically according to Folin-Ciocalteu colorimetric method (18). 50 g of peeled fruit or 25 g peel was homogenized in 50 mL methanol (80%) containing 0.5 g ascorbic acid. The homogenate was filtered thorough cheesecloth. One mL sample extract was introduced into a 100 mL flask, 5 mL of Folin-Ciocalteu reagent, 60 mL of dionized water and 15 mL sodium carbonate (20%) were added, and the content was mixed. Afterwards, the volume of the flask was made up to 100 mL with dH_2O and allowed to stand for 2 hours in the dark. Absorption at 765 nm was measured in a Shimadzu 300 UV-vis spectrophotometer (Shimadzu UV-1700, Kyoto, Japan). The total phenolic content was expressed as gallic acid equivalents in mg/L, using a standard curve generated with 50, 100, 150, 250 and 500 mg/L of gallic acid. Total phenolics were expressed as gallic acid equivalents (mg/L). The measurements were carried out in triplicate and the results are expressed as the means.

Polyphenol oxidase (PPO) activity

20 grams of material were homogenized in 100 mL of 0.1 M sodium phosphate buffer pH 6.8 containing 0.176 g ascorbic acid and 0.5 g polyvinylpyrrolidone, 0.5 mL Triton x-100, 0.0174 phenylmethylsulfonyl fluoride, and extracted with the aid of a magnetic stirrer for 1 h. The crude extract samples were centrifuged at 10000g for 20 min. The process was conducted at the temperature of 4 °C. The supernatant was used as the enzyme source. PPO activity was determined in 1.0 mL assay mixtures in a spectrophotometer by measuring the increase in absorbance at 410 nm at 30 °C. The initial rate was calculated from the slope of the linear part of the absorbance-time curve. The reaction mixture consisted of 0.1 mL of enzyme solution and 0.9 mL of catechol in phosphate buffer (pH 6.8 0). In all experiments, control experiments without enzyme were conducted and no significant oxidation of substrate was observed during the short period employed to measure PPO activity. One unit of enzyme activity was defined as the amount of enzyme that caused an increase of 0.001 in the absorbance value per min under the assay conditions (19).

Statistical analysis

Statistical design was a completely randomized with a factorial arrangement for treatments. There were three replicates per treatment. Treatment effects were examined for significance using a least significant difference test (LSD) at the 5% level, using computational statistical program JUMP.

RESULTS AND DISCUSSION

Firmness

Changes in fruit peel and flesh firmness after 1-MCP and MAP pretreatment is shown in Fig. 1 A and 1 B. Fruit peel and flesh firmness decreased with storage at a faster rate in the untreated fruits than in MAP and 1-MCP treated fruits. However, the banana fruits exposed to 1-MCP had a higher peel and flesh firmness, suggesting that 1-MCP treatment was more effective than MAP in delaying fruit peel and flesh softening. Effect of the

treatments on peel firmness was found to be statistically insignificant at $P < 0.05$, while that on fruit flesh was significant. Ketsa et al. (2013) reported that a combination of 1-MCP and MAP treatment led to a gradual decrease in peel firmness and there was a small change in peel firmness during storage period. They also found that flesh firmness in MAP treated samples declined more rapidly than that of the control and 1-MCP (12).

Vilas-Boas and Kader (2006) reported that exposure to 1 mL/L 1-MCP for 6 h at 14 °C delayed the softening of fresh-cut banana slices stored for 3 days at 10 °C when applied after processing. However, they did not observe any effect of 1-MCP on the firmness of the slices stored for 2 days at 10 °C when it was applied on intact fruits. They asserted that the time between the 1-MCP application and peeling did not suffice to permit the permeation of the 1- MCP from peel to pulp (20). Softening of fruits is related to a change in cell wall component and starch degradation. The starch granules, packed in the tissue of banana flesh give rise to the toughness of the unripe fruit, and are hydrolyzed to sugar while an increase of the cell wall solubility allows water and nutrients to pass in and out of the cells (1). Jiang and Joyce (2003) reported that softening of bananas exposed to 1-MCP for 12 h followed by 5 days of storage in high O_2 atmospheres at 25 °C was enhanced with increasing O_2 concentration between 21 and 100% (21).

Titratable acidiy, total soluble solids

Reduced respiration rate may be reflected in lower changes titratable acidity (TA) and total soluble sugars (TSS) (22). Changes in titratable acidity (TA) of control and 1-MCP and MAP treated fruits are given in Fig. 2 A. As seen in the graph, TA steadily dropped in all groups during storage, with control group having the lowest TA. Effect of the treatments on TA was found to be significant at $P < 0.05$. The drop in TA may be explained by the fact that organic acids in banana were used as substrates for the enzymatic reactions of respiration (22). It was reported that acidity in the banana was at a maximum when the fruits turned yellow (9).

Changes in total soluble solids after MAP and 1-MCP treatment are given in Fig. 2B. TSS in treated and non-treated fruits fluctuated during storage. There was very little change in TSS in the first 2 days. It subsequently reached a peak on day four, followed a by a slight decrease. Effect of the treatments on TSS was not statististically significant at $P < 0.05$. Baez-Sanudo et al. (2009) who studied extending the shelf-life of bananas with 1-MCP and a chitosan-based edible coating reported a decrease of 0.15% titratable acidity and an increase of 8°Brix were observed in all treatments during the experiment (23).

CO_2 and ethylene production

When plant products are harvested, living cells continue to respire, releasing CO_2 at the expense

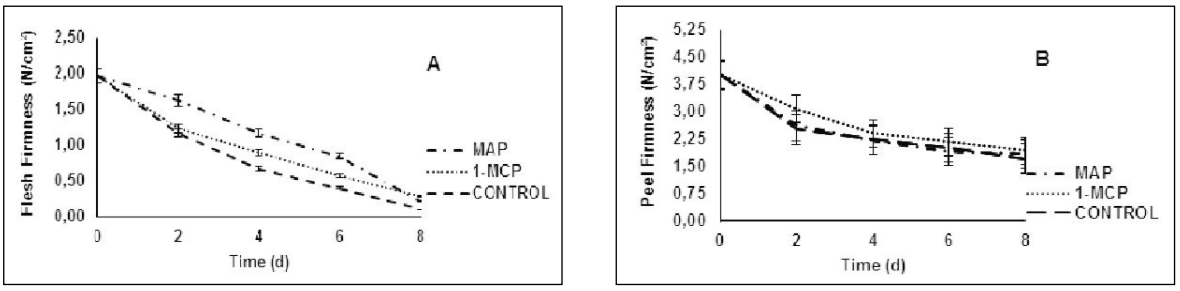


Figure 1. Effect of MAP and 1-MCP on (A) flesh firmness (B) peel firmness (N/cm²) Vertical bars indicate standard deviations of means

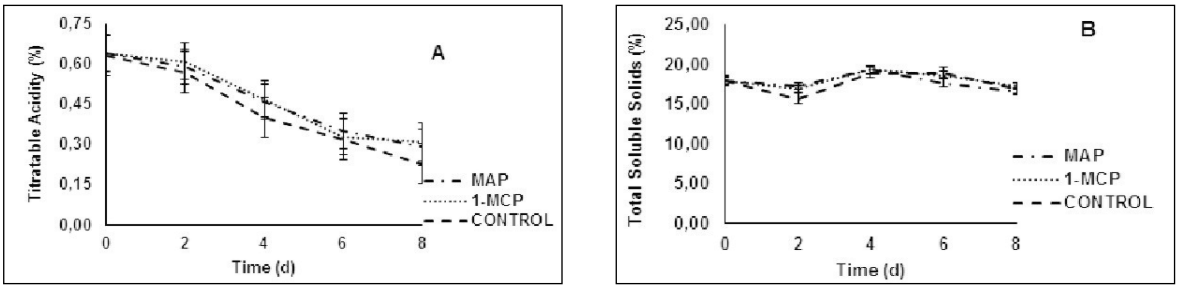


Figure 2. Changes in titratable acidity (A) and total soluble solids (B) of banana flesh after MAP and 1-MCP treatments (%). Vertical bars indicate standard deviations of means

of breaking down some substances in the cell in redox reactions (24). It is a major factor contributing to the post-harvest losses of perishables. The storage air temperature and its composition in terms of O_2, CO_2 and ethylene affect the post-harvest respiratory response of fresh produce. The diminution of enzymatic activities by providing low temperature, low O_2 and slightly high CO_2, in general, reduces utilization rate of substrates (i.e. carbohydrates, organic acid and other reserves) and increases post-harvest life of the fruit beyond its normal span (25).

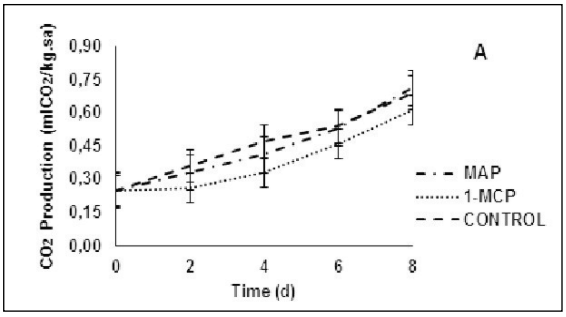
CO_2 and ethylene production by 1-MCP and MAP treated and untreated bananas are shown in Fig. 3A and 3B. CO_2 production steadily increased over time, with the lowest rate in 1-MCP treated samples, indicating that 1-MCP treatment slowed down respiration of the fruits (Fig. 3A). Ethylene production by banana fruit increased in both untreated and 1-MCP and MAP treated fruits over time (Fig. 3B). The ethylene production accelerated from day 4 onwards in all groups. The lowest ethylene production was observed in the fruits treated with 1-MCP, followed by MAP treated samples. Effect of the treatments on CO_2 and ethylene production was not statististically significant at $P < 0.05$.

Golding et al. (1999) reported that application of 1-MCP at 6 and 12 h after propylene treatment resulted in delays in the onset of both ethylene production and respiration. All of the 1-MCP treatments used resulted in an increase in the rate of ethylene production and a decrease in respiration rates during ripening (26). Vilas-Boas and Kader (2006) reported that when fresh-cut banana slices were exposed to different doses of 1-MCP, an increase in ethylene production was observed at third day. However, exposure of fresh-cut banana slices to 1-MCP did not affect their ethylene production during storage at 10 °C. Even as low as 0,1 mL/L 1-MCP (applied on intact fruits) increased the ethylene production of the slices (20).

Peel color (L* values and hue angle)

The experimental results for peel color changes in the treated and non-treated bananas are shown in Fig. 4 A, B. L* values remained almost constant in the first 4 days of storage after which it started to decline. Compared to MAP treatment, exposure to 1-MCP resulted in a higher L* value on day 8 (Fig. 4A).

Hue angle, on the other hand, steadily decrease from day 0 to day 8, with the lowest value in the fruits treated with 1-MCP (Fig. 4 B). Effect of the treatments on L* and hue angle was found to be statistically significant at $P < 0.05$. Decrease in hue angle corresponds to a change in color from green to yellow which is due to the breakdown



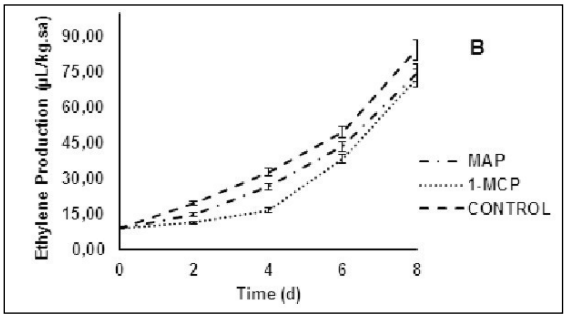
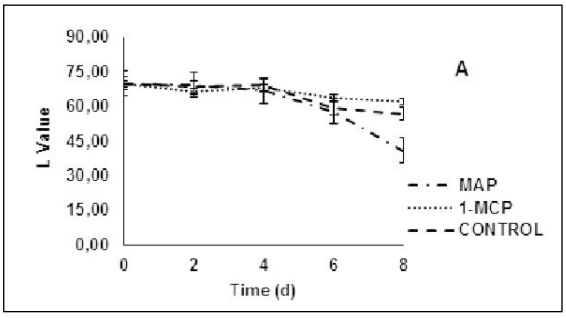


Figure 3. Changes in CO2 (A) and ethylene (B) production in non-treated and MAP and 1-MCP treated banana fruits. Vertical bars indicate standard deviations of means



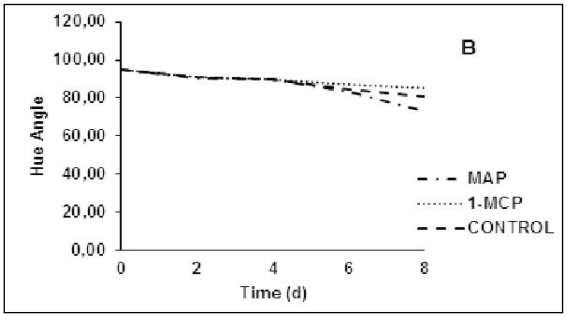


Figure 4. Effects of MAP and 1-MCP treatments on banana peel color. (A) L values. (B) Hue Angle. Vertical bars indicate standard deviations of means.

of the chlorophyll in the peel. From these findings, it can be stated that 1-MCP treated bananas sustained better brightness and green color, measured with L^* and hue angle, respectively, than did the MAP treated and non-treated fruits. In study carried out by Pelayo et al. (2003) it was found that exposure to 1-MCP delayed changes in peel color (27). According to Amiot et al. (1997), L* value was closely related to the amount of phenols degraded (28).

Total phenolics in fruit peel and flesh

Phenolic compounds are the potential main substrate for the browning reactions catalyzed by polyphenol oxidases and peroxidases (29). Change in total phenolics in fruit peel and flesh are illustrated in Fig. 5A and 5B. The level of total phenolics in fruit flesh in all groups dropped in the first two days of storage, which however reached a peak on day six in both MAP and 1-MCP treated samples, followed by a drop on day eight. The concentration of total phenolics in non-treated fruit peel remained almost unchanged during the entire storage period, that in treated fruits dropped on day two, which afterwards increased and then dropped. Effect of the treatments on total phenolics in fruit flesh and peel was statistically significant at $P < 0.05$. Nguyen et al. (2003), who investigated relationship between browning and the activities of polyphenol oxidase and phenylalanine ammonia lyase in banana peel during low temperature storage, reported that the total free phenolics level of Kluai Khai (Musa AA Group) and Kluai Hom Thong (Musa AAA Group) bananas cultivars decreased more rapidly at 6 °C than at 10 °C.

PPO activity in fruit peel and flesh

Polyphenol oxidase (PPO) (EC 1.14.18.1), which is widely distributed in plant and animal kingdom is a copper containing enzyme and is responsible for the enzymatic browning reaction occurring in many plants and vegetables. The enzyme catalyzes the hydroxylation of monophenols to *o*-diphenols to *o*-quinones. The quinones thus formed are highly reactive substances, which normally react further with other quinones, amino acids and proteins to produce dark-colored compounds, resulting in the brown pigments spotting (14, 29-31).

There was only a slight increase in PPO activity in the control fruit flesh in the first 4 days, followed by a decline (Fig. 6 A). The PPO activity in 1-MCP and MAP treated fruit flesh was erratic; after an increase in 1-MCP treated fruits on day 2, it declined until day 6, and then increased again whereas that in MAP treated fruit flesh decreased on day

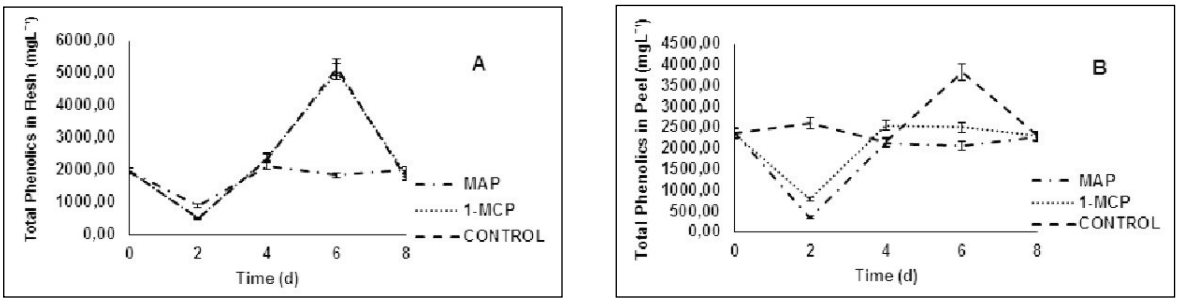


Figure 5. Change in total phenolics in fruits treated with MAP and 1-MCP (mg/L). (A) Fruit flesh (B) In fruit peel. Vertical bars indicate standard deviations of means.

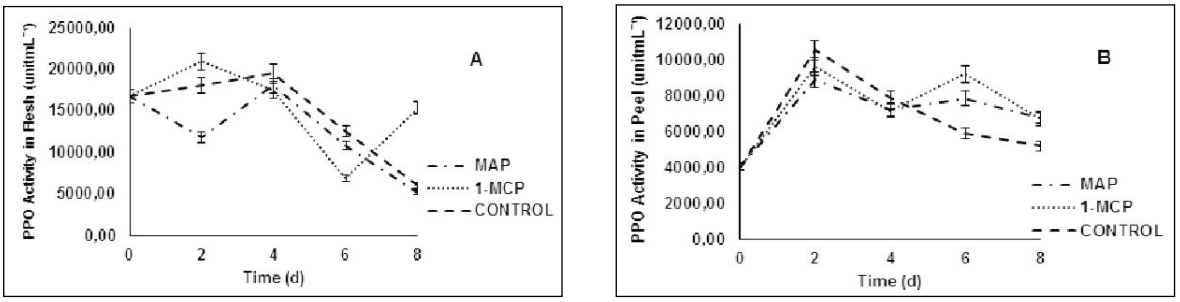


Figure 6. Change in PPO activity in fruits treated with MAP and 1-MCP (g/100 mL). A. Fruit flesh B. Fruit peel. Vertical bars indicate standard deviations of means.

2, followed by an increase on day 4 and then declined until day 8. These results indicate lack of consistency of effect of 1-MCP on PPO activity in fruit flesh, which was also reported by Pelayo et al. (2003) (27).

The PPO activity increased in both treated and non-treated fruit peel during the entire storage period, however, the increase in both 1-MCP and MAP treated fruit peel was more than that in control fruits (Fig. 6B). Effect of the treatments on PPO activity in fruit flesh and peel was found to be statistically significant at $P < 0.05$. Kamdee et al. (2009) also reported an erratic PPO activity in the peel of control banana fruits which tended to decrease during the 6 d of shelf-life (29). Nguyen et al. (2003) studied relationship between browning and the activities of polyphenol oxidase and phenylalanine ammonia lyase in banana peel during low temperature storage Kluai Khai (Musa AA Group) and Kluai Hom Thong (Musa AAA Group) bananas. They reported that with the start of chilling injury PPO activities in the peel increased, and total free phenolics decreased. The decrease in total free phenolic compounds and the increase in PPO activities occurred more rapidly at 6 °C than at 10 °C, in both banana cultivars (32).

CONCLUSIONS

The fruits treated with 1-MCP sustained better brightness and green color, measured with L^* and a^* values, respectively, than did the MAP treated and non-treated fruits. Exposure to 1-MCP delayed changes in skin color and flesh softening of bananas. CO_2 production steadily increased over time, with the lowest rate in 1-MCP treated samples, indicating that 1-MCP treatment slowed down respiration of the fruits. The lowest ethylene production was observed in the fruits treated with 1-MCP, followed by MAP treated samples. The PPO activity in 1-MCP and MAP treated fruit flesh was erratic, indicating lack of consistency of effect of 1-MCP on PPO activity in fruit flesh, which was also reported other researchers.

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