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INFLUENCES OF SOME POST-HARVEST DIPPING TREATMENTS ON GUAVA (*Psidium guajava* L.) FRUIT QUALITY AND THE INCIDENCE OF SKIN BROWNING DURING SHELF LIFE

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ABSTRACT

During 2002 and 2003 seasons the influences of post-harvest dipping treatments of 500 ppm boron (boric acid), 4.0% calcium chloride, 0.25% zinc sulfate, 10 mM sodium benzoate, 2.0 mM salicylic acid, 2.0 mM acetyl salicylic acid, 200 ppm ABG-3168 (ReTain), 0.5% plantacur-E (25.0% alpha-tocopherol) and 3.0% corn starch for 3 min on mature seedy "Balady" guava fruit quality and the incidence of skin browning during 4-days of shelf life at 25°C±3 and 50-60% RH were studied. The results showed that most fruit quality parameters were significantly influenced by the applied treatments. Except the ABG treatment, all the other treatments significantly decreased the incidence of skin browning compared with the control. In this respect, the most effective treatment was sodium benzoate followed by acetyl salicylic acid, salicylic acid, and boron treatments. Except the plantacur-E treatment, all the other treatments significantly decreased the weight loss of fruit compared with the control. Acetyl salicylic acid, sodium benzoate, boron, and calcium chloride treated fruits were significantly firmer than the control. The concentration of fruit acidity was greatly increased in all the treatments during 4-days of shelf life compared to the initial concentration. Acetyl salicylic acid treated fruit contained the highest acidity concentration followed by zinc sulfate and that was significantly higher than all other treatments. Calcium chloride treated fruit contained a significant higher concentration of TSS than all other treatments. There were clear decreases in the concentration of vitamin C (about 24% in the control) during 4-days of shelf life in all the treatments compared to the initial concentration. Boron, calcium chloride, acetyl salicylic acid, zinc sulfate, sodium benzoate, corn, and plantacur-E treated fruit showed significant higher concentrations of vitamin C than the control. However, salicylic acid and ABG treatments significantly decreased the concentration of vitamin C compared with the control. Surprisingly, there was almost no response of guava fruit to ABG application despite, it is a climacteric fruit type. The results concluded that some of the used compounds (especially sodium benzoate, acetyl salicylic acid, salicylic acid, boron, and zinc sulfate) showed clear effects on maintaining quality and decreasing the incidence of browning. However, non of these compounds, at the used concentrations, could completely eliminate the incidence of browning during shelf life. Applying higher concentrations of these compounds might further inhibit browning and better maintain quality of guava fruit during shelf life.

INTRODUCTION

The guava (*Pisidium guajava* L.) is one of the most important and common fruits which have a high nutritive value and exceptionally rich in vitamin C (Siddiqui *et al.*, 1991 and Yadava, 1996). Guavas are consumed fresh at two stages: mature green as a crispy apple-like fruit and ripe soft. Guava is a climacteric fruit (Brown and Wills, 1983 and Reyes and Paull,

1995) with a highly perishable nature and extremely short shelf life of few days, resulting in significant post harvest losses and limiting storability and/or the transportation for long distant markets. The shelf life and quality of fruits varies upon crop, year, region, maturity stage and storage environment (Rathore, 1976 and Yadava, 1996).

The ability to control skin browning and fruit softening after harvesting and thus prolonging shelf life is a critical demand if we planned to market the harvested fruits with minimal physiological and physical disorders. Fruit browning is of major concern during storage and shelf life (Jiang and Fu, 1999 and Kruger, *et al.*, 1999). The browning may render the fruit totally unmarketable or affect the appearance and ultimately the market value. The occurrence of browning increases when the stability of cell walls and particularly the integrity of membranes are disturbed. It is known from studies with other species that both boron and calcium have detrimental effects on the structural and functional integrity of cell walls and plasma membranes (Marschner, 1995; Singh, 1988 and Ferguson and Drobak, 1988). Pre-harvest boron application eliminated the incidence of brown heart in Conference pears during four months of controlled atmosphere storage. Also, boron application reduced membrane permeability at harvest and during storage and increased boron and vitamin C content of inner cortex tissue (Xuan *et al.*, 2001). It has also been reported that foliar application of aqueous solution of zinc sulphate and boron alone or in combination at a concentration of 0.3 or 0.5% improved guava fruit quality and extended their shelf life by reducing weight loss during storage (Chaitany *et al.*, 1997). Dipping guava fruit in 12% wax emulsion or 3% rice starch retained better quality than untreated fruit during 12 days of storage at room temperature (Singh and Mohammed, 1997). They suggested that a 3% rice starch treatment can be used as a substitute for wax emulsion treatments for extending the post-harvest life of fully ripe guava fruits to up to 12 days.

Ethylene is effectively connected with fruit maturity, ripening and post-harvest softening of especially climacteric type of fruits (Awad *et al.*, 2004; Graell and Recasens, 1992; Klee and Clark, 2002). Awad *et al.* (2004) found that pre-harvest application of the ethylene inhibitor ((S)-trans-2-amino-4-(2-aminoethoxy)-3-butenoic acid hydrochloride), that is commercially known as ABG-3168 or ReTain, at 500 ppm on Anna apples greatly delayed maturity and ripening, maintained fruit firmness and green color, and completely eliminated browning during two months of cold storage followed by one week of shelf life. Salicylic acid and its derivative acetyl salicylic acid have also been found to inhibit ethylene production and delay ripening in pear, apple and banana fruits, to increase the antioxidant capacity of plants and to induce multiple stress tolerance in many plants against drought, heat, chilling, and salinity (Leslie and Romani 1988; Srivastava and Dwivedi, 2000 and Senaratna *et al.*, 2000). Moreover, substances which posses antioxidant properties such as vitamin E (alpha-tocopherol) and sodium benzoate would inhibit the oxidation processes and might prolong shelf life of fruit. Vitamin E (alpha-tocopherol) has been reported to be connected with the inhibition of superficial scald (skin browning) in apples (Barden and Bramlage, 1994) and internal browning in Chinese pears (Ju *et al.*, 2000). Sodium benzoate, a free radical scavenger, has been found to retard ripening of many fruits; e.g. apple, avocado and tomato and to delay senescence in carnation flowers, partly via suppression of ethylene production and respiration rate (Baker *et al*., 1977 and 1978).

The aim of the present study was, therefore, to evaluate the influences of post-harvest dipping treatments of boric acid, calcium chloride, zinc sulphate, sodium benzoate, salicylic acid, acetyl salicylic acid, ABG-3168 (ReTain), vitamin E (alpha-tocopherol), and corn starch on mature seedy (Balady) guava (*Pisidium guajava* L.) fruit quality and the incidence of browning during shelf life. This study was an attempt to minimize the post-harvest losses and prolong the shelf life of guava fruit and thereby maintain fruit quality and marketability.

MATERIALS AND METHODS

1. Plant material and experimental design

The present experiment was conducted during the two successive seasons of 2002 and 2003 on seedy guava trees of 20-year-old originated from one mother tree (*Pisidium guajava* L.). Such trees are heavy bearing, flowering during May-June and fruit maturation starts around the end of September until the end of October. The selected trees were mostly uniform, grown in a sandy soil with spacing of 5.0X 5.0m and received the normal cultural practices in a private orchard at Damietta Governorate, Egypt.

Mature green fruit samples of similar size were picked from a uniform group of trees, on the first and the 8^{th} of October in 2002 and 2003 season, respectively. The fruits were immediately, carefully transported to the Horticulture Laboratory at Mansoura University, divided into similar groups of 75-fruits each. Each group were subdivided into three groups of 25-fruits each, and subjected to one of the following dipping treatments at ambient conditions ($25^{o}C\pm3$ and 50-60% RH) for 3 min: 500.0 ppm boron as a boric acid, 4.0% calcium chloride, 0.25% zinc sulfate, 10.0 mM sodium benzoate, 2.0 mM salicylic acid, 2.0 mM acetyl salicylic acid, 200.0 ppm ABG-3168 (ReTain), 0.5% Plantacur-E (a vitamin E formulation containing 25.0% alpha-tocopherol) and 3.0% corn starch. All treatments were combined with 0.1% Tween 20 (polyoxyethylene sorbitan monolaurate) wetting agent. A control treatment in which fruits were dipped in only water plus 0.1% Tween 20 wetting agent. After dipping, the fruits were stored for 4-days at ambient conditions ($25^{o}C\pm3$ and 50-60% RH) in cardboxs. A complete randomized design with 3 replicates (25 fruits each) per treatment was adopted.

2. Fruit quality, total loss in fruit weight and the incidence of skin browning determinations

At harvest, a random sample of 30 fruits were collected and used for the initial fruit quality determinations. After 4-days of shelf life at $25^{o}C\pm3$ and 50-60% RH, the following quality parameters were determined in all treatments. Total loss in weight of fruit and the incidence of skin browning were calculated and expressed as percentage. Fruit firmness was measured on two opposite sides of each fruit by a hand Effgipenetrometer supplemented with probe of 8.0 mm diameter and the results were expressed as Kg/cm^2 (Reyes and Paull, 1995). Total soluble solids (TSS) were measured in juice sample with a hand Refractometer. Titratable acidity was determined in juice sample by titrating with 0.1N sodium hydroxide in the presence of phenolephthalene as an indicator (Ranganna, 1979) and the results were expressed as a percentage of citric acid. Ascorbic acid concentration (vitamin C) was measured by the oxidation of ascorbic acid with 2, 6 dichlorophenol endophenol dye and the results were expressed as mg/100 ml juice (Ranganna, 1979).

3. Statistical analysis

The obtained data were subjected to the analysis of variance (ANOVA) and the treatments means were separated by the least significant difference (LSD) test at the 5% level using the statistical package SAS, release 5, 1996 (USA).

RESULTS

Because of similarity between the results of the two seasons (no significant interactions between seasons), data were presented as the means of both seasons.

1. The incidence of skin browning and total loss in weight during shelf life

There was no decay in any of the treatments throughout the four days of shelf life (data not shown). The data of Table 1 describe the incidence of skin browning during shelf life. Except the ABG treatment, all the other treatments significantly decreased the incidence of skin browning compared with the control.

In this respect, the most effective treatment was sodium benzoate followed by acetyl salicylic acid, salicylic acid, and boron treatments.

Except the plantacur-E treatment, all the other treatments significantly decreased the weight loss percentage compared with the control (Table 1). In this respect, the most effective treatment was calcium chloride followed by salicylic acid and acetyl salicylic acid.

Table 1: The incidence of skin browning and weight loss percentage in "Balady" guava fruits after 4-days of shelf life at ambient conditions (25°C±3 and 50-60% RH) as affected by some post harvest treatments.

Treatments	Skin browning (%)	Weight loss (%)
Control	38.67	9.69
Boron (500 ppm)	28.33	8.63
Calcium chloride (4%)	32.83	7.51
Salicylic acid (2mM)	27.23	7.78
Acetyl salicylic acid (2mM)	25.90	7.79
ABG (200 ppm)	35.83	8.17
Zinc sulfate (0.25%)	33.20	8.68
Sodium benzoate (10mM)	23.67	8.17
Corn starch (3%)	32.33	8.15
Plantacur-E (0.5%)	30.33	9.05
$LSD_{0.05}$	*3.56*	*0.71*

Data are mean of the 2002 and 2003 seasons.

2. Changes in firmness, acidity, TSS, and vitamin C during shelf life

The data of Table 2 shows that fruit firmness greatly decreased during shelf life in all the treatments compared to the initial concentration. Acetyl salicylic acid, sodium benzoate, boron, and calcium chloride treated fruits were significantly firmer than the control. However, in this respect, there were no significant differences between the other treatments and the control (Table 2).

Table 2: Firmness and acidity, TSS and vitamin C concentration of "Balady" guava fruits after 4-days of shelf life at ambient conditions (25°C±3 and 50-60% RH) as affected by some post harvest treatments.

Treatments	Firmness (Kg/cm^2)	Acidity %	TSS %	Vitamin C (mg/100ml Juice)
Initial	3.10	1.11	7.78	130.3
Control	2.27	1.76	7.08	99.2
Boron (500ppm)	2.55	1.76	7.63	100.4
Calcium chloride (4%)	2.55	2.10	8.37	106.2
Salicylic acid (2mM)	2.43	2.01	7.25	92.1
Acetyl salicylic acid (2mM)	2.65	2.34	7.33	112.5
ABG (200 ppm)	2.48	1.93	7.11	94.5
Zinc sulfate (0.25%)	2.30	2.30	7.67	104.8
Sodium benzoate (10mM)	2.57	1.71	7.75	101.4
Corn starch (3%)	2.47	1.77	7.00	105.3
Plantacur-E (0.5%)	2.28	1.87	7.25	107.6
$LSD_{0.05}$	*0.25*	*0.23*	*0.46*	*4.7*

Data are mean of the 2002 and 2003 seasons.

The concentration of fruit acidity was greatly increased in all the treatments during 4-days of shelf life compared to the initial concentration. Acetyl salicylic acid treated fruit contained the highest acidity concentration followed by zinc sulfate treated fruit and that was significantly higher than all other treatments. Calcium chloride, salicylic acid, and ABG treated fruits contained significant higher concentrations of acidity than the control. However, boron, plantacur-E corn starch, and sodium benzoate treated fruit showed no significant differences in acidity concentration than the control.

There were slight changes in TSS concentration during shelf life compared to the initial concentration (Table 2). Calcium chloride treated fruit contained a significant higher concentration of TSS than all other treatments. Boron, zinc sulfate, and sodium benzoate treated fruit showed significant higher concentrations of TSS than control. However, in this respect, there were no significant differences between the other treatments and the control.

There were clear decreases in the concentration of vitamin C (about 24%) during shelf life in all the treatments compared to the initial concentration. Boron, calcium chloride, acetyl salicylic acid, zinc sulfate, sodium benzcate, corn starch, and plantacur-E treated fruit showed significant higher concentrations of vitamin C than control. However, salicylic acid and ABG treatments significantly decreased the concentration of vitamin C compared with the control (Table 2).

DISCUSSION

Generally, fruit browning is due to oxidation process of phenolic substances. This process might be enzymatic (via oxidase enzymes e.g. polyphenol oxidase) and/ or non-enzymatic (via free radicals or the reactive oxygen species) and lead to the formation of the melanin pigment which is responsible for the brown color (Nicolas *et al.*, 1994: Kruger, *et al.*, 1999 and Jiang and Fu, 1999). Therefore, it is logic that compounds which posses antioxidant properties would inhibit the occurrence of fruit browning. Our results showed clear effects for most of the applied compounds on decreasing the incidence of skin browning and decreasing weight loss of guava fruit during shelf life (Table 1). The most effective treatment was sodium benzoate followed by acetyl salicylic acid, salicylic acid, and boron treatments. These results might be attributed to the antioxidant properties of sodium benzoate, salicylic acid and its derivative acetyl salicylic acid (Baker *et al.*, 1977 and 1978; Leslie and Romani, 1988; Srivastava and Dwivedi, 2000 and Senaratna *et al.*, 2000).

The occurrence of fruit browning increases when the stability of cell walls and particularly the integrity of membranes are disturbed. Our results on boron, partly, confirm those of Xuan *et al.* (2001) on Conference pear. They reported that pre-harvest boron application eliminated the incidence of brown heart in Conference pears during four months of controlled atmosphere storage. Moreover, boron application reduced membrane permeability at harvest and during storage and increased boron and vitamin C content of inner cortex tissue. Also, our results concerning effects of zinc sulfate on guava fruit quality confirm those of Chaitany *et al.* (1997), who found that foliar application of aqueous solution of zinc sulfate and boron alone or in combination at a concentration of 0.3 or 0.5% improved guava fruit quality and extended their shelf life by reducing weight loss during storage. In the present study plantacur-E (a formulation containing alpha tocopherol) slightly, but still significantly, decreased the incidence of skin browning of guava fruit. Vitamin E (alpha-tocopherol) has been reported to be connected with the inhibition of superficial scald (skin browning) in apples (Barden and Bramlage, 1994) and internal browning in Chinese pears (Ju *et al.*, 2000).

The concentration of fruit acidity greatly increased and vitamin C clearly decreased in all the treatments during 4-days of shelf life compared to the initial concentration (Table 2). These results confirm those of Siddiqui *et al.* (1991) who found progressive increase in acidity and decrease in vitamin C concentrations of guava fruit with the increase in storage time at ambient conditions. Corn starch treatment did not affect firmness, acidity and TSS of guava fruit but maintained slightly higher vitamin C concentration and decreased browning and weight loss compared with the control (Tables 1 and 2). These results, partly, confirm those of Singh and Mohammed (1997) who suggested that a 3% rice starch treatment can be used as a substitute for wax emulsion treatments for extending the post-harvest life of fully ripe guava fruits to up to 12 days. Possibly, different starch types have different effects on guava fruit quality parameters. Acetyl salicylic acid, sodium benzoate, boron, and calcium chloride treated fruits were significantly firmer than the control. These are expected results since it is known from studies with other species that both boron and calcium have detrimental effects on the structural and functional integrity of cell walls and plasma membranes (Marschner, 1995; Singh, 1988 and Ferguson and Drobak, 1988). Calcium is essential for the maintenance of cell wall structure, where it is involved in cross-linkage of pectic molecules in the middle lamella (Poovaiah, 1988). Also, acetyl salicylic acid and sodium benzoate is known to increase the antioxidant capacity of plants and that connected with the integrity of both cell walls and membranes (Baker et al., 1977 and 1978; Leslie and Romani, 1988; Srivastava and Dwivedi, 2000; Senaratna *et al.*, 2000). The decrease in TSS of the control fruits and other treatments compared to the initial concentration might be explained by the higher rate of catabolism of some fruit components, especially sugars, by respiration (Crisosto *et al.*, 1993).

Surprisingly, in our study there was almost no response of guava fruit to ABG application despite, it is a climacteric fruit type (Tables 1 and 2) (Brown and Wills, 1983 and Reyes and Paull, 1995). Moreover, in another study we found no response of guava fruit to the pre-harvest application of ABG (un-published data). In contrast, apple fruit showed a strong response to ABG (Awad and de Jager, 2002; Awad *et al.*, 2004). This phenomenon might be attributed to differences between the nature of fruit tissues and the sensitivity of enzymes involved in the ripening process to ABG.

In conclusion, some of the used compounds (especially sodium benzoate, acetyl salicylic acid, salicylic acid, boron, and zinc sulfate) showed clear effects on maintaining quality and decreasing the incidence of browning. However, non of these compounds, at the used concentrations, could completely eliminate the incidence of browning during shelf life (Table 1). Applying higher concentrations of these compounds might further inhibit browning and better maintain quality of guava fruit during shelf life.

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تأثير بعض معاملات النقع بعد الحصاد على جودة ثمار الجوافة وحدوث التلون البنى أثناء التخزين تحت ظروف الغرفة

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تم دراسة تأثير نقع ثمار الجوافة المكتملة النمو فى مركبات البورون (٥٠٠ جزء فى المليون)، كلوريد الكالسيوم (٤%)، سلفات الزنك (٠,٢٥%)، بنزوات الصوديوم (١٠ ملليمول)، حمض السلسيليك (٢ ملليمول)، أستيل حمض السلسيليك (٢ ملليمول). الريتان (ABG) (٢٠٠ جزء فى المليون)، بلانت أكيور-E (0.5%) ونشا الذره (٣%) كل بمفرده على خواص الجودة وحدوث التلون البنى خلال ٤-أيام من التخزين تحت ظروف الغرفة وذلك خلال موسمى ٢٠٠٢، /٢٠٠٣م.

وقد أوضحت النتائج أن معظم خصائص جودة الثمار قد تأثرت بالمعاملات المستخدمة. فيما عدا مركب الريتان (ABG) أدت جميع المعاملات الى تقليل حدوث التلون البنى معنويا مقارنة بالكنترول. وفى هذا الخصوص كانت أفضل المعاملات هى بنزوات الصوديوم، حمض السلسيليك، أستيل حمض السلسيليك والبورون. وفيما عدا معاملة بلانت أكيور-E أدت جميع المعاملات الى تقليل الفقد فى وزن الثمار معنويا مقارنة بالكنترول. وفى هذا الخصوص كانت أفضل المعاملات هى كلوريد الكالسيوم، حمض السلسيليك، أستيل حمض السلسيليك. الثمار المعاملة بأستيل حمض السلسيليك، حمض السلسيليك وكلوريد الكالسيوم.

الثمار المعاملة بمركبات أستيل حمض السلسيليك، بنزوات الصوديوم، البورون، وكلوريد الكالسيوم كانت أكثر صلابة بالمقارنة بالكنترول. أثناء التخزين ازداد تركيز الحموضة بشكل واضح فى كل المعاملات مقارنة بالتركيز الأولى عند بداية التخزين. احتوت الثمار المعاملة بأستيل حمض السلسيليك على أعلى تركيز من الحموضة متبوعا بمعاملة سلفات الزنك وكلاهما كانتا أعلى معنويا من الكنترول. أعطت الثمار المعاملة بكلوريد الكالسيوم أعلى نسبة من مجموع المواد الصلبة الذائبة مقارنة بباقى المعاملات. أثناء التخزين انخفض تركيز فيتامين ج بشكل واضح فى كل المعاملات (٢٤% فى الكنترول) مقارنة بالتركيز الأولى عند بداية التخزين. أظهرت معاملات البورون، كلوريد الكلسيوم، أستيل حمض السلسيليك، سلفات الزنك، وبنزوات الصوديوم، نشا الذرة و بلانت أكيور-E تركيزا مرتفعا من فيتامين ج مقارنة بالكنترول. وعلى العكس من ذلك قللت معاملات الريتان وحمض السلسيليك معنويا تركيز فيتامين ج مقارنة بالكنترول. ومن المفاجى أن ثمار الجوافة لم تستجيب تقريبا لمعاملة الريتان (ABG) على الرغم من أنها ثمار كلايمكتيرية.

وعموما فانه لم تؤدى أى من المعاملات عند التركيز المستخدم فى هذه الدراسة الى المنع النهائى لحدوث التلون البنى فى ثمار الجوافة أثناء ٤-أيام من التخزين تحت ظروف الغرفة. ولذلك فان استخدام تركيزات مرتفعة من هذه المركبات ربما يؤدى الى التحكم فى حدوث التلون البنى وحفظ خصائص جودة الثمار أثناء التخزين.