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Research Article

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Export- and Storage Potentiaility Improvement of Murcott Mandarins by Chitosan and Salicylic Acid

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Abstract The present study was conducted during 2015 and 2016 seasons to study the effect of postharvest chitosan and salicylic acid either alone or in combination dipping on fruit chemical quality attributes and export- and storability after 4 weeks (for simulating export conditions) and 10 weeks (simulating storage conditions) of cold storage at 7± 2°C and 85-90 % RH of Murcott mandarin fruits. The experimental design was RCBD analysed by one-way analysis of variance. Obtained results showed that chitosan and salicylic acid applied either alone or in combinations generally maintained the fruit acidity, TSS and carotenes percent, V.C, pH, and TSS/acidity after 4 and 10 weeks during both seasons. In the meantime, all postharvest dipping treatments enhanced fruits export- and storagability as it decreased the percentages of fruit weight loss, decay, unmarketable and total quantitative losses and electrolyte leakage percent during cold storage in 2015 and 2016 seasons.

Keywords agrochemicals, exportability, postharvest, storagability, quality

Introduction

Mandarins are highly perishable citrus fruits that must be harvested at full maturity to achieve maximum quality in terms of visual appearance, texture, flavor and nutritional value which should be maintained until the final consumption. For instance, maintaining vitamin C content in fruits after harvest is crucial for human health, due to its antioxidant function and also due to human disability of vitamin C synthesis [1]. Harvested fruits however, faces several physiological disorders mainly weight and dry matter losses due to respiration, as well as fruit decay due to pathogens attack which have negative influences on the fruit store- and marketability. Mandarins are the second main citrus fruits produced in Egypt and suffer from such postharvest disorders.

Giovannoni [2] stated that although citrus fruits are non-climacteric and ethylene is not required for the coordination of its ripening, ethylene would have obvious impact on the fruit peel senescence process and thus fruit deterioration after harvest. Tumminelli et al. [3] recorded ethylene increases in the albedo tissues of mandarin fruits contributing to the weakening of peel structure and leading to rind senescence and to final demise of fruits. Accordingly, delaying rind senescence and decreasing pathological disorders is a main object for the maintenance of the postharvest quality characteristics and for the extension of the postharvest life of the fruits. In the last decades, next to cold storage, synthetic fungicides were often used to preserve fruit quality characteristics after harvest. Synthetic fungicides nowadays are not appreciable to be applied in the postharvest handling process and more restrictions on their applications to the fresh product are made. This is not only because of the emergence of new race of pathogens diversity as indicated by Sompong et al. [4], but also to the public concern about food safety on human health and environment. Therefore, interests are being tried to find effective alternatives to fungicides in order to control fruit postharvest diseases and to enhance the shelf life.

Additionally, Halliwell and Gutteridge [5] reported that fruit ripening and senescence is directly associated with reactive oxygen species (ROS) such as superoxide radical (O^{2-}), hydrogen peroxide (H_2O_2) and hydroxyl radicals (OH^-) accumulation leading to membrane deterioration, lipid peroxidation, DNA mutation and ultimately economical quality and quantity losses. To overcome oxidative stress and delay fruits deterioration, and to, maintain the fruit sensory and nutritional quality after harvest, fruits cells employ an antioxidant system [6] and an effective antioxidant system in fruits could be achievable by treating the fruits with environmentally friendly safe compounds such as chitosan and salicylic acid [7-8].

Chitosan is reported as an alternative to synthetic fungicides for preventing fruit postharvest decay, extending fruit storage life and retaining the overall postharvest fresh fruit quality [9]. It has been approved by the United State Food and Drug Administration (USFDA) as a Generally Retained as Safe (GRAS) food additive. Postharvest application of chitosan and salicylic acid are evident in inducing fruit resistance and the inhibitory effects against fungal pathogens during storage as reported by Liu et al. [10]. Salicylic acid has been indicated to interfere with ethylene signaling and to inhibit the perception of exogenous ethylene which is known to be the primitive influence of the fruit senescence process and thus slowing down fruit senescence and extend its postharvest life [11].

In this view, the present study was conducted to investigate the effect of postharvest dipping application of chitosan and salicylic acid applied either alone or in combinations on the fruit transport- and storagability, as well as quality attributes maintenance especially the nutritive value of Murcott mandarins which is becoming nowadays the most planted cultivar in Egypt for internal and external marketing because of its high appearance and internal quality compared to the common local cultivar the Baladi mandarin.

Materials and Methods

Experimental site and plant materials

The present study was carried out during 2015 and 2016 seasons on Murcott mandarin fruits (*Citrus reticulata* Blanco) from 4-5-year trees budded on Volkameriana rootstock grown in sandy soil under drip irrigation system in a private orchard in Al-Hammam area, Marsa Matrouh governorate. Uniform as possible fruits (500 Kg) sorted based on size, color and free of physical injuries or disease infection were manually harvested at the commercial harvest date (end of February) The fruits were collected in filed plastic boxes then transported immediately to the postharvest laboratory of the Faculty of Agriculture, Alexandria University.

Treatments and statistical design

Murcott mandarin fruits were washed and disinfected with a 100-ppm chlorine solution for 10 minutes at 30 ±2 °C, air dried and sorted to remove any unsuitable fruits (injured and discolored). Selected fruits were divided into 9 groups presenting the treatments with 4 replicates /treatment. The fruits of each group were dipped for 3-4 minutes in water, chitosan at 3 and 6 g 1^{-1} in the first season and 2 and 4 g l^{-1} in the second one (Chi_1 and Chi_2, respectively) and salicylic acid at 2 and 4 g l^{-1} in both seasons (SA_1 and SA_2, respectively) either alone or in combinations. Treated fruits were left to dry, placed in plastic boxes (40 × 30 × 15 cm) and laid out in a Randomized Complete Block Design (RCBD) in the storage room at 7± 2°C and 85-90 % RH either for 4 (simulating transport conditions) and 10 (simulating storage conditions) weeks.

Fruit chemical characteristics

Fruit chemical characteristics were determined in a sample of 20 fruits per replicate taken after 4 and 10 weeks of cold storage, respectively. Fruit juice total soluble solids content (TSC%) recorded using a hand refractometer model ATAGO, model. N-1e. Japan and fruit pH by hand pH meter model PNS044 Hanna. Titratable acidity expressed as citric acid (g/100 ml juice) determined by titration with 0.1 N sodium hydroxide using phenolphthalein dye, and vitamin C (mg VC/100 ml juice) by titration with 2, 6-dichlorophenol-indophenol dye according to A.O.A.C. [12] and t total juice carotenes content measured at 460 nm according to Britton et al. [13].

Fruit export - and storagability

In addition to the percentage of fruit peel electrolyte leakage (EL%) determined according to Whilton et al, [14] the percentages of fruit physiological weight loss, decay, unmarketable fruits and total quantitative losses were estimated during 4 and 10 weeks of cold storage in order to express the fruit export- and storagability, respectively as follow: A sample of 10 fruits per replicate was taken, put in a plastic mesh bag and weighed at 0, 4 and 10 weeks and the physiological weight loss was calculated. Decayed fruits were separated by visual observations every week during cold storage and the percentage of fruit decay was calculated.

Also, any fruits that not acceptable for marketing including decayed, shriveled, pitted, browned were recorded in order to calculate the percentage of unmarketable fruits. Finally, total quantitative losses percent was calculated by adding percentages of unmarketable fruits plus physiological fruit weight loss.

Statistical analysis

A one-way analysis of variance (ANOVA) was set to investigate the effect of the different treatments on the measured parameters either after 4 (exportability) or 10 (storagability) weeks. Treatments means were separated and compared according to Snedecor and Cochran [15] using the least significant differences (LSD) at 0.05 level of significance. The statistical analysis was performed using SPSS (Statistical Analysis System) version 18, 2016.

Results

Fruit export- and storagability

Results presented in Tables 1 and 2 showed that all treatments, except SA_1 significantly maintained (2015) or decreased (2016) fruit electrolyte leakage during 4 weeks cold storage. Also, significant reduction in leakage percent occurred after 10 weeks cold storage by $Chi_1 \times SA_1$ and $Chi_1 \times SA_2$ (both seasons) and by SA_2, Chi_1 and Chi_2 (2016).

All treatments were effective in decreasing fruit weight loss during 4 and 10-weeks cold storing. The SA_2 treatment indicated the least weight loss percentage during 4 weeks (both seasons) and 10 weeks (2016), while, the SA_1 treatment revealed the least weight loss percent during 10 weeks cold storage (2015) compared to all other treatments.

In general, all treatments significantly decreased the percentage of fruit decay during 4 and 10 weeks of cold storage. Meanwhile, no decay incidence occurred by $Chi_1 \times SA_1$, $Chi_2 \times SA_1$ and $Chi_2 \times SA_2$ after 4 weeks and the least decay percentage during 10 weeks obtained by dipping in $Chi_2 \times SA_1$. Nevertheless, unmarketable fruits and total quantitative losses percentages were clearly decreased by all treatments during 4- and 10-weeks cold storage and in general, the treatments did not show significant difference among each other.

Table 1: Chitosan and salicylic acid dipping influence on fruit export -and storagability after 4 and 10 weeks in 2015 season

Treatments	Electrolyte leakage%		Weight loss %		Decay %		Un-marketable %		Total losses %	
Weeks	4	10	4	10	4	10	4	10	4	10
Control	54.68^{b}	61.74^{ab}	6.89^{a}	18.76^{a}	1.14^{a}	8.53^{a}	5.64^{a}	25.17^{a}	8.65^{a}	43.94^{a}
SA_1	58.20^{a}	58.53^{bc}	6.04^{abc}	12.84^{f}	0.24^{bc}	2.21^{cd}	1.07^{b}	7.78^{b}	8.36^{ab}	23.07^{bc}
SA_2	55.32^{ab}	59.97^{cbc}	5.21^{c}	15.29^{bc}	0.41^{bc}	2.55^{cd}	0.95^{b}	7.59^{b}	6.29^{cd}	22.88^{bc}
Chi_1	54.90^{ab}	60.86^{abc}	5.58^{c}	15.09^{bc}	0.43^{bc}	4.32^{b}	0.51^{b}	8.73^{b}	6.73^{cd}	23.82^{b}
Chi_2	57.68^{ab}	59.7^{8bc}	5.71^{bc}	14.48^{bcde}	0.38^{bc}	1.51^{cd}	0.21^{b}	6.71^{b}	7.04^{cd}	21.18^{c}
$Chi_1 \times SA_1$	55.97^{ab}	57.83^{c}	5.93^{abc}	15.98^{b}	0.0^{c}	1.83^{cd}	0.38^{b}	6.17^{b}	5.93^{d}	$22.14b^{c}$
$Chi_1 \times SA_2$	55.54^{ab}	57.83^{c}	6.79^{ab}	13.97^{def}	1.83^{a}	4.72^{b}	1.80^{b}	9.10^{b}	7.19^{bc}	20.98^{c}
$Chi_2 \times SA_1$	55.50^{ab}	61.18^{abc}	6.24^{abc}	13.52^{ef}	0.00^{c}	1.01^{d}	1.20^{b}	7.46^{b}	7.37^{bc}	20.62^{c}
$Chi_2 \times SA_2$	57.98^{ab}	62.28^{a}	5.62^{c}	14.11^{cde}	0.00^{c}	1.39^{cd}	1.36^{b}	7.88^{b}	6.36^{cd}	21.99^{bc}
LSD 0.05	**3.5**	**3.5**	**1.15**	**1.28**	**1.05**	**1.45**	**3.70**	**2.99**	**1.25**	**2.56**

Table 2: Chitosan and salicylic acid dipping influence on fruit export -and storagability after 4 and 10 weeks in 2016 season

Treatments	Electrolyte leakage%		Weight loss %		Decay %		Un-marketable %		Total losses %	
Weeks	4	10	4	10	4	10	4	10	4	10
Control	45.49^{a}	52.98^{a}	8.00^{a}	16.57^{a}	1.83^{a}	6.70^{a}	1.30^{a}	18.09^{a}	9.297^{a}	34.66^{a}
SA_1	43.59^{ab}	49.98^{ab}	5.10^{b}	12.74^{bc}	0.30^{b}	2.27^{cd}	0.30^{a}	4.00^{bc}	5.397^{b}	16.74^{bc}
SA_2	42.37^{b}	47.66^{b}	4.75^{b}	12.25^{c}	0.39^{b}	2.54^{cd}	1.11^{a}	4.65^{bc}	5.869^{b}	16.89^{bc}
Chi_1	41.39^{b}	48.68^{b}	5.39^{b}	12.81^{bc}	0.83^{ab}	3.67^{bc}	0.83^{a}	4.30^{bc}	6.225^{b}	17.10^{bc}
Chi_2	42.11^{b}	48.20^{b}	5.21^{b}	12.39^{bc}	0.36^{b}	1.58^{d}	0.36^{a}	2.84^{c}	5.576^{b}	14.93^{c}
$Chi_1 \times SA_1$	41.64^{b}	47.91^{b}	4.99^{b}	12.51^{bc}	0.0^{b}	1.83^{d}	0.00^{a}	5.95^{b}	4.991^{b}	15.35^{c}
$Chi_1 \times SA_2$	41.85^{b}	47.93^{b}	5.10^{b}	12.40^{bc}	0.30^{b}	4.71^{b}	1.83^{a}	2.54^{c}	6.447^{b}	18.50^{b}
$Chi_2 \times SA_1$	42.58^{b}	52.48^{a}	6.45^{ab}	14.61^{b}	0.0^{b}	1.01^{d}	0.00^{a}	3.90^{bc}	6.929^{b}	18.35^{bc}
$Chi_2 \times SA_2$	42.46^{b}	50.22^{ab}	5.56^{b}	13.33^{bc}	0.0^{b}	1.39^{d}	0.00^{a}	2.94^{bc}	5.556^{b}	16.26^{bc}
LSD 0.05	3.32	3.12	2.03	2.33	1.08	1.58	2.2	3.10	2.11	3.13

Fruit chemical characteristics

Data presented in tables 3and 4 indicated that fruits treated with either chitosan and /or salicylic acid had high TSS content after 4 weeks (both seasons) and 10 weeks (2015). Fruit TSS content was high in $Chi_1 \times SA_1$ treated fruits and was maintained by the rest of treatments after 10 weeks cold storing in 2016.

The treatments SA_2 (after 4 weeks) and $Chi_2 \times SA_1$ (after 10 weeks) indicated higher acidity content than the control in the first season only, while, TSS/acidity ratio was not affected neither after 4 nor 10 weeks.

All treatments (except $Chi_2 \times SA_1$ after 10 weeks) in 2015 led to higher pH value than the untreated control fruits, while in 2016, only the Chi_1 and $Chi_1 \times SA_1$ (after 4 weeks) and Chi1 (after 10 weeks) significantly increased the fruit pH.

A significant high vitamin C content was found in fruits dipped in Chi_2 solution after 4 weeks cold storing, as well as in fruits dipped in all chitosan and salicylic acid combinations (except $Chi_1 \times SA_1$) after 4 (both seasons) and 10 (2015) weeks cold storing. However, fruit carotenoids content maintained unaffected.

Table 3: Chitosan and salicylic acid influence on fruit chemical characteristics after 4 and 10 weeks in 2015

Treatments	TSS %		Acidity %		TSS/acidity		pH		V.C %		Carotenes %	
Weeks	4	10	4	10	4	10	4	10	4	10	4	10
Control	12.5^{b}	13.25^{b}	0.73^{b}	0.66^{b}	17.12^{a}	20.08a	3.82^{b}	4.08^{d}	97^{b}	91^{b}	22.8^{a}	29.34^{a}
SA_1	12.9^{ab}	14.15^{a}	0.80^{ab}	0.68^{ab}	16.13^{a}	20.81^{a}	4.07^{a}	4.30^{bc}	97^{b}	95^{ab}	23.7^{a}	31.10^{a}
SA_2	13.10^{a}	14.10^{a}	0.84^{a}	0.68^{ab}	15.60^{a}	20.74^{a}	4.00^{a}	4.29^{c}	99^{b}	99^{ab}	22.7^{a}	29.81^{a}
Chi_1	13.20^{a}	14.05^{a}	0.75^{ab}	0.66^{b}	17.60^{a}	21.29a	4.06^{a}	4.44^{ab}	102^{ab}	92^{b}	23.1^{a}	31.13^{a}
Chi_2	13.15^{a}	14.00^{a}	0.75^{ab}	0.66^{b}	17.53^{a}	21.21^{a}	4.11^{a}	4.52^{a}	112^{a}	99^{ab}	23.6^{a}	30.15^{a}
$Chi_1 \times SA_1$	13.25^{a}	14.20^{a}	0.75^{ab}	0.66^{b}	17.67^{a}	21.52^{a}	4.02^{a}	4.26^{c}	103^{ab}	99^{ab}	22.6^{a}	30.36^{a}
$Chi_1 \times SA_2$	13.10^{a}	14.20^{a}	0.75^{ab}	0.66^{b}	17.47^{a}	21.52^{a}	4.05^{a}	4.31^{bc}	110^{a}	103^{a}	23.7^{a}	30.13^{a}
$Chi_2 \times SA_1$	13.02^{a}	14.15^{a}	0.82^{ab}	0.73^{a}	15.88^{a}	19.38^{a}	3.98^{a}	4.19^{cd}	110^{a}	103^{a}	25.3^{a}	30.89^{a}
$Chi_2 \times SA_2$	13.10^{a}	14.27^{a}	0.78^{ab}	0.70^{ab}	16.79^{a}	20.39^{a}	4.05^{a}	4.32^{bc}	112^{a}	105^{a}	24.1^{a}	29.83^{a}
LSD 0.05	0.45	0.37	0.10	0.07	2.47	2.25	0.15	0.15	10.8	10.8	5.5	5.5

Table 4: Chitosan and salicylic acid influence on fruit chemical characteristics after 4 and 10 weeks in 2016

Treatments	TSS %		Acidity %		TSS/acidity		pH		V.C `%		Carotenes %	
Weeks	4	10	4	10	4	10	4	10	4	10	4	10
Control	11.55^{c}	12.80^{b}	0.71^{ab}	0.64^{a}	16.55^{ab}	20.00^{a}	3.67^{b}	3.92^{b}	76^{c}	68^{ab}	21.79^{a}	26.40^{a}
SA_1	12.23^{ab}	12.95^{ab}	0.75^{ab}	0.69^{a}	16.31^{ab}	18.77^{a}	3.54^{b}	4.05^{ab}	81^{abc}	68^{ab}	21.72^{a}	24.25^{a}
SA_2	12.45^{a}	13.00^{ab}	0.71^{ab}	0.66^{a}	17.54^{ab}	19.70^{a}	3.47^{b}	4.04^{ab}	78^{bc}	70^{ab}	21.47^{a}	23.80^{a}
Chi_1	12.05^{bc}	12.95^{ab}	0.71^{ab}	0.62^{a}	16.97^{ab}	20.89^{a}	3.97^{a}	4.14^{a}	78^{bc}	70^{ab}	24.15^{a}	26.82^{a}
Chi_2	12.15^{ab}	13.00^{ab}	0.71^{ab}	0.69^{a}	17.11^{ab}	18.84^{a}	3.54^{b}	4.00^{ab}	86^{ab}	70^{ab}	22.77^{a}	24.62^{a}
$Chi_1 \times SA_1$	12.40^{ab}	13.15^{ab}	0.69^{b}	0.62^{a}	17.97^{a}	21.21^{a}	3.97^{a}	4.02^{ab}	86^{ab}	70^{ab}	21.22^{a}	23.79^{a}

$Chi_1 \times SA_2$	12.45^{ab}	13.15^{ab}	0.78^{ab}	0.62^{a}	15.96^{ab}	21.21^{a}	3.54^{b}	4.05^{ab}	86^{ab}	75^{a}	20.67^{a}	22.42^{a}
$Chi_2 \times SA_1$	12.45^{ab}	13.20^{ab}	0.80^{a}	0.71^{a}	15.56^{b}	18.59^{a}	3.69^{b}	4.05.^{ab}	86^{ab}	66^{ab}	21.32^{a}	23.55^{a}
$Chi_2 \times SA_2$	12.60^{a}	13.35^{a}	0.76^{ab}	0.69^{a}	16.58^{ab}	19.35^{a}	3.64^{b}	4.04^{ab}	88^{a}	70^{ab}	19.57^{a}	22.54^{a}
LSD 0.05	0.52	0.52	0.10	0.08	2.27	3.25	0.23	0.23	8.9	8.9	4.9	4.9

Discussion

Positive influences of chitosan and salicylic acid postharvest treatments were generally addressed in the present work on the different parameters expressing the export- and storagability of the Murcott mandarins. Electrolyte leakage, physiological fruit weight loss, decay, unmarketable fruits and the total quantitative losses percent were reduced. Previous studies stated the enhancement of postharvest quality of various fruits by chitosan [16] and salicylic acid [18] applications.

Fruit weight loss through transpiration is a major cause of quality deterioration in fresh horticultural crops after harvest as it alters fruit appearance and textural quality such as softening and loss of juiciness. The primary mechanism of moisture loss from fresh fruits is by vapor-phase diffusion driven by a gradient of water vapor pressure between internal fruit and external environment [19]. If weight loss is more than 10%, fruit surface becomes prone to quality defects like wilting and shriveling and becomes sensitive for marketing [20]. Chitosan coating is indicated to form a semi-permeable protective film on the fruit surface which decreases transpiration losses and acts as a selective permeability barrier to water vapor transference through its hydrophilic part, as well as it reduces stomatal conductance, size and aperture, thus slows down fruit shriveling and maintains the fresh appearance of fruits leading to its maximal storability [21-22]. Similarly, salicylic acid is evident to close stomata and to reduce activities of respiratory pathways such as glycolysis and related enzymes and thus it suppresses respiration rate and minimizes weight loss of different fruits [23-25]. Furthermore, chitosan and salicylic acid are well-known as natural inducers of disease resistance and boosters of defense-related enzymes activities in plants [26-27]. Chitosan is evident to induce losses in the cytoplasm of fungal hyphae, inhibit mycelial growth and damage the spore plasma membrane of several pathogens. This reduction in fruit weight loss and decay percent would give impact on reducing the number of shriveled and unmarketable fruits leading to lower total quantitative losses.

In the meantime, a common feature accompanying senescence is increased membrane permeability, expressed as increasing leakage of ions [28]. Therefore, electrolyte leakage has been recommended as a valuable criterion for identification of fruit quality as it is reported to express injury degree of harvested fruits [29]. In the present investigation, the chitosan and/ or salicylic acid dipping generally lowered the electrolyte leakage or kept it from rising. This would be because of their influence in lowering activity of cell wall-degrading enzymes and maintaining membrane stability [27, 30-32].

In general, chitosan and/or salicylic treated fruits had high or maintained the TSS, acidity and carotenoids contents as well as the pH value after 4- or 10-weeks cold storage. These results are in line with findings of Zhu et al., [33] for chitosan and of Razavi et al., [18] and Ahmad et al., [34] for salicylic acid. Seehanam et al., [35] stated the effect of fruit coating in decreasing the respiration rate by modifying the fruit internal atmosphere i.e. decreasing O_2 and increasing CO_2. As a result of decreasing the respiration process, the use of organic acids as substrates for respiratory metabolism is minimized [36] and ascorbic acid (VC) oxidation is decreased [37]. Accordingly, this might clear the positive effect of the different treatments on maintaining the content of TSS, VC and acidity in the present study and thus maintaining the fruit nutritive quality.

Conclusion

Accordingly, from the previous mentioned it is cleared that the different treatments helped in maintaining the fruit visual and chemical quality of the Murcott mandarin during both exportation and storing. Therefore, it could be concluded that fruit dipping in chitosan and/or salicylic acid would be used successfully as postharvest treatments to be commercially applied not only to maintain the fruit quality during marketing and storing as visual appeal, but also to provide additional health benefits (enhancing levels of bioactive compounds such as VC in fruits) which all would result in improving the fruit marketability.

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