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Research Article

Evaluating the Efficacy of Moringa Aqueous Leaf Extract as A postharvest Treatment for Preserving the Quality of Valencia Oranges

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Abstract:

The objective of this study was to evaluate the impact of postharvest treatment with moringa aqueous extract on the quality attributes of Valencia oranges under refrigerated condition (18±2°C and 85 % RH). Valencia oranges were treated with moringa aqueous extract (0.0, 0.5 1.0 and 3.0 %). Cleaned healthy two hundred and forty Valencia oranges were selected randomly and divided into 4 groups (60 oranges for each) and every group was divided to 4 subgroups (for the four storage periods). The treated Valencia oranges were stored at refrigerated condition for 45 days. This experiment was arranged in a factorial design with three replicates. The data pointed to Juice content, titratable acidity and ascorbic acid were decreased, while weight loss was increased by prolonging storage period. All moringa aqueous concentrations proved efficiency in maintaining quality of Valencia oranges during refrigerated condition. Moringa aqueous extract 1.0 % showed the higher effect on keeping quality attributes of Valencia oranges up to 45 days at 18±2°C and 85 % RH + 3 days shelf life (25±2°C and 75 % RH).

1. Introduction

Citrus family considered as a best source of vitamin C (Lee and Kader, 2000) that varies with respect to climate, variety, horticultural practices storage conditions and maturity stage. Sweet orange has some medicinal importance. Oranges contain huge amount of citrus limonoids, which fight against cancer (skin, lung, breast, stomach and colon) (Milind et al., 2012). Citrus fruits classified as non-climactic and have low respiration and ethylene release during the ripening phase, thus, can be stored for up to eight weeks, depending on the variety (Reference). Citrus fruits can shrink and lose excess moisture in environmental atmosphere after harvest (Li et al., 2008). Grierson and Ben-Yehoshua et al. (1986) noted that harvesting is the most important factor affecting fruit quality during storage and transportation. Since ripe citrus fruits do not ripen again, it is best to pick them when they are fully ripe. Citrus Postharvest deterioration is caused by cultural treatments, including harvest techniques and timing, as well as pre- and post-harvest treatments (Reference). Due to the development of both physiological and pathological diseases that cause loss of storage mass and juice, fruit deterioration and peel decay, citrus fruits decay faster (Esna and Zokaee, 2011).

Poor handling of fruits during transport to market, careless harvest date and effects of temperature and storage conditions are all associated with changes in physicochemical properties (Iqbal et al., 2012). Various techniques have been used to prolong the storage of citrus fruits and reduce changes in fruit quality, such as controlling storage conditions and applying a safe and environmentally friendly edible coating.

In recent years, consumers prefer natural food with fresh taste with fewer preservatives. Consumers are trying partial or complete replacement of the synthesized preservatives due to their negative health effects. This matter has led to an increasing interest in improving natural, harmless and eco-friendly alternatives to food preservatives in order to extend the shelf life and safety of foods. Besides, substitute processing technologies has emerged in order to produce foods with little nutritional, physicochemical organoleptic changes in the food industry (El-Kady et al., 2015).

Postharvest decay represents major losses of fruits. The application of fungicides is the most effective method to control postharvest diseases. However, chemical control faces imminent problems of residues thus, there is a growing need to develop bioactive substances to control the decay (Reference). Post-harvest weight changes in fruits are usually due to the loss of water through transpiration. This loss of water can lead to wilting and shriveling which both reduce a commodity's marketability (Reference). Edible coating the aim to inhibit ripening mechanisms and preserve the fruit from water loss and spoiling may be a viable approach to extend product shelf life (Reference). Therefore, coating fruits have become more resistance to pathogens and increasing their storage and marketing (Park, 1999). Coatings can also offer a possibility to extend the shelf life by providing a semi-permeable barrier to gases and water vapor and therefore, they can reduce respiration, enzymatic browning, and water loss that leads to reduction of water loss, oxidation reaction rates and metabolic activities (Guilbert, 1986, Baldwin et al. 1995). In this respect, natural substances in moringa extracts which rich in antioxidants had a role as an electron donor produces free radical which decrease normal respiration and transpiration as well as stomata closing (Manthe et al., 1992, Sartaj et al., 2013).

Moringa (*Moringa oleifera* Lam.) is one of the most useful trees in the tropics and subtropics of Asia and Africa. The leaf, bark, sap, root, flower and seed extracts of moringa plants possess antimicrobial and antioxidant activity, contributed by a high concentration of phenolics, vitamins and carotenoids (Busani et al., 2012; Tesfay et al., 2016). Moringa plant is the most credible but cheap alternative for not only coating fruit but also providing antimicrobial effects during postharvest storage. Moringa as a medicinal tree has numerous economic applications and utilizations in human consumption (Kasolo et al., 2010). Moringa leaves can be utilized in the food industry as a natural preservative, that can be used as an alternative to synthetic preservatives in the future, it can also be utilized to market healthier products without synthetic additives (Bukar et al., 2010). The extract of the leaves has a significant antimicrobial activity, and it is effective in preventing growth of fungi (Farooq et al., 2012).

According to previous views, the objective of this study was to investigate the effect of edible coating of Valencia oranges by moringa aqueous leaf extract concentrations, for maintaining fruit quality attributes of under refrigerated room condition (18°C and 85% RH) followed by 3 days at shelf life (25°C and 75 % RH).

Fruit physical attributes

2. Materials and Methods

The present study was carried out at the experimental laboratory of the Horticulture Department, Faculty of Agriculture, Tanta University during two consecutive seasons (2020-2021) to study the effect of post-harvest treatment of moringa aqueous leaf extract for maintaining fruit quality attributes of Valencia oranges under refrigerated Shelf life. Valencia orange fruits were picked and transferred to the Horticultural laboratory, Tanta University, immediately (during 2 hours) at ambient temperature (25°C) in plastic boxes (15 kg capacity). On arrival, the fruits were cleaned, sorted, and graded, and then the detective fruits including wounded and other disorders were excluded. The sound fruits at the same maturity stage were washed with 0.01% sodium hypochlorite solution for 2 min and then the fruits were air dried at a temperature of 45°C by air –dryer for two minutes until visible moisture on fruit surfaces disappeared completely.

2.1. Preparation of moringa leaf extract

Moringa (*Moringa oleifera* L.) leaves were brought from research farm, Faculty of Agriculture Tanta university, Egypt. Moringa leaves were air-dried under shade. The aqueous extract of moringa leaves was prepared according to the method described by Thanaa et al., (2017). Soaking 100 g of air-dried moringa leaves in 1 liter of distillated water for 24 hours. Moringa air-dried leaves were blended using a blender as suggested by (Makkar and Becker, 1996). The suspension was stirred using a homogenizer to help maximize the amount of the extract. The extract was purified by filtering twice through (Whatman No. 2) filter paper. The suspension was stirred using a homogenizer to help maximize the amount of the extract. The extract was purified by filtering twice through (Whatman No. 2) filter paper, then filtered and diluted with water to 0.5%, 1%, 3% and. Tween-20 at 0.01% was added as a surfactant.

Cleaned healthy two hundred and forty Valencia oranges were selected randomly and divided into 4 groups (60 oranges for each) and every group was divided to 4 subgroups (for the four storage periods). Every group has 3 replications (5 oranges for each) that subjected to one of the following treatments:

1. Control treatment: distilled water.
2. Moringa extract (0.5%).
3. Moringa extract (1.0%).
4. Moringa extract (3.0%).

Fruits in control and all moringa concentrations were dipped in the for 5 minutes and then left for 30 minutes to dry by an electric fan at room temperature, then all fruits sprayed with water wax (FOMESA FRUITECH) (mixed by Thiabendazole (TBZ) 0.5% and Imazalil (IMZ) 0.25% and left it to air dry.

The fruits of each treatment were filled in plastic boxes with holes. Five fruits were placed in each replicate. The treated fruits were stored at refrigerated conditioned (18±2°C and 85% RH) for 45 days. The fruits physical and chemical properties were recorded in the initial time and after 15, 30 and 45 days of refrigerated room condition (18°C and 85% RH) followed by 3 days at shelf life (25°C and 75 % RH).

2.2. Fruit physical attributes

Fruit weight loss (%)

Fruit weight loss was calculated every 15 days during storage period using a bench-top digital scale Model PC-500 (Doran scales, Batavia, IL, USA) and calculated by the following formula: Fruit weight loss (%) = [100 * (Fruit weight before storage - fruit weight after each refrigerated condition period +shelf life)/ (Fruit weight before storage)].

Juice percentage: Juice percentage was determined according to the following formula:

Juice (%) = [100× (Juice volume per fruit (ml))/(fruit weight (g)]

2.2.3. Fruit chemical attributes

Ascorbic acid (AsA) content

AsA was determined according to AOAC (2005). Briefly, samples of juice were used, oxalic acid solution was added to each sample and titrated with 2,6-dichlorophenol-indophenol dye solution and expressed as a milligram of AsA and calculated as mg/100 ml of the juice.

Total Soluble solids (TSS)

TSS was measured using a digital hand-held refractometer, 0-35% Brix Model PAL-1 3810 (Atago Co., Ltd., Tokyo, Japan) at 20°C, and the results were expressed in °Brix according to AOAC (2005).

Titratable acidity (TA%)

Titratable acidity was assayed based on the method of adopting the procedure described by AOAC (2005). Aliquot of fruit juice was taken and titrated against 0.1 N NaOH in the presence of phenolphthalein as an indicator to the end point and was calculated as grams of citric acid per 100 ml of juice.

TSS/TA ratio

TSS/TA was calculated from the values recorded for fruit juice TSS and TA percentages were determined.

2.4. Statistical analysis

This experiment was arranged in a factorial design with three replicates. The effects of treatments and cold storage period on different attributes were analyzed statistically by analysis of variance (ANOVA) using the MSTAT-C statistical package. Then means comparisons were compared using Dunkan Multiple Reng Test (DMRT) at probability ≤ 0.05.

3. Results

3.1.1. The effect of Moringa ratios on the quality of oranges during the refrigerated shelf life (18 °C and 85% RH)

Weight loss (%)

The results in Table 1 confirm that all the used moringa aqueous treatments significantly reduced Valencia oranges weight loss compared to the control group. The least weight loss was detected in oranges treated with Moringa 0.5% (0.83%), followed by fruits treated with Moringa 1% (0.94%), and Moringa 3 % (0.25%). Regarding to storage periods, the results showed that the weight loss of Valencia oranges was increased significantly with extension of the storage period, and the lowest weight loss was recorded at the initial time before storage (0.44%) and increased to 0.89% after 15 days of storage period. It increased to 1.21% after 30 days. Orange weight loss reached its peak (2.14%) after 30 days of storage.

Across all storage periods, oranges treated with 0.5% Moringa and 1% Moringa aqueous extract appeared to be superior treatments as they reduced weight loss of oranges at all periods. The lowest weight loss was detected in oranges treated with 1% after 15 days of storage period (0.88%). The results showed negative significant correlation between Weight loss and juice content (r=-0.351) (Table 7).

Table 1. Effect of moringa aqueous leaf extract treatment and refrigerated period (18±2°C and 85% RH) + 3 days shelf life (25°C and 75 % RH) on weight loss of Valencia oranges

Storage period (days)	Moringa aqueous leaf extract (%)				Means
	Control	0.5	1.0	3 .0	
0	0.46 g	0.40 g	0.43 g	0.46 g	0.44 D
15	1.09 e	0.65fg	0.88ef	0.92 e	0.89 C
30	1.93 c	0.92 e	0.94 e	1.07 e	1.21 B
45	3.15 a	1.35 d	1.50 d	2.56 b	2.14 A
Means	1.66 A	0.83 C	0.94 C	1.25 B	

Means followed by the same letters in the storage periods or treatments and interaction treatments are not significantly different at level P≤0.05 according to Dunkan Multiple Rang Test (DMRT).

Juice percentage

The data in Table 2 show a decrease in Valencia oranges juice content was increased under all used treatments compared to the control group. The highest percentage of juice was recorded in Valencia oranges treated with Moringa 0.5% (47.60%), followed by Moringa 1% (47.38%), while the lowest juice percentage was recorded in oranges treated with water (control) (45.07%).

Regarding the effect of storage periods on the orange juice content, the results in Table 2 showed that the juice percentage was decreased by advancing the storage periods compared to the initial time before storage. The lowest percentage was recorded at the last time of storage (45 days storage) (44.45%).

In all of storage periods, oranges that treated with Moringa 0.5% or under control recorded the highest juice percentages.

Table 2. Effect of moringa extract treatment and refrigerated period (18±2°C and 85% RH) + 3 days shelf life (25°C and 75 % RH) on juice content of Valencia oranges

Storage period (days)	Moringa aqueous leaf extract (%)				Means
	Control	0.5	1.0	3 .0	
0	47.02b-f	48.55bc	51.46 a	48.85 b	48.97A
15	47.37b-f	46.12ef	46.13ef	45.69ef	46.33 B
30	46.69c-f	48.12bcd	46.40d-f	40.38g	45.40 C
45	39.19 g	47.60b-e	45.53 f	45.50 f	44.45 D
Means	45.07 B	47.60A	47.38A	45.10 B	

Means followed by the same letters in the storage periods or treatments and interaction treatments are not significantly different at level P≤0.05 according to Dunkan Multiple Rang Test (DMRT).

Ascorbic acid (AsA) content

The results in Table 3 show that the ascorbic acid content of Valencia oranges showed a significant increase in the present of moringa treatments especially at the concentrations of 1 and 3 %

compared by the control group.

Also, the data shows that the ascorbic acid content was gradually and significantly decreased by extending the refrigerated shelf life up to 45 days as compared to the initial time, the highest content of the ascorbic acid was recorded at the initial time before the storage period (70.20 mg / 100 ml), and the lowest content was recorded at the last storage period (45 days) (51.40 mg /100 ml).

Interaction effect, control detected the lowest AsA value comparing with the other treatments while the highest value was recorded by Moringa 3% treatment after 15 days storage period (61.40 mg/100 ml). our data revealed negative highly significant correlation between AsA content and oranges weight loss r= -0.629) (Table 7).

Table 3. Effect of moringa extract treatment and refrigerated period (18±2°C and 85% RH) + 3 days shelf life (25°C and 75 % RH) on ascorbic acid content of Valencia oranges

Storage period (days)	Moringa aqueous leaf extract (%)				Means
	Control	0.5	1.0	3 .0	
0	69.32a	75.20a	71.60a	67.04a	70.20A
15	55.52c-e	57.16bc	55.04cd	61.40b	56.96B
30	51.44c-f	49.44d-f	56.20bc	55.40b-d	53.12C
45	47.32f	48.32ef	53.32c-f	56.68cb	51.40C
Means	55.56C	56.92BC	59.16AB	60.12A	

Means followed by the same letters in the storage periods or treatments and interaction treatments are not significantly different at level P≤0.05 according to Dunkan Multiple Rang Test (DMRT).

Total soluble solid (TSS)

The data indicate that the TSS showed no significant affect by all the moringa extract concentrations compared to the control (Table 4). the results confirmed that TSS increased significantly with the advancing of storage period and the highest TSS was detected after 45 days of storage period (11.82%). Interaction effect, moringa extract at 0.5 and 1 % treatments recorded the highest TSS values at 45-day than the other treatments.

Table 4. Effect of moringa extract treatment and refrigerated period (18±2°C and 85% RH) + 3 days shelf life) (25°C and 75 % RH) on total soluble solid (TSS) of Valencia oranges

Storage period (days)	Moringa aqueous leaf extract (%)				Means
	Control	0.5	1.0	3 .0	
0	10.83b	11.33ab	11.33ab	11.33ab	11.21BC
15	10.67b	10.83B	10.67b	11.00b	10.79C
30	11.33ab	11.67ab	11.33ab	11.67ab	11.50AB
45	11.67an	12.00a	12.00a	11.67ab	11.83A
Means	11.13A	11.46A	11.33A	11.42A	

Means followed by the same letters in the storage periods or treatments and interaction treatments are not significantly different at level P≤0.05 according to Dunkan Multiple Rang Test (DMRT).

Titratable acidity (TA)

The results in Table 5 confirm that the TA showed no significant affect by the used treatments compared to the control. While TA showed significant reduction by advancing the storage period and recorded the lowest value (1.03%) at 45 days. All the concentrations showed significant higher reduction in TA at 45 days storage than the other periods.

Table 5. Effect of moringa extract treatment and refrigerated period (18±2°C and 85% RH) + 3 days shelf life (25°C and 75 % RH) on titratable acidity of Valencia oranges

Storage period (days)	Moringa aqueous leaf extract (%)				Means
	Control	0.5	1.0	3 .0	
0	0.97a	0.94a	0.96a	0.88ab	0.93A
15	0.78cd	0.76cd	0.75cd	0.81bc	0.77B
30	0.80bc	0.75cd	0.75cd	0.69d	0.75B
45	0.20e	0.23e	0.21e	0.19e	0.21C
Means	0.69A	0.67A	0.67A	0.64A	

Means followed by the same letters in the storage periods or treatments and interaction treatments are not significantly different at level P≤0.05 according to Dunkan Multiple Rang Test (DMRT).

TSS/TA

The data presented in Table 6 indicate that the TSS/TA is significantly affected by the used treatments compared to the control. All used treatments resulted in a significant slight reduction in the TSS/TA compared to the control group. Furthermore, TSS/TA significantly increased with advancing of refrigerated room condition period, where the lowest value of TSSA/TA was recorded at the initial time (2.80), while the highest TSS/TA was recorded after 45 days of storage period (11.62). Exception 45 days storage period, all the treatments showed no significant different in TSS/TA, while showed significant higher value at 45 storage period comparing with TSS/TA values in the other periods.

Table 6. Effect of moringa extract treatment and refrigerated period (18±2°C and 85% RH) + 3 days shelf life (25°C and 75 % RH) on TSS/TA of Valencia oranges

Storage period (days)	Moringa aqueous leaf extract (%)				Means
	Control	0.5	1.0	3 .0	
0	11.16c	12.12c	11.87c	12.93c	12.06C
15	13.66c	14.31c	14.26c	13.56cc	13.99BC
30	14.22c	15.51c	15.07c	16.96c	15.50B
45	59.60a	51.41b	58.17a	61.98a	34.87A
Means	24.77AB	23.54B	24.88AB	26.48A	

Means followed by the same letters in the storage periods or treatments and interaction treatments are not significantly different at level P≤0.05 according to Dunkan Multiple Rang Test (DMRT)

Table 7. The personal correlations between the different characters of Valencia oranges during refrigerated period (18±2°C and 85% RH) + 3 days shelf life) (25°C and 75 % RH) and treated with moringa aqueous leaf extract

	Weight Loss	Juice %	TSS	TA	TSS/ TA	AsA
Weight Loss	1			.		
Juice %	-.351*	1				
TSS	.382**	-.337*	1			
TA	.222	-.186	.458**	1		
TSS/TA	-.165	.138	-.375**	-.971**	1	
AsA	-.629**	.232	-.129	.329*	-.424**	1

*. Correlation is significant at the 0.05 level (2-tailed).

**. Correlation is significant at the 0.01 level (2-tailed).

4. Discussion

Weight loss percentage increased with extending cold storage periods. Normally this occurs during the fruit storage due to respiratory process, the transference of humidity and some processes of oxidation (Ayranci and Tunc 2003). In this study, the weight of the oranges treated with moringa leaf extracts as well as the untreated fruits (control) decreased during the storage period. However, significantly lower weight loss was observed in Valencia oranges dipped in the extract of the plant than the untreated (control) fruits (Table 1). The Moringa leaf extract was able to form a coating on the oranges which reduced the rate of respiration in the fruit and therefore lowered the weight loss because an increase in respiration results in an increase in metabolic rate and higher weight loss due to the expiration of moisture from the fruit (Liamngee et al., 2019).

Ascorbic acid (AsA) is lost due to the activities of phenol oxidase and ascorbic acid oxidase enzymes during storage (Salunkhe et al., 1991). Also, Ascorbic acid is highly sensitive to oxygen and is readily oxidized when exposed to it (Hossain et al., 2006). Coatings create a modified atmosphere and limit the exchange of gases thus reducing the amount of oxygen reaching to the interior of fruit that prevents the oxidation of ascorbic acid (Baldwin et al., 1999). Moringa extracts as antioxidant coating treatments were effective in reducing the AsA loss in mango fruits during all periods of storages due to its low oxygen permeability which reduced the activity of the enzymes and prevented oxidation of AsA.

Khan et al. (2020) reported that the aqueous extract of moringa increased the AsA content of grape vines. The effect of coating on soluble solid content is probably due to reducing respiration and metabolic activity, which delays the ripening process, adjusts the internal atmosphere by reducing oxygen and/or raising carbon dioxide and suppressing ethylene evolution (Dong et al., 2004). Furthermore, the higher levels of total soluble solids in fruits coated with moringa leaf extract may be due to a reduction in the protective oxygen barrier of oxygen supply at the fruit surface, which inhibits respiration (Yonemoto et al., 2002). The titratable acidity is directly related to the concentration of organic acids present in the fruits (Kays, 1997) and the decreasing trend in the fruit acidity with the increasing storage period might be due to the oxidation of organic acid and its further utilization in metabolic processes (Obenland et al., 2011). Also, the decline in total acidity was probably due to the slow rate of respiration and metabolic processes converting citric acid into sugars as a function of applied coating with antioxidant materials (Martínez-Esplá et al., 2017, and Chanikan et al., 2015). Our data revealed that TSS/TA was significantly more correlated to TA (r=-0.971) than TSS (r=-0.375) Table 7).

Conclusion

In conclusion, this study revealed that juice content, titratable acidity and ascorbic acid were decreased, while the weight loss was increased by prolonging storage period of Valencia oranges. Postharvest applications of moringa aquas extract (0.5 1.0 and 3.0 %) proved efficiency in maintaining quality of Valencia oranges during storage period. Moringa aquas extract 1.0 % showed the higher effect on the most customary values of Valencia oranges quality attributes at refrigerated shelf life (18±2°C and 85 % RH) for 45 days.

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