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Effects of Ozonated Water Treatment and Modified Atmosphere Package on the Decay and Color of Wax Apple Fruit (*Syzygium samarangense* [Blume] Merrill & L. M. Perry) during Storage

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Wax apple is very perishable fruit at room temperature. The aim of this study is to extend its storage limit by using modified atmosphere package (MAP) and ozonated water which is a highly destructive agent for microbial. Wax apples were immersed in ozonated water (0.1, 0.2 and 0.3 ppm) for 4, 8 and 12 minutes, then placed in cardboard boxes, stored at 12°C and evaluated the effect of ozonated water on the fruit quality. Ozonated–water–treated wax apples were packed into low–density polyester (0.03 and 0.1 mm thick) or high–density polyester (0.01 mm thick) bags and stored at 12°C for the MAP storage test. Quality characteristics including color, decay and chilling injury symptom were checked every week during storage. Results showed that treatment with 0.2 ppm of ozonated water for 4 minutes is good for the low temperature storage of wax apple. The low–density polyester bag (0.1 mm thick) has an advantage over others for the storage of wax apple at 12°C. The combination of ozonated water pretreatment and MAP with 6% O₂, 10% CO₂ and 84% N₂ appeared to be the most prospective for the storage of wax apple at 12°C.

Key words: color, decay, MAP, ozonated water, wax apple

INTRODUCTION

Wax apple (Syzygium samarangenese [Blume] Merrill & L. M. Perry) has become economically important in Taiwan. The production of wax apple fruit was about 84,991 tons in 2015. Wax apple is very perishable fruit at room temperature. Postharvest deterioration of wax apple leads to unacceptable appearances and quality losses that raise serious concerns commercially. Chilling injury, mechanical injury and water loss are the three major disorders. It has a shelf life of only three to five days under ambient temperature after harvesting (Lai et al., 2012). Wax apples show pitting and skin scald after 4 days at 2°C, while slight injury occurs after 4 days at 10°C (Horng and Peng, 1983). Thin skin and dehydrated fruit are the main limiting factors in the export of wax apple. The maximum period recommended for its transportation is from 5 to 7 days, and the temperature of $12 - 15^{\circ}C$ (Lai et al., 2012). A conservative period of recommendation, 12 to 14°C with 90 to 95% RH, was suggested also by Paull and Chen (2014). Because the fruit has delicate skins outside and plenty of water inside, it is easy to crack, bruise, rot and perish. Therefore, the perishable fruit is not resistant either to long distance transportation or long-term storage.

The quality of fruit during storage depends on the storage environment and fruit microorganisms, since the activity of microorganisms can cause fruit decay. The injured areas are easily infected, facilitating rot and

decay by pathogens such as Phytophthora palmivora, Colletotrichum gloeosporioides, Penicillium digitatum, Fusarium spp., and Mucor spp. (Shu et al., 2011). Bacterial isolates of Enterobacter sakazakii, Klebsiella pneumoniae, K. planticola, Pantoea agglomerans, Chromobacterium violaceum, and Streptomyces roseochromogenus and fungal isolates of Mucor Penicillium purpurogenum, hiemalis, Aspergillus niger, A. fumigatus, and Candida tropi*calis* were identified as spoilage organisms of wax apple (Esua et al., 2017).

Ozone is a gas with a pungent, characteristic smell. It is a strong oxidant that makes ozone very effective in destroying microorganisms. In most countries, ozone has also been used in different applications in the food industry, and more recently an expert panel has recommended a GRAS (generally recognized as safe) classification of ozone as disinfectant or sanitizer for foods in the United States (Graham, 1997). Research reports that ozone is high reactivity and no harmful disinfectant to assure the quality and microbiological safety of food (Kim et al., 1999). According to reports about storage period, ozone extends fruits or vegetables shelf lives without affecting their sensory values (Wysok et al., 2006). Bactericidal effect of ozone is due to two major mechanisms. One is oxidation of sulfhydryl groups and amino acids of enzymes, peptides and proteins, and the other is based on oxidizing polyunsaturated fatty acid (Victorin, 1992). It has been proven that ozone destroys viruses inducing hepatitis A, influenza A, vesicular stomatitis, and infectious bovine rhinotracheitis (Güzel et al., 2004). The bactericidal properties of ozone have also been demonstrated in the case of Gram-positive and Gram-negative microorganisms, in both the spores and vegetative cells (Güzel et al., 2004; Restaino et al., 1995). Several

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researches have shown that treatment of ozone is a suitable method to ensure fruit and vegetable quality (Liew and Prange, 1994; Pérez *et al.*, 1999; Zhang *et al.*, 2005; Zhang *et al.*, 2011). Ozone at the concentration of 0.04 ppm appears to have potential for extending the storage life of broccoli and seedless cucumbers stored at 3°C (Skog and Chu, 2001). Ogawa *et al.* (1990) reported the inactivation of *Botrytis cinerea* spores after ozone treatment of tomato fruit. Ozone storage of blackberries suppressed fungal development for 12 days, while 20% of the control showed decay (Barth *et al.*, 1995).

The aim of modified atmosphere packaging (MAP) is to change the composition of the atmosphere around the product so that the storage life of the product can be extended. Oxygen, CO₂ and N₂ are most often used in MAP. In the modified atmosphere, recommended level of O_2 fell in the range between 1 and 5% for safety and quality of fruits and vegetables (Kader, 1986). Most fruits and vegetables aged less quickly when the level of oxygen in the atmosphere surrounding them was reduced. The significant reduction of microorganisms in the chambers with the modified atmosphere was due to the low content of oxygen, as under these conditions aerobic microorganisms, as well as molds, do not develop (Jacxsens et al., 2001). Raising the level of carbon dioxide to levels of 2% or more was also beneficial. Successful suppression of respiration of the products and activity of microorganisms by MAP could result in the fruit or vegetable products with high organoleptic quality and less decay.

To the best of our knowledge, very limited research was carried out with ozonated water as a disinfectant for wax apple. Thus, we firstly evaluated the effect of ozonated water on the quality of wax apples. Then, effects of ozonated water and modified atmospheres on the decay and color changes of wax apples during cold storage were evaluated.

MATERIALS AND METHODS

Materials

Fresh wax apple was purchased from local market. Cardboard boxes and fruit net were obtained from commercial resources.

Methods

Ozonated water treatment

Ozone was generated with an ozone generator (Gioyell ozone purifier, Taiwan). Ozone concentration was regulated at 0.1, 0.2 or 0.3 ppm with an eXact Micro 7⁺ Water Meter (Industrial Test Systems, Inc., Rock Hill, SC., U.S.A.). The generated ozone was contained in distilled water in a closed acrylic container (7L) lined with tempered glass ventilating plate. Fresh wax apple individuals were immersed into ozonated water for 4, 8 and 12 minutes in different concentrations of ozonated water. In the control, wax apple individuals were immersed into tap water for 2 minutes. Wiping the surface of the wet fruit individuals, they were wrapped with fruit net. Ten individuals each were placed in cardboard

boxes, and were stored at an 12°C room. Changes for fruit quality were monitored every week.

MAP storage

Ten-ozonated-water-treated (0.2 ppm for 4 min) wax apple individuals each were put into 25×35 -cm bags comprised of 0.03 and 0.1-mm-thick low density polyester (LDPE) film and 0.01-mm-thick high density polyester (HDPE) film, and sealed. The air of the polyester bags was replaced with a desired air composition (mixture range of 4–8% O₂, 5–15% CO₂ and 77–91% N₂). The desired gas mixture was instilled in quantity into the package after removing the air by vacuum. Samples were stored at the 12°C room and quality analysis was conducted every week.

Quality analysis

Quality parameters, including decay, color of skin and storage limit were assessed objectively. Color of fruit was measured by using a color differential meter (ZE– 2000, Nippon Denshoku, Japan) to determine CIE L* value (lightness or brightness), a* value (redness or greenness) and b* value (yellowness or blueness). Measurements were carried out to take 10 samples in triplicate and the average of L*, a* and b* values were recorded. The colorimeter was warmed up for 30 minutes and calibrated with a white standard tile: L=95.87, a=-0.86, and b=2.47.

Statistical analysis

Data were subjected to analysis of variance (ANOVA) and the means were separated by Fisher's Least Significant Difference (LSD) test at p < 0.05 significant level with three replications.

RESULTS

Effect of ozonated water treatment on decay and color of wax apple

Table 1 showed that the decay of wax apple individuals treated with ozonated water except for 0.3 ppm for 4 and 8 minutes were significant (p < 0.05) lower than non-treated samples at the first week. On the 14th day of storage, the decay of non-treated wax apples reached 90%. The decay rates of wax apple treated with 0.2 ppm of ozonated water for 4 and 8 minutes were significantly (p < 0.05) lower than those of the non-treated and treated at 12°C. It revealed that ozonated water treatment was effective on the reduction of decay of wax apple stored at 12°C. Wax apple individuals treated in 0.2 ppm of ozonated water for 4 minutes showed the lowest decay among all treatments. It can be said that ozonated water was effective in reducing bacteria on wax apple, thus increasing storage limit of wax apple.

Skin color of the wax apples was reveled as L* value (Lightness), a* value (red to green) and b* value (yellow to blue). Table 2 showed that the color difference indicator L* decreased in the low temperature during storage. It suggested that the wax apple peel darkened as storage time increased. Wax apple treated with 0.3 ppm

Treatment	Decay % of wax apples in indicated storage period (day)						
(ppm/min)	7	14					
Non-treated	$43.33^{ab} \pm 1.92$	$90.00^{a} \pm 5.77$					
0.3/4	$43.33^{ab} \pm 1.92$	$93.33^{a} \pm 3.85$					
0.3/8	$46.67^{\circ} \pm 1.92$	$90.00^{a} \pm 3.33$					
0.3/12	$26.67^{\circ} \pm 3.85$	$70.00^{\circ} \pm 8.82$					
0.2/4	$33.33^{\text{abc}} \pm 1.92$	$50.00^{\circ} \pm 3.33$					
0.2/8	$30.00^{\text{bc}} \pm 3.33$	$63.33^{\text{bc}} \pm 1.92$					
0.2/12	$33.33^{\text{abc}} \pm 5.09$	$70.00^{\circ} \pm 3.33$					
0.1/4	$26.67^{\circ} \pm 1.92$	$70.00^{\circ} \pm 3.33$					
0.1/8	$33.33^{\text{abc}} \pm 3.85$	$66.67^{\circ} \pm 3.85$					
0.1/12	$30.00^{\text{bc}} \pm 3.33$	$76.67^{\circ} \pm 5.09$					

Table 1. Effect of ozonated water on decay % of wax apples stored at 12°C

Means \pm SD in the same column with the same letters are not significantly different ($P \le 0.05$) by Fisher's protected LSD test.

Table 2. Effect of ozonated water treatment on CIE L*, a* and b* value of wax apples stored at 12°C

Treatment	L*, a* and b* value of wax apple in indicated storage period (days)									
(ppm/min)		0			7		14			
	L*	a*	b*	L*	a*	b*	L*	a*	b*	
Non-treated	$36.58^{a} \pm 2.57$	$4.51^{\circ} \pm 2.96$	$8.84^{\circ} \pm 0.94$	$35.92^{ab} \pm 2.66$	$6.24^{d} \pm 3.03$	$8.61^{\text{bc}} \pm 0.85$	_	_	_	
0.3/04	$36.58^{\circ} \pm 2.58$	$4.51^{\text{a}} \pm 2.96$	$8.84^{\text{a}} \pm 0.94$	$34.81^{\text{cde}} \pm 2.35$	$8.93^{\text{bc}} \pm 3.39$	$8.16^{\circ}\pm0.78$	-	-	-	
0.3/ 08	$36.58^{\circ} \pm 2.59$	$4.51^{\text{a}} \pm 2.96$	$8.84^{\text{a}} \pm 0.94$	$36.37^{\circ} \pm 2.79$	$7.83^{\rm bcd} \pm 5.11$	$8.64^{\rm b}\pm1.17$	$35.99^{\circ} \pm 2.53$	$7.93^{\circ} \pm 5.11$	$8.36^{\rm a}\pm1.51$	
0.3/12	$36.58^{\circ} \pm 2.60$	$4.51^{\circ} \pm 2.96$	$8.84^{\text{a}} \pm 0.94$	$34.12^{\circ} \pm 2.05$	$10.94^{\circ} \pm 3.94$	$8.12^{\scriptscriptstyle e}\pm 0.78$	$33.73^{\circ} \pm 2.34$	$10.85^{\circ} \pm 3.74$	$7.88^{\mathrm{b}} \pm 0.99$	
0.2/04	$36.58^{\circ} \pm 2.61$	$4.51^{\text{a}} \pm 2.96$	$8.84^{\text{a}} \pm 0.94$	$34.81^{\rm cde}\pm2.36$	$9.3^{\rm ab}\pm4.14$	$8.28^{\scriptscriptstyle de}\pm0.83$	$34.28^{\text{\tiny de}} \pm 2.55$	$8.90^{\rm b}\pm3.99$	$7.9^{\circ} \pm 1.30$	
0.2/08	$36.58^{\circ} \pm 2.62$	$4.51^{\text{a}} \pm 2.96$	$8.84^{\rm a}\pm0.94$	$36.24^{\circ} \pm 2.61$	$7.26^{\rm cd}\pm3.54$	$8.71^{\rm ab}\pm0.94$	$35.63^{\text{ab}} \pm 2.63$	$7.25^{\scriptscriptstyle b}\pm 3.29$	$8.36^{\text{a}} \pm 1.32$	
0.2/12	$36.58^{\circ} \pm 2.63$	$4.51^{\circ} \pm 2.96$	$8.84^{\text{a}} \pm 0.94$	$35.38^{\text{abcd}} \pm 2.84$	$9.34^{\rm ab}\pm3.96$	$8.33^{\scriptscriptstyle{\mathrm{cde}}} \pm 1.03$	$35.18^{\text{abc}} \pm 2.48$	$8.84^{\scriptscriptstyle b}\pm 3.74$	$8.26^{\rm ab}\pm0.96$	
0.1/04	$36.58^{\circ} \pm 2.64$	$4.51^{\circ} \pm 2.96$	$8.84^{\text{a}} \pm 0.94$	$34.54^{\scriptscriptstyle de}\pm3.88$	$8.56^{\text{bc}} \pm 3.53$	$8.53^{\rm bcd}\pm0.77$	$34.58^{\rm cde}\pm2.48$	$8.33^{\circ} \pm 3.22$	$8.05^{\rm ab}\pm1.06$	
0.1/08	$36.58^{\circ} \pm 2.65$	$4.51^{\circ} \pm 2.96$	$8.84^{\text{a}} \pm 0.94$	$34.96^{\rm bcde}\pm2.38$	$8.75^{\text{bc}} \pm 3.65$	$8.62^{\scriptscriptstyle bc}\pm 0.83$	$34.84^{\scriptscriptstyle bcd}\pm 2.26$	$8.23^{\scriptscriptstyle b}\pm 3.69$	$8.23^{\rm ab} \pm 1.02$	
0.1/12	$36.58^{a} \pm 2.66$	$4.51^{\circ} \pm 2.96$	$8.84^{\text{a}} \pm 0.94$	$35.57^{\rm abc}\pm2.80$	$4.47^{\circ} \pm 3.36$	$8.99^{\rm a}\pm0.89$	$34.77^{\rm cd}\pm2.47$	$3.89^{\circ} \pm 3.18$	$8.25^{\rm ab}\pm1.20$	

Means \pm SD in the same column with the same letters are not significantly different ($P \le 0.05$) by Fisher's protected LSD test.

of ozonated water for 8 minutes has the highest L* value on the seventh and 14th day of strage. Almost all a* values were increased during storage at 12°C. The a* value of non-treated samples significantly differed from those treated with ozonated water on the seventh day of storage. The result showed that wax apple treated with 0.3 ppm of ozonated water for 12 minutes has the highest a* value on the seventh and 14th day of strage. The b* value decreased after two weeks of storage; however, the changes for it were limited.

Effect of MAP storage on decay and color of wax apple

Ozonated-water-treated wax apple individuals were put into low-density polyester (0.03, and 0.1 mm of thickness) or high-density polyester (0.01 mm of thickness) bags, packed and stored at 12°C. Results showed that low-density polyester bag (0.1 mm of thickness) has an advantage over others for the storage of wax apple treated in 0.2 ppm of ozonated water for 4 minutes at 12°C. The combination of ozonated water pretreatment and MAP showed the best result for prolong the storage limit of wax apple to two weeks with 6.67% decay at 12°C (Table 3). Results of color changes were showed in Table 4. From the first week, the lightness became lower for those without MAP than all the MAP packed wax apple individuals. The lightness of packed samples increased during storage. Wax apple individuals packed in polyester bag of 0.1 mm thickness showed the highest lightness among all samples. During three weeks of storage, the red color of the skin of wax apple had no significant change but samples packed in bags with 0.03 mm thickness showed change. Unpacked wax apples became more yellowish than others on the 21th day of storage Based on these results, we found that low (Table 4). density polyethylene bags with 0.1 mm thickness is the best packaging material for wax apple in MAP storage at 12°C. The gas in MAP composed by 6% O₂, 10% CO₂, and

Thickness	Decay % of wax apples in indicated storage period (day)							
(mm)	7	14	21					
0.01	6.67^{a}	23.33ª	36.67^{a}					
0.03	6.67ª	13.33^{b}	43.33ª					
0.10	$3.33^{ m b}$	6.67°	36.67ª					

Table 3. Decay % of wax apples packed in bags of different thickness during the storage at 12°C

Values in the same column with the same letters are not significantly different ($P \le 0.05$) by Fisher's protected LSD test.

Table 4. CIE L*, a* and b* values of wax apples packed in bags of different thickness during the storage at 12°C

	Each of L*, a* and b* values of wax apple in indicated storage period (days)									
Thickness (mm)	7			14			21			
(11111)	L*	a*	b*	L*	a*	b*	L*	a*	b*	
Unpacked	30.45°	19.66ª	11.20ª	31.48^{b}	19.78ª	11.64ª	31.56 ^b	20.05ª	12.13ª	
0.01	32.26ª	18.20^{a}	11.44^{a}	33.26ª	18.43^{a}	11.36ª	31.58^{b}	19.74^{ab}	11.04^{b}	
0.03	31.72^{ab}	$16.83^{\rm b}$	11.21ª	32.59^{ab}	19.15^{a}	11.10^{a}	32.22^{b}	$18.01^{\rm b}$	$11.00^{\rm b}$	
0.10	31.42^{ab}	18.19^{a}	11.55^{a}	33.36ª	19.14^{a}	11.27^{a}	33.98ª	$19.52^{\rm ab}$	11.26^{b}	

Values in the same column with the same letters are not significantly different ($P \le 0.05$) by Fisher's protected LSD test.

Table 5. Decay % of wax apple in MAP with mixed gas during the storage at 12°C

Gas composition	Decay % of wax apple in indicated storage period (day)							
O ₂ /CO ₂ /N ₂ (%)	7	14	21					
Air	$26.67^{\text{b}} \pm 1.92$	43.33°±1.92	$66.67^{a} \pm 5.09$					
4/5/91	$23.33^{\text{b}} \pm 3.85$	40.00°±3.33	$50.00^{\text{b}} \pm 3.33$					
4/10/86	$20.00^{\text{b}} \pm 3.33$	33.33°±3.85	$66.67^{\circ} \pm 1.92$					
4/15/81	$16.67^{\text{b}} \pm 1.92$	33.33°±5.09	63.33ª±3.85					
6/5/89	$20.00^{\text{b}} \pm 5.77$	$36.67^{\circ} \pm 3.85$	$53.33^{b} \pm 8.39$					
6/10/84	$10.00^{\circ} \pm 0.00$	$10.00^{\circ} \pm 5.77$	36.67°±5.09					
6/15/79	$23.33^{\text{b}} \pm 1.92$	$40.00^{\circ} \pm 5.77$	$70.00^{\circ} \pm 3.33$					
8/5/87	$20.00^{\text{b}} \pm 3.33$	33.33°±7.70	70.00°±3.33					
8/10/82	$36.67^{\circ} \pm 1.92$	$36.67^{\circ} \pm 6.94$	63.33°±5.09					
8/15/77	13.33°±5.09	$26.67^{\text{b}} \pm 1.92$	$60.00^{ab} \pm 6.67$					

Mean \pm SD in the same column with the same letters are not significantly different ($P \leq 0.05$) by Fisher's protected LSD test.

84% N_2 showed the lowest decay of wax apple among treatments during storage (Table 5). The colors of wax apples during storage were shown in Table 6. At the end of storage, wax apples in MAP with mixed gas of 6% O_2 + 10% CO_2 + 84% N_2 showed a lighter and redder color as well as a smaller color difference of skin than those in MAP with air. Instead of the air in MAP, mixed gas has the advantage of improving the decay rate and color of MAP wax apple individuals stored at 12°C. Modified atmosphere package with 6% O_2 , 10% CO_2 and 84% N_2 appeared to be most prospective for the storage of wax apple at 12°C.

DISCUSSION

The microorganisms usually responsible for vegetable spoilage are gram-negative bacteria, particularly *Erwinia* and *Pseudomonas* (Barriga *et al.*, 1991). Inadequate field practices and postharvest handlings may result in serious infections caused by microbes and decays of horticultural crops during storage. Postharvest treatments with synthetic chemicals are commonly used to minimize postharvest losses (Palou *et al.*, 2015). According to principles of sustainability and the concern of environmental protection, the consumers require a safer and healthier food production nowadays. To reduce

Table 6. CIE L*, a*, and b* values of wax apple in MAP with mixed gas during storage at 12°C

Gas	Each of L*, a* and b* values of wax apple in indicated storage period (days)									
composition	mposition 7			14			21			
O ₂ /CO ₂ /N ₂ (%)	L*	a*	b*	L*	a*	b*	L*	a*	b*	ΔE^*
Air	30.79 ± 3.39	15.45 ± 2.61	12.09 ± 3.16	34.06 ± 2.46	17.43 ± 3.27	11.80±1.79	31.86 ± 3.73	15.51 ± 3.48	12.16 ± 2.45	3.21
4/5/91	32.24 ± 4.03	17.34 ± 4.03	11.55 ± 4.09	33.78 ± 2.80	17.65 ± 3.48	11.78 ± 2.12	31.42 ± 3.21	13.48 ± 3.10	11.20 ± 1.88	3.96
4/10/86	31.07 ± 3.87	14.04 ± 3.43	10.90 ± 2.27	34.75 ± 1.91	16.81 ± 3.72	11.47 ± 1.28	32.67 ± 2.59	14.31 ± 4.08	10.31 ± 1.99	1.73
4/15/81	31.67 ± 3.23	13.13 ± 3.27	10.39 ± 1.85	33.90 ± 2.34	17.36 ± 3.41	10.60 ± 1.69	31.81 ± 2.76	13.58 ± 4.65	9.70 ± 2.74	0.83
6/5/89	31.53±3.82	14.51 ± 3.96	9.56 ± 1.69	32.61±2.84	14.07 ± 3.77	10.07 ± 3.13	33.09 ± 3.19	13.82 ± 4.31	10.28 ± 2.09	1.86
6/10/84	32.14±3.00	14.57 ± 3.61	10.34 ± 2.60	31.62 ± 2.13	14.50 ± 2.88	10.34 ± 2.77	33.33±2.22	16.32 ± 4.62	10.97 ± 1.90	2.21
6/15/79	31.85 ± 2.85	13.89 ± 4.31	10.51 ± 1.99	32.98 ± 2.52	13.71 ± 4.08	10.19 ± 2.13	32.76 ± 2.97	13.37 ± 3.91	11.80 ± 3.39	1.66
8/5/87	33.34±3.71	16.14 ± 5.08	10.91 ± 1.67	33.40±3.63	16.05 ± 3.61	10.57 ± 1.98	33.71 ± 2.96	13.66 ± 4.30	11.34 ± 4.31	2.54
8/10/82	33.25 ± 2.90	16.48 ± 3.95	11.26 ± 2.47	34.85 ± 2.26	16.36 ± 3.00	11.53±1.76	33.84 ± 3.54	12.68 ± 3.54	10.59 ± 2.53	3.90
8/15/77	34.59 ± 2.46	16.04 ± 3.18	12.76±1.52	35.02±2.10	16.65 ± 2.82	11.73±1.40	33.14±2.99	14.25 ± 3.96	11.03 ± 2.38	2.87

 ΔE^* : color difference between the color of wax apples at the beginning and the end of storage.

the use of pesticides, efforts are devoted to reducing postharvest losses of horticultural crops by using chemical and physical methods such as ozone treatments, heat treatments, and storage technologies. Postharvest heat treatments such as hot water treatment and hot air treatment reduce rot development, enhance fruit resistance to chilling injury in cold-sensitive cultivars, and retain fruit quality during cold storage and shelf life. However, the complete control of decay is rarely accomplished by heat therapy alone, especially when fruits are subjected to cold storage prior to marketing (Schirra et al., 2011). Ozone is a strong oxidizer that is lethal to microorganisms at low concentrations. Ozonated water was highly effective in killing both gram-negative and gram-positive food associated bacteria (Restaino et al., 1995). In the aqueous form, the antimicrobial efficacy of ozone as a postharvest treatment has been exhibited on a wide variety of fruit and vegetable commodities including iceberg lettuce (Akbas and Olmez, 2007), red bell peppers, strawberries and watercress (Alexandre et al., 2011) and blueberries (Crowe et al., 2007).

Red color plays a very important role when wax apple fruit is purchased (Pan and Shu, 2007). As more is paid for the red-colored fruits, factors influencing red color during storage are much interested. Anthocyanin is the primary red pigment for the red color in wax apple fruit (Supapvanich et al., 2011). Degradation of anthocyanins in the presence of ozone could be due either to direct reaction with ozone or indirect reaction because of secondary oxidants (Tiwaria et al., 2009). According to the Criegee mechanism (Criegee, 1975) where ozone molecules undergo 1,3-dipolar cycloaddition with double bonds present, leading to the formation of ozonides (1,2,4-trioxolanes) from alkenes and ozone with aldehyde or ketone oxides as decisive intermediates, all of which have finite lifetimes (Cullen et al., 2009). This leads to the oxidative disintegration of the ozonide and formation of carbonyl compounds, while oxidative workup leads to carboxylic acids or ketones. Significant reduction in anthocyanin content (98.2%) was observed at an ozone concentration of 7.8% w/w and a treatment time of 10 minutes (Tiwaria *et al.*, 2009). Results from Table 1 showed that the 'a' value of treated wax apples increased on day 7 and then decreased slightly on day 14. Therefore, ozonated water treatment is not able to cause serious degradation of anthocyanin in wax apple fruit individuals treated with relatively low concentration of ozone water. Similar results were found on the surface color of blackberries that was better retained in 0.1– and 0.3–ppm–stored berries by five days and in 0.3 ppm berries by 12 days (Barth *et al.*, 1995).

The principal spoilage mechanisms affecting the quality of chilled fresh produce are enzymatic discoloration, microbial growth and moisture loss. Good manufacturing and handling practices along with the appropriate use of MAP are very effective on inhibiting these spoilage mechanisms, thereby extending the shelf life of prepared produce items (Day, 1992). Properly chosen composition of the gas mixture in the storage environment prolongs storage time by reducing development of microorganisms, and ensuring their microbiological safety. Juhneviča et al. (2011) mentioned that a modified atmosphere reduced the development of bacteria, yeasts and molds on the surface of apple stored at 2°C. The total amount of bacteria was reduced by 50% in the used modified atmosphere, in comparison to samples stored in a cooler. The experimental finding that low O₂MAP is capable of decreasing decay of wax apple under storage can be explained by the inhibition of aerobic microbial growth. Storage in high CO₂ and/or low O₂ results in reduced loss of chlorophyll as well as reduced accumulation of other pigments including anthocyanin, lycopene, xanthophylls and carotenoids (Barth et al., 1993; Barth and Hong, 1996). Results showed that the CIE L* a* b* values of MAP wax apple were not changed a lot during storage. It may due to the low content of chlorophyll and high content of anthocyanin in the surface of wax apple so that small amounts of the reduction of chlorophyll

loss and anthocyanin accumulation are not able to lead to a considerable change of color during storage.

Conclusion

Ozonated water treatment and modified atmosphere are both effective on the extension the storage limit of wax apple at 12°C. The combination of ozonated water treatment and MAP storage not only decreased the decay of fruits, but also maintained the color of the skin of fruit in good quality. Thus, the combination of ozonated water treatment and MAP provided an opportunity of extending the storage limit of wax apple. More research will be conducted to improve the storage limit of wax apple at 12°C including active MAP by the use of gas absorbers, such as oxygen, carbon dioxide or ethylene absorbers.

AUTHOR CONTRIBUTIONS

Conceptualization: JJS. Data curation: JJS. Formal analysis: JJS. Funding acquisition: JJS. Investigation: JJS. Methodology: JJS TCH AW. Project administration: JJS. Resources: JJS. Software: JJS. Supervision: JJS TCH AW. Validation: JJS. Visualization: JJS. Writing – original draft: JJS. Writing – review & editing: JJS TCH AW.

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