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Synergistic effect of calcium chloride and 1-Methylcyclopropene on storage of watermelon (*Citrullus lanatus* (Thunb.) Matsum. & Nakai)

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ABSTRACT

Watermelon (*Citrullus lanatus* (Thunb.) Matsum. & Nakai) was a climacteric variety with a high respiration rate and ethylene accumulation. Therefore the fruit matures and softeness quickly during post-harvest period. Calcium chloride was popularly utilized as stabilizing agent. 1-methylcyclopropene (1-MCP) has been known to be highly effective inhibitor of ethylene reaction. This research evaluated the synergistic effect of CaCl₂ and 1-methylcyclopropene (1-MCP) treatment to weight loss, firmness, total soluble solid, carotenoid, ascorbic acid and decay rate of watermelon during storage. Results showed that a combination of 2.5% CaCl₂ and 0.6 ppm 1-MCP in 20 min of immersion could extend watermelon shelf life for 15 days. After 15 days of ambient storage, the weight loss (1.43 ± 0.02 %), firmness (4.38 ± 0.00 N), total soluble solid (13.60 ± 0.01 °Brix), carotenoid (16.31 ± 0.02 µg/100g), ascorbic acid (13.36 ± 0.03 mg/100g), decay rate (0.47 ± 0.02 %) were clearly presented. Meanwhile, the treatment of 2.5% CaCl₂ alone showed the weight loss (2.11 ± 0.02 %), firmness (3.03 ± 0.02 N), total soluble solid (12.97 ± 0.03 µg/100g), ascorbic acid (9.57 ± 0.02 mg/100g), decay rate (2.14 ± 0.01 %). The incorporation of CaCl₂ and 1-MCP created a synergistic effect on the improved quality of watermelon fruit during ambient storage.

Introduction

Calcium chloride was widely known as stabilizing agent to reinforce cellular membrane, cell turgor, membrane integrity, tissue firmness; slow down senescence; retarded membrane lipid catabolism; prolonged shelf life of fruits and vegetables (1). Post harvest treatment by CaCl₂ dipping or spraying was proven without detrimental impact on overall acceptance (2). 1-methylcyclopropene (1-MCP) has been proven to be greatly beneficial inhibitor of ethylene reaction (3). 1-MCP highly reactive at trace concentration without toxic residue (4). It retarded fruit senescence, pigment degradation, respiration rate, but preserved texture hardness and quality properties, prevented fruit weight loss, prolonged shelf life of different fruits (5). Effectiveness of 1-MCP to fruit shelf life based on numerous parameters such as cultivar, maturity, ratio, temperature, duration of exposure, technique and storage (6).

Watermelon (Citrullus lanatus (Thunb.) Matsum. & Nakai) belongs to the family Cucurbitaceae. Its pulp contained high moisture content, and a rich source of phytochemical components such as lycopene, βcarotene, ascorbic acid (7). Lycopene and β -carotene natural were lipophilic carotenoids having antioxidant properties against free radicals with beneficial effects on human health. Studies are there on coated watermelons with alginate: chitosan and preserved under ambient temperature for 15 days (8). It was found that watermelon coated by alginate: chitosan (1.0: 1.0%) revealed the smallest change in weight loss, highest retention of firmness and increased total soluble solids and beta-carotene contents. By this coating, watermelon could be extended to 15 days. Objective of the study examined the synergistic effect of CaCl₂ and 1-MCP treatment to weight loss, firmness, total soluble solid, carotenoid, ascorbic acid and decay rate of watermelon during storage.

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Materials and Methods

Material

Watermelon fruits were synchronous in maturity and free from defects. They were harvested in Soc Trang province, Vietnam. Chemical reagents were of analytical grade.

Researching method

Watermelon fruits were randomly separated into 3 groups:

Group I: Fruits were immersed in $CaCl_2$ in different concentrations (1.0÷3.0%) in different durations (5÷25 min). Fruits were monitored for firmness and weight loss after 3 days at ambient storage.

Group II: Fruits were immersed in 2.5% CaCl₂ + 1-methylcyclopropene (0÷0.8 ppm) for 20 min.

Chemical reagents were all of analytical grade. Fruits were monitored for firmness and weight loss for 15 days in 3 day-interval at ambient storage.

Group III: Fruits were immersed in 2.5% CaCl₂ and 0.6 ppm 1-methylcyclopropene for 20 min and then stored for 15 days of ambient storage. In 3 day-interval, fruits were sampled to measure total soluble solid, carotenoid, vitamin C and decay rate.

Physicochemical determination

Firmness (N) was estimated by penetrometer. Weight loss (%) was evaluated by comparison of weight before and after treatment. Total soluble solid (°Brix) was measured by hand-held refractometer. Carotenoid (μ g/100 gm) was quantified by highperformance liquid chromatography following the standard methodology (9). Vitamin C (mg/100 gm) was determined by using a 2,6-dichlorophenol indophenol visual titration method. Decay rate (%) was counted by number of rotten fruits per total number of tested fruits.

Statistical analysis

The experiments were run in triplicate with different groups of samples. The data were presented as mean±standard deviation. Statistical analysis was performed by the Statgraphics Centurion version XVI.

Results and Discussion

Effect of $CaCl_2$ concentration and immersion time on firmness and weight loss of watermelon after 3 days at ambient storage

Watermelon fruits (without treatment) after 3 days at ambient storage had texture firmness (2.28 ± 0.03 N) and weight loss (3.51 ± 0.02 %). Weight loss and firmness were significantly improved by CaCl₂ treatments during 3 days at ambient storage (Table 1 and 2). It's obviously noticed that the watermelon fruits treated by 2.5% CaCl₂ in 20 min and stored at ambient temperature for 3 days had higher firmness (4.62 ± 0.00 N) and lower weight loss (1.62 ± 0.03 %) compared to control. Weight loss in watermelon fruits was mainly caused by respiration and transpiration induced by a vapour pressure deficit between the internal tissue and the environment.

 Table 1. Firmness (N) of watermelon after 3 days at ambient storage

CaCl ₂ (%)	Immersion time (min)						
	5	10	15	20	25		
1.0	3.76 ± 0.01^{d}	$3.89{\pm}0.03^{\text{cd}}$	$3.95{\pm}0.02^{\text{cd}}$	$4.08{\pm}0.01^{\rm bc}$	$4.09\pm0.00^{\mathrm{bc}}$		
1.5	$3.92{\pm}0.03^{cd}$	$4.02\pm0.02^{\circ}$	$4.13{\pm}0.00^{\text{bc}}$	$4.27\pm0.03^{\text{b}}$	4.30±0.01 ^b		
2.0	4.01±0.00 ^c	$4.18{\pm}0.01^{\rm bc}$	$4.26{\pm}0.03^{\rm b}$	4.39 ± 0.02^{b}	4.40±0.03 ^{ab}		
2.5	$4.19{\pm}0.02^{\rm bc}$	$4.33\pm0.00^{\mathrm{b}}$	4.49 ± 0.01^{ab}	4.62 ± 0.00^{a}	4.65±0.02ª		
3.0	4.21±0.01 ^{bc}	4.35±0.02 ^b	4.52 ± 0.00^{ab}	4.64±0.03 ^a	4.69±0.01ª		

Note: the values were expressed as the mean of twenty two samples; the same characters (denoted above), the difference between them was not significant ($\alpha = 5\%$).

Table 2. Weight loss (%) of watermelon after 3 days at ambient storage

CaCl ₂ (%)	Immersion time (min)							
	5	10	15	20	25			
1.0	2.45 ± 0.02^{a}	2.23 ± 0.01^{ab}	$2.14{\pm}0.00^{\rm b}$	$2.03{\pm}0.01^{\rm bc}$	2.01 ± 0.02^{bc}			
1.5	2.21 ± 0.01^{ab}	$2.13\pm0.00^{\text{b}}$	$2.01{\pm}0.03^{\rm bc}$	1.90±0.02°	1.88±0.03 ^c			
2.0	$2.12 \pm 0.03^{\text{b}}$	$2.00{\pm}0.02^{\rm bc}$	1.85±0.01°	$1.77{\pm}0.00^{cd}$	1.75±0.01 ^{cd}			
2.5	2.00 ± 0.00^{bc}	1.81±0.01 ^c	1.73±0.02 ^{cd}	1.62 ± 0.03^{d}	1.60 ± 0.02^{d}			
3.0	$1.97{\pm}0.02^{\rm bc}$	1.78±0.03 ^{cd}	1.72±0.00 ^{cd}	1.60 ± 0.01^{d}	1.58.±0.00 ^d			

Note: the values were expressed as the mean of twenty two samples; the same characters (denoted above), the difference between them was not significant ($\alpha = 5\%$).

Their firmness became soft from crisp. Their taste decreased, and resistant capacity against physical and microbial pathogen also came down. However, high concentration of CaCl₂ could create a bitter aftertaste. In fruit softening, protopectin was hydrolyzed into soluble pectin, thus the physical structures of pectin, cellulose and hemicellulose were depolymerized and the fruit turned loose. Calcium prevented the action of cell-wall degrading enzymes. It also created an antagonistic effect on ethyleneinduced ripening of fruit. The retention of firmness in calcium treated fruit might be due its accumulation in the cell walls leading to facilitation in the cross linking of the pectic polymers which accelerated wall strength and cell cohesion (2). It was revealed that apple treated by CaCl₂ decreased softening and kept firmness during storage (10). It was also noticed that the apple fruit dipped in CaCl₂ increased firmness percentage (11). There are studies revealing pear fruit treated with CaCl₂ had minor weight loss compared to non treated fruit (12). CaCl₂ decreased the respiration rate and ethylene accumulation of netted melon (13). It was reported that CaCl₂ significantly prevented the alteration of cell wall to preserve the fruit firmness of apple (14). Post-harvest CaCl₂ treatments inhibited fruit softening and reduced weight loss in apple (15). In a study it was proved that CaCl₂ enhance grapefruit texture (16). There were investigation on 1methylcyclopropene on Rosa sterilis D. shi fruits (17). They showed that postharvest treatment with 1.0 μ L/ L 1-MCP suppressed the respiration rate, ethylene production rate and peroxidase activity in Rosa sterilis D. shi fruits, inhibited the transport of reducing sugar and the increase of cellulose, delayed the increase in polyphenol oxidase activity, and retarded the decrease in ascorbic acid. Studies also proved that 2.5% $CaCl_2$ solution immersion for 15 min significantly delayed the ripening of papaya fruit (18). Synergistic effect of $CaCl_2$ and 1methylcyclopropene concentration on firmness and weight loss of watermelon during 15 days of ambient storage.

Watermelon fruits (only treated by 2.5% CaCl₂) after 3 days at ambient storage were noticed at firmness (4.62 ± 0.00 N) and weight loss (1.62 ± 0.03 %). Weight loss and firmness were significantly affected by CaCl₂ combined 1-MCP treatments during 15 days at ambient storage (Table 3, 4). It's obviously noticed that firmness decreased and weight loss increased

Table 3. Firmness (N) of watermelon during 15 days at ambient storage by incorporation of 1-methylcyclopropene (ppm) into 2.5% CaCl₂ in 20 min of immersion

2.5% CaCl ₂ + 1-methylcyclopropene (ppm) in 20 min of immersion							
0	0.2	0.4	0.6	0.8			
4.62±0.00°	4.95 ± 0.03^{b}	5.16 ± 0.02^{ab}	5.29±0.01 ^a	5.30±0.03ª			
4.43±0.01°	$4.79\pm0.00^{\mathrm{b}}$	$4.97{\pm}0.01^{\text{ab}}$	5.08 ± 0.03^{a}	5.10±0.00 ^a			
4.17±0.03°	$4.58\pm0.01^{\text{b}}$	$4.79{\pm}0.03^{\text{ab}}$	$4.94{\pm}0.01^{a}$	4.96±0.02ª			
4.01±0.00°	4.35 ± 0.02^{b}	4.50 ± 0.00^{ab}	4.66±0.02 ^a	4.69±0.01ª			
3.03±0.02°	$4.09\pm0.03^{\text{b}}$	4.22 ± 0.01^{ab}	4.38 ± 0.00^{a}	4.40±0.03 ^a			
	0 4.62±0.00 ^c 4.43±0.01 ^c 4.17±0.03 ^c 4.01±0.00 ^c	0 0.2 4.62±0.00 ^c 4.95±0.03 ^b 4.43±0.01 ^c 4.79±0.00 ^b 4.17±0.03 ^c 4.58±0.01 ^b 4.01±0.00 ^c 4.35±0.02 ^b	0 0.2 0.4 4.62±0.00 ^c 4.95±0.03 ^b 5.16±0.02 ^{ab} 4.43±0.01 ^c 4.79±0.00 ^b 4.97±0.01 ^{ab} 4.17±0.03 ^c 4.58±0.01 ^b 4.79±0.03 ^{ab} 4.01±0.00 ^c 4.35±0.02 ^b 4.50±0.00 ^{ab}	0 0.2 0.4 0.6 4.62±0.00 ^c 4.95±0.03 ^b 5.16±0.02 ^{ab} 5.29±0.01 ^a 4.43±0.01 ^c 4.79±0.00 ^b 4.97±0.01 ^{ab} 5.08±0.03 ^a 4.17±0.03 ^c 4.58±0.01 ^b 4.79±0.03 ^{ab} 4.94±0.01 ^a			

Note: the values were expressed as the mean of twenty two samples; the same characters (denoted above), the difference between them was not significant ($\alpha = 5\%$).

Table 4. Weight loss (%) of watermelon during 15 days at ambientstorage by incorporation of 1-methylcyclopropene (ppm) into 2.5% $CaCl_2$ in 20 min of immersion

Storage (days)	2.5% CaCl ₂ + 1-methylcyclopropene (ppm) in 20 min of immersion								
	0	0.2	0.4	0.6	0.8				
3	1.62 ± 0.03^{a}	$1.15\pm0.00^{\text{b}}$	$1.01{\pm}0.01^{\rm bc}$	$0.84{\pm}0.03^{\circ}$	0.81±0.00°				
6	$1.76\pm0.01^{\mathrm{a}}$	$1.27\pm0.03^{\text{b}}$	$1.13{\pm}0.00^{\rm bc}$	$0.93 \pm 0.02^{\circ}$	0.90±0.03°				
9	1.85 ± 0.00^{a}	1.41 ± 0.01^{b}	$1.22{\pm}0.03^{\rm bc}$	1.04±0.00°	1.02±0.01°				
12	1.97 ± 0.03^{a}	$1.62\pm0.00^{\text{b}}$	$1.50{\pm}0.02^{\rm bc}$	1.32±0.01 ^c	1.30±0.00°				
15	2.11 ± 0.02^{a}	1.73 ± 0.03^{b}	$1.61{\pm}0.01^{\rm bc}$	1.43±0.02 ^c	1.41±0.03°				

Note: the values were expressed as the mean of twenty two samples; the same characters (denoted above), the difference between them was not significant ($\alpha = 5\%$).

during storage. After 15 days of preservation, watermelon fruit treated by 2.5% CaCl₂ combined 0.6% 1-MCP had higher firmness (4.38 ± 0.00 N) and lower weight loss (1.43 ± 0.02 %) compared to samples treated by 2.5% CaCl₂ alone (firmness: 3.03 ± 0.02 N; weight loss: 2.11 ± 0.02 %). Ethylene enhanced the fruit ripening. 1-Methylcyclopropene (1-MCP) was an inhibitor of ethylene performance by competitively capturing to ethylene receptor and blocking ethylene signaling, therefore prevented post-ripening. 1-MCP suppressed the performance of cell wall-lysing enzymes, thus lowering the softening of apple fruit. The retardation of weight loss in 1-MCP treated fruit

might be attributed to slow respiration rate and maintenance of tissue rigidity of the fruit (19). It was demonstrated that 1-MCP suppressed internal ethylene concentration, reduced weight loss, delayed softening and maintained the acidity and total soluble solid in apple (3). It was observed that 1-MCP can decrease the respiratory rate and ethylene release of Yate kiwifruit (20). 1-MCP could suppress the activity of cell wall-lysing enzymes, thereby delaying the sofening of apple fruits. In a treatment chloride with calcium (CaCl₂) and 1methylcyclopropene (1-MCP) (21). They found that the respiration rate, ethylene formation, the activity and gene expression of pectinases were strongly decreased by 0.18 mol/L CaCl₂ and/or 1µl/L 1-MCP.

Quality properties of watermelon under synergistic effect of 2.5% CaCl₂ and 0.6 ppm 1-methylcyclopropene during 15 days of ambient storage

Total soluble solid (°Brix), carotenoid (μ g/100 gm), vitamin C (mg/100 gm) of watermelon treated by 2.5% CaCl₂ and 0.6 ppm 1-MCP slightly decreased during 15 days at ambient storage. Meanwhile decay rate slightly increased (Table 5). The degradation of total soluble solid in fruit during storage was due to transpiration and the substrate metabolism of respiration (21).

Fruit decay was primarily originated by the infection of decay-causing pathogens. Fruit softening was highly beneficial to the accumulation and spreading of these pathogens, therefore accelerating the decay rate of fruit (21). CaCl₂ and 1-MCP effectively retarded the growth and propagation of pathogens by delaying the fruit softening thus limiting the decay rate and extending the fruit stability. Researchers examined the influence of 1methylcyclopropene and calcium salt to quality of fresh-cut muskmelon stored in ambient condition for 18 days (22). They confirmed that Ca-chelate synergistically enhanced the effect of 1-MCP on firmness after nine days of storage, while calcium chloride improved firmness. It was showed that the incorporation of 1-MCP and CaCl₂ significantly reduced polygalacturonase and polyphenoloxidase activities on jujube fruit (23). The ripening of mango was delayed by 1-MCP and shelf life of the fruits was extended with increase by the concentration of 1-MCP (24). Meanwhile the physicochemical properties such as weight loss, total soluble solid and colour were slightly accelerated and ascorbic acid was strongly decreased. It was proved that softer melon flesh was more conducive to the growth and reproduction of decay-causing microorganisms (21). The utilization of 1-MCP at 100 ppm prolonged the physicochemical and phytochemical quality of mabolo fruit, including weight loss, firmness, ascorbic acid, total soluble solid, total phenolic, flavonoid during 8 days of preservation (25).

Conclusion

Watermelon contained high water content and thin dark green rind. Its shelf-life was relatively short under ambient storage. Calcium chloride created **Table 5.** Quality properties of watermelon under synergistic effect of 2.5% $CaCl_2$ and 0.6 ppm 1-methylcyclopropene at 20 min ofimmersion during 15 days of ambient storage

Storage (days)	Total soluble solid (°Brix)		Carotenoid (µg/100g)		Vitamin C (mg/100g)		Decay rate (%)	
	2.5% CaCl ₂	2.5% CaCl ₂ + 0.6 ppm 1- MCP	2.5% CaCl ₂	2.5% CaCl ₂ + 0.6 ppm 1-MCP	2.5% CaCl ₂	2.5% CaCl ₂ + 0.6 ppm 1-MCP	2.5% CaCl ₂	2.5% CaCl ₂ + 0.6 ppm 1-MCP
3	$14.12 \pm 0.02^{\rm b}$	14.25 ±0.01 ^a	16.83 ±0.03 ^B	16.83 ±0.00 ^A	13.79 ± 0.01^{N}	14.23 ±0.02 ^M	1.65 ±0.00 ^x	0.17 ±0.01 ^Y
6	$13.65 \pm 0.00^{\rm b}$	14.08 ± 0.03^{a}	15.10 ±0.01 ^B	16.62 ±0.02 ^A	12.25 ± 0.00^{N}	13.88 ±0.03 ^M	1.79 ± 0.03^{x}	0.24 ±0.02 ^y
9	12.72 ± 0.03^{b}	13.91 ±0.02 ^a	14.39 ±0.00 ^B	16.57 ±0.01 ^A	11.16 ±0.03 ^N	13.60 ±0.01 ^M	1.92 ± 0.02^{x}	0.30 ±0.00 ^Y
12	$11.21 \pm 0.01^{\rm b}$	13.77 ±0.00ª	13.74 ±0.02 ^B	16.48 ±0.00 ^A	10.29 ±0.01 ^N	13.47 ±0.00 ^M	2.05 ±0.00 ^x	0.39 ±0.01 ^Y
15	$10.83 \pm 0.02^{\rm b}$	13.60 ±0.01 ^a	12.97 ±0.03 ^B	$16.31 \pm 0.02^{\rm A}$	9.57 ± 0.02^{N}	13.36 ±0.03 ^M	2.14 ± 0.01^{x}	0.47 ±0.02 ^y

Note: the values were expressed as the mean of twenty two samples; the same characters (denoted above), the difference between them was not significant ($\alpha = 5\%$).

firming effect on watermelon, stabilized membrane, formed calcium-pectate complex rendering rigidity to the middle lamella and cell wall. 1-MCP prolonged postharvest stability and quality of watermelon by inhibiting ethylene action. CaCl₂ combined 1-MCP had a great possibility to extend the shelf life and improve post harvest quality of watermelon fruit. The synergistic effect created from CaCl₂ and 1-MCP could retard lignin and cellulose accumulation, and delay senescence.

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Authors' contributions

Nguyen Phuoc Minh was the single author of this article.

Conflict of interests

The authors declared that the present study was performed in the absence of any conflict of interest.

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