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EFFECTS OF VACUUM INFILTRATION, DIPPING IN CALCIUM CHLORIDE AND POSTHARVEST HEAT TREATMENT ON FIRMNESS AND CALCIUM CONTENT OF APPLES

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ABSTRACT

Apples (*Malus domestica* Borkh. 'Red Delicious') were vacuum infiltrated (250 mm Hg) for 30 s with 0, 1.5, 3 and 6% solutions of calcium chloride ($CaCl_2$) and stored at 0°C for 6 mo, followed by 1 wk storage at 20°C. Calcium chloride solutions at 3 and 6% concentrations significantly increased fruit firmness tested either immediately after removal from storage or after being held for 1 wk at 20°C. The correlation coefficient between calcium (Ca) content and fruit firmness (r=75) was highly significant. In another experiment, 'Red Delicious' apples were treated by dip infiltration in 0 and 3% $CaCl_2$ solution, for 3 min, heat treatment for 4 d at 38°C and dip infiltration (3% $CaCl_2$) plus heat treatment, before storing at 0°C for 6 mo. Heat treatment and the combination of dip infiltration plus heat treatment significantly increased fruit firmness either immediately after removal from storage or after being held for 1 wk at 20°C.

Key words: Calcium chloride, Heat treatment, 'Red Delicious' apples, Vacuum infiltration.

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اثرهای نفوذ در خلاء، غوطه وری در کلریدکلسیم و تیمار گرمایی بر سفتی و میزان کلسیم سیب

مجید راحمی و ابوالقاسم بحرانی

به ترتیب، دانشیار و دانشجوی پیشین کارشناسی ارشد بخش علوم باغبانی دانشکده کشاورزی دانشگاه شیراز، شیراز، جمهوری اسلامی ایران.

چکیده

سیب رقم 'رد دلیشس' برای ۳۰ ثانیه در محلول کلرید کلسیم صفر، ۱/۵، ۳ و ۶ ٪ نفوذ در خلاء (۲۵۰ میلی متر جیوه) شده و در انبار سرد صفر درجه سانتی گراد برای مدت ۶ ماه و در پی آن یک هفته در دمای ۲۰ درجه سانتی گراد نگهداری شد. محلول کلرید کلسیم ۳٪ و ۶٪ به طورمعنی داری سفتی گوشت میوه را بی درنگ پس از خروج از سردخانه و پس از یک هفته نگهداری در دمای ۲۰ درجه سانتی گراد، افزایش داد. ضریب همبستگی بین میزان کلسیم و سفتی گوشت میوه (r =۰/۷۵) در سطح ۰/۱٪ معنی دار بود در آزمایشی دیگر، سیب رقم 'رد دلیشس' در محلول کلرید کلسیم صفر و ۳٪ برای ۳ دقیقه به روش غوطه ور کردن، گرما دهی برای ۴ روز در ۳۸ درجه سانتی گراد و غوطه ور کردن در محلول کلرید کلسیم ۳٪ همراه با گرما دهی، پیش از نگهداری در سردخانه صفر درجه سانتی گراد، تیمار شدند. تیمار گرما دهی و تیمار کلرید کلسیم به طور معنی داری باعث سفتی گوشت میوه بی درنگ پس از خروج از انبار یا قرار گرفتن به مدت یک هفته در دمای ۲۰ درجه سانتی گراد شد.

INTRODUCTION

'Red Delicious' apples soften at a faster rate than 'Golden Delicious' during prolonged storage in Iran. There are methods for preventing or reducing this phenomenon. Several investigators have studied the effects of $CaCl_2$ sprays and dips on Ca concentration in apples and on delay of softening during prolonged storage (2, 11, 14, 15, 16).

Studies on infiltration of Ca by vacuum suggest that migration of Ca solution into the intercellular spaces of a submerged apple could be induced by establishing a positive atmospheric pressure differential between the ambient and the intercellular spaces of the fruit cortex (3, 22). Methods of positive pressure and partial vacuum for infiltrating fruit with Ca have proven to be highly effective in controlling fruit firmness, bitter pit and senescence (18).

Pre-storage heat treatments have been used for controlling fungal disease and insect infestation to delay softening of apple fruits. Porritt and Lidster (19) exposed 'Spartan' and 'Golden Delicious' apples to 38°C for 4 to 6 d and stored them at -1°C for 4 to 7 mo. Fruit softening was suppressed and naturally occuring decay was reduced. Klein *et al.* (9) heated 'Anna' or 'Granny Smith' fruit at 38°C for 4 d and dipped the fruit in 2% $CaCl_2$ at either 20°C or 38°C, or heated fruit at 38°C for 4 d after dipping them in 2% $CaCl_2$ at 20°C. The best heat treatment was exposure to 38°C for 4 d; a $CaCl_2$ dip before heating appreciably enhanced the effect of heating. Lurie *et al.* (12) showed that following a pre-storage exposure of apples to 38°C no damage was found on preclimacteric apples and the fruit firmness was maintained during post storage ripening.

This report describes the effects of pre-storage heat treatment and vacuum infiltration of $CaCl_2$ on softening, and shelf life of 'Red Delicious' apples.

MATERIALS AND METHODS

Experiment 1

'Red Delicious' apples (*Malus domestica* Borkh.) were harvested at physiological maturity from a commercial orchard near Sepidan, Fars province. The fruits were transferred to the postharvest laboratory of the

Department of Horticultural Science, College of Agriculture, Shiraz University, Shiraz, Iran. The apples were vacuum infiltrated with 0, 1.5, 3 and 6% solutions of calcium chloride ($CaCl_2$, 98%). Starch (1%) and corn flour (2.5%) were added to $CaCl_2$ solution. Vacuum infiltration was performed by placing the fruits in $CaCl_2$ solution for 30 s under 250 mm Hg in a vacuum oven. Then, the fruits were allowed to drain and air dry for 30 min. Treated fruits were divided into random samples of 20 fruits and kept in perforated polyethylene bags each with 24 holes of 5 mm diameter and stored at 0°C under a RH of 85-90% for 6 mo, followed by 1 wk storage at 20°C. Samples of treated fruits from storage were evaluated at monthly intervals.

Fruit firmness was measured in 4 replications of 20 fruits per treatment, 10 fruits immediately after removal from storage and 10 fruit at the end of 7 d at 20°C with an Effegi pressure tester using a 11 mm tip which penetrated to a depth of 8.0 mm.

Calcium content of the apple tissue was determined by removing the peel and samples of flesh tissue on both side of fruit were then removed with a knife. The flesh tissue was dried at 90°C for 7 d. Tissue samples, 0.5±0.0005 g, were ashed at 450°C overnight and the residue was dissolved in 5 ml of 2N HCl. The samples were then diluted and analyzed for Ca content with an atomic absorption spectrophotometer (Shimadzu AA670). All Ca values were reported as mg kg^{-1} on a dry weight basis. The factorial experiments were arranged in a completely randomized design with four replications.

Experiment 2

Fruits were harvested as described previously. To test the combination of $CaCl_2$ and heating, a completely randomized design with three replications of each of the following treatments was used: 1) no treatment (control), 2) dipped for 3 min in 3% $CaCl_2$ solution, 3) heated 4 d at 38° C, and 4) dipped for 3 min in 3% $CaCl_2$ solution plus 4 d at 38°C. The calcium content and firmness were measured as in the previous experiment. Samples of treated fruits from storage were evaluated at 2-month intervals. The data were analyzed using MSTATC software and the means compared using Duncan's Multiple Range Test (DMRT).

RESULTS AND DISCUSSION

Experiment 1

Firmness. Fruit firmness of 'Red Delicious' vacuum infiltrated with $CaCl_2$ was influenced by $CaCl_2$ solution concentrations (Table 1). Treatment with $CaCl_2$ at 3 and 6% concentration progressively increased flesh firmness of the apple fruits. Treated fruits after 6 mo storage at 0°C followed by holding at 20°C for 7 d, were significantly firmer than untreated fruits (Table 2). There was a positive linear relationship between $CaCl_2$ concentration and flesh firmness, and flesh firmness was correlated positively with Ca concentration (Fig. 1). The effect of $CaCl_2$ on firmness was consistent with effects previously found with apples (1, 4, 5, 6, 20).

Table 1. Effects of vacuum infiltration of $CaCl_2$ on flesh firmness (kg) of 'Red Delicious' apples after removal from storage at 0°C.

$CaCl_2$ solution (%)	Storage period (mo)						Mean
	1	2	3	4	5	6	
0.0	8.01 b†	6.78 c	6.71 bc	6.32 b	6.18 bc	6.05 c	6.69 C
1.5	7.70 b	7.45 b	6.45 c	6.19 b	5.96 c	5.87 c	6.61 C
3.0	8.83 a	7.69 ab	6.92 b	6.77 a	6.40 ab	6.47 b	7.18 B
6.0	8.13 b	7.91 a	7.51 a	7.01 a	6.46 a	6.87 a	7.32 A
Mean	8.17 A	7.48 B	6.90 C	6.57 D	6.25 E	6.31 E	

† Mean separation within columns and rows by DMRT, at 5% level.

Table 2. Effects of vacuum infiltration of $CaCl_2$ on flesh firmness (kg) of 'Red Delicious' apples after removel from storage plus 7 d at 20°C

$CaCl_2$ solution (%)	Storage period (mo)						Mean
	1	2	3	4	5	6	
0.0	6.85 b†	6.41 c	5.97 c	6.9 bc	5.71 bc	5.57 bc	6.10 C
1.5	6.85 b	6.38 c	5.46 d	5.77 c	5.44 c	5.35 c	5.88 D
3.0	7.56 a	6.81 b	6.22 b	6.38 ab	5.88 b	5.94 ab	6.47 B
6.0	7.53 a	7.30 a	7.13 a	6.71 a	6.68 a	6.39 a	6.96 A
Mean	7.20 A	6.73 B	6.20 C	6.24 D	5.93 D	5.81 D	

† Mean separation within columns and rows by DMRT, at 5% level.

Free carboxyl groups on polygalacturonate polymers play an important role in stabilizing and maintaining wall integrity. Calcium application may prevent dessication and Ca binding may strengthen tissue and make it more resistant to hydrolytic enzyme attack, or the presence of Ca may decrease hydrolytic enzyme activity.

Calcium content. Uptake of Ca by apples from vacuum infiltration with $CaCl_2$ solutions increased as $CaCl_2$ concentration increased up to 6%. $CaCl_2$ at 6% significantly increased flesh Ca level by 469.8 mg kg^{-1} after 6 mo of storage (Table 3). There was a linear relationship between $CaCl_2$ concentration and flesh Ca level, and the correlation coefficient (r=0.51) was significant at 5% level (Fig. 2). A positive correlation also existed between flesh Ca level and fruit firmness (Fig. 3). The results of this study were in agreement with previous reports (3, 4, 8, 10). Glenn *et al.* (7) investigated the pathways of Ca penetration into apple fruit and reported that lenticels had a significant positive effect on Ca penetration. They found a dense network of structures resembling cracks that seemed to provide pathways for cuticular Ca penetration. Lidster *et al.* (11) reported that moderate relative humidity (87%) resulted in the most rapid uptake of Ca by the tissues.

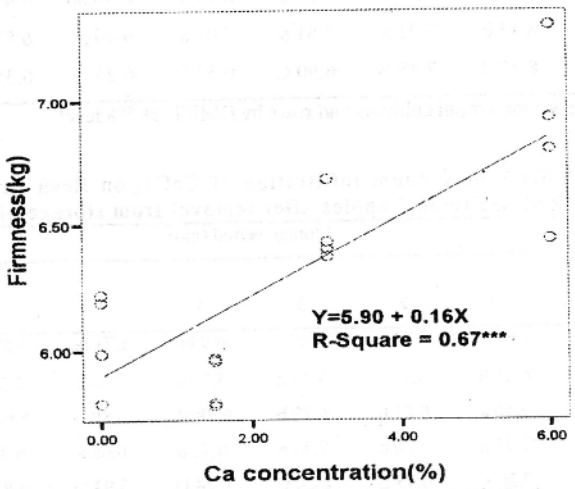

Fig. 1. Relationship between Ca concentration and firmness of 'Red Delicoius' apples after 6 mo removal from storage at 0°C.
*** Significant at 0.0001 level.

Experiment 2

Fruit firmness. 'Red Delicious' apples were treated after harvest by dipping in 3% $CaCl_2$ solution, heated for 4 d at 38°C or the two treatments combined, before stored at 0°C. Dipping apples in 3% $CaCl_2$ alone slightly reduced fruit firmness at removal from storage and at the end of holding for 1 wk at 20°C. Heating apples before storage significantly increased fruit firmness (Tables 2 and 3).

Table 3. Effects of heat treatment and dipping in $CaCl_2$ solutions on fruit firmness of 'Red Delicious' apples during 6 mo of storage at 0°C.

Treatment	Storage period (mo)			Mean
	2	4	6	
Control	6.72 b†	6.71 b	6.34 b	6.59 C
$CaCl_2$ 3%	6.56 b	6.09 c	5.95 c	6.20 D
4 d at 38 °C	7.29 a	6.72 b	6.85 a	6.95 B
$CaCl_2$ 3%+ 4 d at 38° C	7.46 a	7.36 a	6.89 a	7.24 A
Mean	7.01 A	6.72 B	6.51 C	

† Mean separation within columns and rows by DMRT, at 5% level.

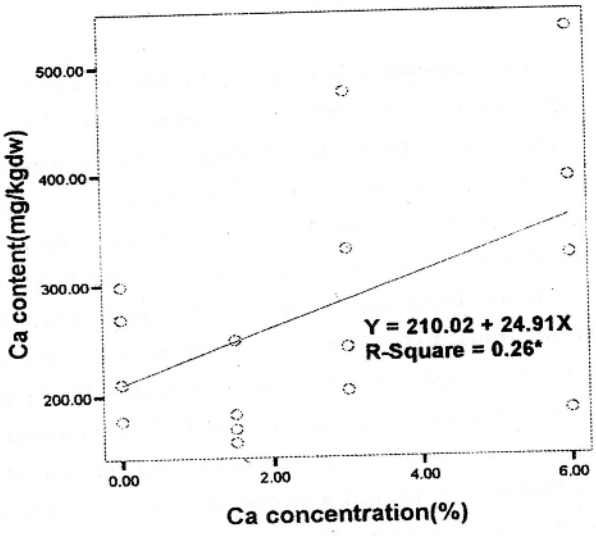

Fig. 2. Relationship between Ca concentration and Ca content of 'Red Delicious' apples after 6 mo removal from storage at 0°C.
* Significant at 0.05 level.

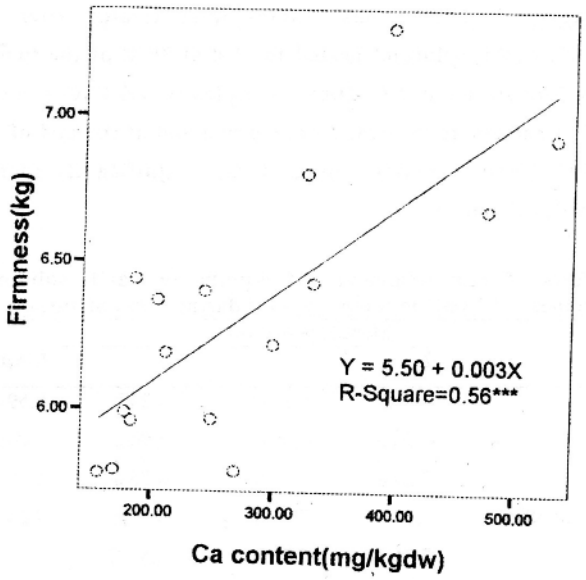

Fig. 3. Relationship between Ca content and firmness of 'Red Delicious' apples after 6 mo removal from storage at 0°C.
*** Significant at 0.001 level.

The combined treatment of heating plus Ca dipping was more effective for retention of flesh firmness than either treatments, alone. Lurie and Klein (13) reported that heat treatment alone maintained fruit firmness of 'Anna' apples, while $CaCl_2$ alone had no effect on fruit quality. The combined treatment maintained fruit firmness best at removal from storage and holding 1 wk at 20°C. The rate of softening of 'Red Delicious' apples increased when heated fruits were returned to 20°C, but it was still less than that of nonheated fruits. Even after 6 mo of storage at 0°C and subsequent storage at 20°C for 7 d, apples that had been held at 38°C for 4 d pre-storage were firmer than nonheated fruits (Tables 3 and 4). The main benefit of heat treatment was reduction in softening. The reasons for the lack of softening may be due to inhibition of the cell wall-degrading enzymes. Klein *et al.* (9) reported that the insoluble pectin fraction remained larger in the heated apples compared with unheated apples during storage. They also examined the activity of softening-related enzymes in other fruits after heat treatment. In avocado, cellulase and polygalacturonase were inhibited by 4 d at 38°.

Lurie and Klein (13) repoted that heated apples contained less soluble pectin and more insoluble pectin than the control. At harvest, 80% of the galacturonic acid was in the insoluble pectin fraction, while after storage followed by holding at 20°C for 7 d, it dropped to 50% in the control apples, 55% in heated apples and 59% in the combined treatment.

Table 4. Effects of heat treatment and dipping in $CaCl_2$ solution on fruit firmnessof 'Red Delicious' apples after removal from storage plus 7 days at 20°C.

Treatment	Storage period (mo)			Mean
	2	4	6	
Control	6.02 ab†	5.95 b	6.11 a	6.03 B
$CaCl_2$ 3%	5.71 b	5.54 c	5.59 b	5.61 C
4 d at 38°C	5.89 b	6.12 b	6.16 a	6.06 B
$CaCl_2$ 3% + 4 d at 38°C	6.30 a	6.67 a	6.27 a	6.42 A
Mean	5.99 A	6.07 A	6.04 A	

† Mean separation within columns and rows by DMRT, at 5% level.

Calcium content. Heating 'Red Delicious' apples for 4 d at 38°C and dipping in 3% $CaCl_2$ solution had no significant effect on Ca content of fruit after 6 mo of storage at 0°C. The concentration of $CaCl_2$ and length of dipping time may not have been sufficient to increase penetration of Ca through the skin and flesh of apple. Wieneke and Benson (24) did not find ^{45}Ca to penetrate into the flesh of mature 'Jonathan' fruit by a postharvest dippig in 1% $CaCl_2$. Mika *et al.* (17) reported that the penetration of Ca from the skin into the flesh was greatest in apples that were warm at the dipping time. Methods of application is also important for penetration of Ca into the flesh of fruit. Scott and Wills (22, 23) reported that vacuum and pressure infiltration of Ca salt solutions was especially effective in getting Ca into the apple.

CONCLUSIONS

This study showed that the rate of softening of 'Red Delicious' apple was related to the Ca status of the flesh. Application of Ca and heat treatment had a great effect on fruit firmness. These methods, can be used in postharvest industry to improve the quality of apples during handling and storage in Iran.

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