Journal of Horticultural Science & Ornamental Plants 4 (2): 155-168, 2012
ISSN 2079-2158

Effect of Pre- and Post-Harvest Calcium and Magnesium Compounds and Their Combination Treatments on "Anna" Apple Fruit Quality and Shelf Life

Karim M. Farag and Neven M.N. Nagy

Department of Horticulture (Pomology), Faculty of Agriculture,
Damanhour University, P.O.Box 22516, Damanhour, Egypt

Abstract: This work aimed at investigating the role of Ca^{2+} and Mg^{2+} compounds or sources on firmness and quality of "Anna" apple fruits whether applied as preharvest or postharvest treatments. Studies on the role of Mg^{2+} on these characteristics are rare. Since calcium is immobile nutrient, spray treatment may be less efficient due to the partial deposition and uptake by apple leaves. During 2009 and 2010 seasons, "Anna" apple trees were sprayed ten days before harvest with Ca^{2+} and, or Mg^{2+} sources in the form of chloride or sulphate in addition to the control. In the postharvest study, collected apples from untreated trees from the same orchard were dipped in $MgCl_2$, $CaCl_2$ each at 1% or 2% (w/v) alone or in combinations, Ethrel at 100 ppm and (control) and then held at room temperature (22±2°C) for 5 days. The preharvest study provided evidence that $CaCl_2$ alone or combined with $MgCl_2$ resulted in the highest firmness of apple fruits followed by $MgSO_4$ plus $CaSO_4$. Moreover, the highest values of TSS were found in fruits treated with $CaSO_4$ alone or combined with $MgSO_4$ when compared with untreated fruits. In addition, $CaCl_2$ alone or plus $MgCl_2$ resulted in higher acidity and vitamin C than other used treatments. The preharvest applications of various Ca^{2+} or Mg^{2+} sources indicated that Ca^{2+} in the chloride form performed better than Ca^{2+} in the sulphate form with regard to electrolyte leakage. Meanwhile, the combined form of $MgCl_2$ plus, $CaCl_2$ resulted in less leakage as compared with just using $MgCl_2$ alone. However, $CaCl_2$ was still superior to its combination with $MgCl_2$ in terms of reducing such leakage. Carotene content data as another quality attribute of “Anna” apples revealed that apples treated with the sulphate form of Mg^{2+} had greater carotene content in the skin as compared with those treated with chloride form. The postharvest treatments of various concentrations of $CaCl_2$ and $MgCl_2$ (1% or 2% w/v), retarded the loss of firmness whether applied individually or in combinations. While Ethrel lead to an adverse effect on flesh firmness in both seasons when compared with the control. Meanwhile, there was no added advantage from increasing the concentration of $CaCl_2$ or $MgCl_2$ from 1% to 2% (w/v) with regard to many fruit characteristics such as TSS, acidity, chlorophylls a and b and anthocyanin and carotenes in both seasons. It could be concluded that calcium in the chloride form was mostly effective over the sulphate form while the later was better with regard to enhancing carotene formation. Moreover, it is preferred to treat “Anna” apples after harvest with $CaCl_2$ plus $MgCl_2$ (1% w/v) and avoid using Ethrel even at low concentration since it accelerates all senescence parameters.

Key words: Apple · Calcium · Magnesium · Coloration · Firmness · Plasma membrane integrity · Cell wall breakdown · Tissues senescence

INTRODUCTION

Apple (*Malus domestica*.L) is one of the most important crops widely grown in the world. Moreover, the consumers acceptance of apples has been increasing especially with increasing the public health awareness which emphasizes the need for better quality fruits with prolonged shelf-life. Firmness is one of the most important attributes of apple quality that tends to be greatly influenced by many preharvest and postharvest factors. Apple consumers demand a crispy flesh in addition to the special qualities of each cultivar. Obtaining and maintaining apple fruit firmness from orchard to the consumers, tends to be one of the major issues facing

Corresponding Author: Karim M. Farag, Department of Horticulture (Pomology), Faculty of Agriculture, Damanhour University, P.O.Box 22516, Damanhour, Egypt. E-mail: karimmfarag@hotmail.com.

apple producers [1]. Preharvest foliar treatment of Golden Delicious, Delicious and Cox's Orange Pippin apples with calcium have been shown to be firmer than the respective non-treated apples [2, 3]. Postharvest Ca^{2+} dip increases fruit Ca^{2+} content and reduces firmness loss for many apple cultivars [4, 5]. The source of Ca^{2+} may also influence its efficiency preserving fruit firmness [6]. Increasing the Ca^{2+} content of apples maintains fruit firmness, decreases the incidence of disorders such as water core, bitter pit and internal breakdown [7]. Postharvest Ca^{2+} infiltration represents a safe and potentially effective method for increasing the content of fruit tissue and storage life of apples [8]. Calcium was found to increase cell membrane integrity by binding to the polar head groups of phospholipids. Obviously, many studies have been focusing on the role of calcium in maintaining fruit apple quality since it binds to the cell wall polymers. The available information, however, about the role of Mg^{2+} in obtaining and maintaining quality of apple fruits is rare. Although, Mg^{2+} is bi-valent cation as Ca^{2+} and possible for this element that can be linkaged between pectic substances within the cell wall or linkaged between the polar heads in the plasma membrane. Thus, magnesium may maintain cell wall and plasma membrane integrity. Meanwhile, calcium is important but immobile nutrient thus spray treatment of calcium solutions in the field before harvest may be less efficient than expected from direct dipping of apple fruit after harvest. The reason behind this conclusion could be ascribed to losing some of the sprayed calcium that deposited on the apple leaves. Thus, the aims of this research were to study the effect of Ca^{2+} and Mg^{2+} compounds alone or in combinations, when applied as pre or postharvest treatments, also, the study investigated the effect of the type of salts of these elements when applied as sulphate or chloride as preharvest treatments on maintaining fruit firmness at harvest or during the shelf-life either compared with untreated fruits or Ethrel treated fruits as a practice conducted on a commercial scale leading to accelerating senescence due to the release of ethylene.

MATERIALS AND METHODS

"Anna" apple trees (*Malus domestica*, Borkh) grown in a private orchard at Tahrir region, Behira governorate, having four years old, grown in a sandy soil, spaced at 4x4m, under drip irrigation system, similar in growth and received common horticulture practices, were selected for this investigation which included two studies, the first one is preharvest treatments by spraying and the second one is postharvest treatments, by dipping fruits in treatment solutions.

The First Study: Preharvest Treatments: Three "Anna" apple trees were selected for each treatment in this part of the study and sprayed with one of the following solutions 10 days before harvest (spraying date was on the mid of June during 2009 and 2010, seasons):- Control, Mg SO_4 (1%), Ca SO_4 (1%), $MgCl_2$ (1%), $CaCl_2$ (1%), Mg SO_4 (1%) plus Ca SO_4 (1%) and $MgCl_2$ (1%) plus $CaCl_2$ (1%).

All treatments solutions were calculated as weight per volume (w/v), while Tween-20 (as a surfactant agent with 0.05% v/v) was added to all these treatments solutions. Then, four apple fruits were collected from each sprayed-tree (replicate) per each treatment and these fruits were free from apparent pathogen infection, uniform in shape and color. Some physical and chemical analysis were carried out on these collected fruits.

The Second Study: Postharvest Treatments: "Anna" apple fruits were harvested (on twenty five of June during both seasons) from other unsprayed - apple trees. The collected fruits were undamaged mature, free from obvious pathogen infection, uniform in shape, weight and color and transported to the laboratory then washed with tap water, dipped for 3 min. in sodium hypochloride (0.05%), for surface sterilization, washed with distillated water, dried in air then immersed for 20 min. in one of the following solutions: Control (water), $CaCl_2$ (1%), $MgCl_2$ (1%), $CaCl_2$ (2%), $MgCl_2$ (2%), $CaCl_2$ (1%) + $MgCl_2$ (1%), $CaCl_2$ (2%) + $MgCl_2$ (2%) and Ethrel (100 ppm) as a commercial ripening agent.

Tween-20 (as a surfactant agent with 0.05%) was added to these solutions and all concentrations of the used solutions were calculated as weight / volume.

The Shelf Life Test: The treated-apple fruits were dried in air, weighed (initial weight), packed in foam dishes with three replicates for each treatment (12 fruits per treatment, with 4 fruits / replicate) and then left at room temperature (22±2°C) for 5 days. Physical and chemical quality assessments were assessed at zero time of shelf life (initial values) and after the end of shelf life (5 days).

Quality Assessments: Fruit weight readings were recorded initially and after 5 days and calculated as a percentage of the initial weight according to Ghoname [9]. Fruit firmness were determined on red and green cheek by using pressure tester and the values of firmness were expressed as Newton units.The percentage of total soluble solids was measured at 20°C using hand refractometer. Titratable acidity was determined as gms malic acid per 100 ml of fruit juice according to the method of Spayed and Morris [10]. Vitamin C was determined as mgs of ascorbic acid per 100 ml fruit juice according to Egan *et al*. [11]. The percentage of electrolyte leakage was calculated according to Gary [12] and Ahrenes and Ingram [13]. Anthocyanin content of apple peel was expressed as mgs / 100 gm fresh weight according to Fuleki and Francis [14]. Carotene and chlorophyll a and b contents of apple peel were extracted, calculated and recorded as mgs / litre according to Wintermans and Mots [15].

Experimental Design and Statistical Analysis: The preharvest treatments was laid out as completely randomized design (CRD) but the second type of experiments which applied as postharvest treatments was laid out as factorial (two factors) design. All the data were subjected to the analysis of variance. The least significant difference to compare the means (LSD) was calculated as outlined by Steel and Torrie [16].

RESULTS AND DISSCUSSUION

The First Study: Preharvest Treatments of "Anna" Apple Fruits

Physical Characteristics

Fruit Weight: Fruit weight data of Anna apple cultivar as influenced by spraying before harvest with magnesium or calcium in the forms of chloride or sulphate alone or used as a combination of both nutrients as compared with control treatment was reported in Table 1. The results revealed that in the first season, $MgSO_4$- treated fruits either alone or in combination with calcium sulphate had similar effect on fruit weight. Moreover, calcium sulphate alone or in combination with $MgSO_4$ did not significantly vary in their influence on fruit weight. In the second season, most used treatments led to similar fruit weight at harvest. Even the control fruits did not significantly vary from all used treatments except Mg^{2+} and the combination of Mg^{2+} and Ca^{2+} in the chloride salt. This latter combination also resulted in lowest fruit weight in both seasons. Moreover, there was no added advantage from using the combination of Mg^{2+} and Ca^{2+} solution as compared with the individual application of each in the form of sulphate salt.

Fruit Firmness (The Red Cheek): Tissue firmness of the red cheek of "Anna" apple fruit was determined at harvest and the data was tabulated and shown in Table 1. The data indicated that calcium chloride treatment alone or in combination with magnesium chloride caused a significant increase of tissue firmness as compared with control in both seasons with significant differences. Moreover, a significant increase in tissue firmness at the red cheek was found with use of calcium in the form of chloride as compared with calcium solution in the form of sulphate in both seasons. Meanwhile, $CaSO_4$ treatment resulted in lower firmness of apple red cheek when compared with other used treatments. Furthermore, the combination of Ca^{2+} and Mg^{2+} in the chloride salt had greater influence on maintaining greater firmness when compared with combining the same nutrients but in the form of sulphate in both seasons.

Table 1: Some physical characteristics of "Anna" apple fruit as influenced by preharvest treatments with calcium, magnesium as chloride or sulphate and their combinations during the two seasons 2009 and 2010

	Physical Characteristics					
	Fruit Weight (g)		Firmness of Red Cheek (Newton)		Firmness of Green Cheek (Newton)	
Treatments	2009	2010	2009	2010	2009	2010
Control	735.00 ab*	741.00 a	32.26 e	31.89 e	33.37 e	35.33 d
$MgSO_4$	750.00 a	753.00 a	40.04 c	46.75 c	48.06 bc	53.50 b
$CaSO_4$	728.30 ab	732.00 a	37.28 d	44.46 d	45.84 d	51.00 c
$MgCl_2$	690.00 ab	696.00 b	38.93 cd	46.06 c	47.40 c	52.75 bc
$CaCl_2$	721.67 ab	737.00 a	47.16 a	53.27 a	54.47 a	60.14 a
$MgSO_4$ + $CaSO_4$	740.00 a	747.67 a	40.26 c	47.77 b	49.04 b	54.57 b
$MgCl_2$ + $CaCl_2$	676.67 b	682.33 b	45.02 b	52.60 a	53.85 a	59.44 a

* Values, within the same column, of similar letters were not significantly different when compared according to the least significant difference (LSD at 0.05 level).

Fruit Firmness (The Green Cheek): "Anna" apple firmness at the green cheek as influenced by preharvest treatments was recorded in Table 1. The data revealed that $CaCl_2$ treatment resulted in the greatest tissue firmness at harvest as compared with all other used treatments and the control. Furthermore, the incorporation of $MgCl_2$ to $CaCl_2$ did not make a significant alterations in tissue firmness at the green cheek in both seasons. On the other hand, a greater firmness was also obtained when either Ca^{2+} or Mg^{2+} solutions were applied in the sulphate form relative to the control. Meanwhile, there was no added advantage from mixing $MgSO_4$ to $CaSO_4$ in terms of their influence on tissue firmness at the green cheek as compared with just spraying $MgSO_4$ alone. Generally, the firmness results presented in this investigation agreed with those obtained by Bramlage *et al*. [17]; Bramlage *et al*. [18]; Davenport and Peryea [19]; Kadir [20] ; Raese and Drake [2] and Watkins *et al.* [3]. In addition, the obtained data agreed with that found by Wei Wei *et al*. [21] who indicated that foliar Ca^{2+} spraying and applying $CaSO_4$ into soil significantly increased the fruit firmness. The same trend was obtained by other studies such as Verela *et al*. [22] who found that treatment with $CaCl_2$ effectively retarded or avoided tissue softness in the apple fruits. This was explained on the basis that $CaCl_2$ treatment firms apple tissue by strengthening the cell wall and middle lamella [23-25]. Finally, the rate of calcium on increasing apples fruit firmness can be explained by that Ca^{2+} enters the fruit through lenticles and through the calyx [25, 26]. Moreover, cell wall-bound Ca^{2+} is involved in maintaining cell wall integrity by binding carboxyl groups of polygalacturonate chains, which are mainly present in the middle lamella and primary cell wall [27]. The role of Mg^{2+} could be also explained in increasing fruit firmness was similar to the role of Ca^{2+} in increasing cell membrane integrity by binding to the polar head groups of phospholipids. On the other hand, chloride salt treatments were more effective in improving fruit firmness than sulphate salt treatments, these results may be due to that chloride salt had greater permeability than sulphate salt, the amounts of calcium and magnesium increased thus the firmness values were consequently increased [28].

Chemical Characteristics

Total Soluble Solids (TSS): Changes in TSS in "Anna" apple fruits in response to various preharvest treatments were recorded in Table 2. The data revealed that $CaSO_4$- treated fruits had higher TSS values than that treated with $CaCl_2$. However, TSS of "Anna" apples was similar whether they were treated with Mg^{2+} chloride or sulphate in both seasons. However, all calcium solutions whether in the form of chloride or sulphate caused a significant increase in TSS as compared with the control. In addition, the chloride form of Ca^{2+} and Mg^{2+} in a combination did not lead to TSS values different from the single application of each one. On the contrary, the combination of Mg^{2+} and Ca^{2+} in the sulphate form had a significant effect on TSS as compared with the individual application of MgSO4 in both seasons. The presented data in this study agreed with that found by Kadir [20] who showed that preharvest tree sprays with Ca^{2+} have been used commercially to improve ratio of soluble solid concentration to titralable acidity of apple fruits.

Juice Acidity: Response of juice acidity of "Anna" apple fruits to various applied treatments before harvest was recorded in Table 2. The data indicated that such acidity was greater in control and $CaCl_2$ – treated fruits than that found in fruits of other treatments in both seasons of study. However, juice acidity of $CaSO_4$ – treated fruits was lower than that of the control in a consistent manner. The application of $MgCl_2$, on the other hand, resulted in lower acidity than both the control and $CaCl_2$ –treated fruits in both seasons. However, the combination of $MgCl_2$ plus $CaCl_2$ did not lead to a different acidity when compared with the individual application of $CaCl_2$ alone. Similarly, the combination of $MgSO_4$ plus $CaSO_4$ did not result in a considerable different juice acidity when compared with the application of $CaSO_4$ alone in both seasons. Thus, the individual application of Mg^{2+} in the form of sulphate did not really cause a significant difference in juice acidity as compared with its application in the form of chloride.

Vitamin C: Preharvest applications of various used treatments were recorded in Table 2 in terms of their influence on Vitamin C. The results revealed that the combination of both $MgCl_2$ and $CaCl_2$ caused a significant increase in vitamin C content of “Anna” apple juice as compared with the other applied treatments, also $CaCl_2$ and untreated fruits had significantly higher content of vitamin C in both seasons of study without a significant difference among both treatments only in the first season. $CaSO_4$ treatment whether used alone or in combination plus $MgSO_4$ were equally affective in vitamin C content only in the first season but in the second one, $MgSO_4$ plus $CaSO_4$- treated fruits had vitamin C significantly

Table 2: Some chemical characteristics of "Anna" apple fruit as influenced by pre-harvest treatments with calcium, magnesium as chloride or sulphate and their combinations during two seasons 2009 and 2010

	Chemical Characteristics															
	TSS (%)		Acidity (%)		Vitamin C (mg/100ml)		Electrolyte leakage (%)		Anthocyanin (mg/100ml)		Carotene Content (mg/l)		Chlorophyll a (mg/l)		Chlorophyll b (mg/l)	
Treatments	2009	2010	2009	2010	2009	2010	2009	2010	2009	2010	2009	2010	2009	2010	2009	2010
Control	10.53 c*	9.87 c	0.40 a	0.43 a	6.30 b	5.50 c	51.17 a	48.53 a	3.40 f	3.76 f	0.453 c	0.469 g	0.189 a	0.178 a	0.130 b	0.119 a
$MgSO_4$	11.20 b	10.67 bc	0.30 b	0.33 e	4.90 d	3.70 ef	32.68 b	30.00 c	4.93 c	5.62 c	0.833 b	0.902 c	0.133 c	0.126 c	0.086 bc	0.080 c
$CaSO_4$	12.80 a	12.33 a	0.32 b	0.35 cde	5.00 cd	4.00 e	33.94 b	31.00 b	6.13 a	7.53 a	0.947 a	1.000 a	0.113 e	0.104 e	0.060 c	0.055 e
$MgCl_2$	11.20 b	10.53 bc	0.31 b	0.34 de	4.60 d	3.50 f	33.46 b	30.58 b	4.23 e	4.72 e	0.457 c	0.500 f	0.159 b	0.144 b	1.020 a	0.095 b
$CaCl_2$	11.40 b	10.87 b	0.39 a	0.41 ab	6.90 b	6.20 b	27.73 d	26.17 e	4.83 cd	5.31 d	0.510 c	0.563 d	0.130 c	0.119 cd	0.091 bc	0.085 c
$MgSO_4$ + $CaSO_4$	12.87 a	12.47 a	0.32 b	0.37 bcd	5.60 c	4.60 d	30.81 c	28.17 d	5.60 b	7.08 b	0.883 b	0.963 b	0.120 d	0.114 d	0.079 bc	0.073 d
$MgCl_2$ + $CaCl_2$	11.60 b	11.27 b	0.38 a	0.39 bc	7.80 a	7.20 a	29.40 cd	27.80 d	4.47 de	5.14 d	0.493 c	0.521 e	0.131 c	0.123 cd	0.087 bc	0.079 cd

* Values, within the same column, of similar letters were not significantly different when compared according to the least significant difference (LSD at 0.05 level).

higher than those treated with $CaSO_4$ only. $MgSO_4$ and $MgCl_2$ treatments were similar in their influence in reducing the content of vitamin C of “Anna” apple juice in both seasons of study when compared with other used treatments.

Electrolyte Leakage: Response of electrolyte leakage to various preharvest treatments of "Anna" apples was reported in Table 2. The data indicated to a remarkable increase in electrolyte leakage was found with control as compared with all-other used treatments in both seasons. Moreover, the application of Ca^{2+} in the chloride form led to a considerable reduction in electrolyte leakage of the tissue as compared with its application in the sulphate form. However, the application of Mg^{2+} either in the chloride or sulphate led to similar leakage of electrolyte in the first season. The combination in the form of sulphate led to a significant decrease in electrolyte leakage as compared with just $CaSO_4$ alone or $MgSO_4$ alone in both seasons. The obtained results may be due to the roles of Ca^{2+} and Mg^{2+} in increasing cell membrane microviscosity and maintaining cell membrane integrity that are characteristics of senescent apple fruit [29-35]. Consequently there was a negative correlation between the percentage of electrolytes leakage and firmness [12, 13].

Anthocyanin Content: The data in Table 2 showed the influence of various used treatments before harvest on anthocyanin content in the skin of “Anna” apples. It was apparent that the greatest anthocyanin content was obtained with $CaSO_4$ treatment in both seasons. Moreover, the combination of $MgSO_4$ plus $CaSO_4$ led to a significant increase in anthocyanin content when compared with using $MgSO_4$ alone. When the two salts of $MgCl_2$ and $CaCl_2$ were combined and compared with $CaCl_2$ as individual, there was no difference in skin anthocyanin. In a similar manner, $MgCl_2$ or $CaCl_2$-treated apples caused a significant increase in anthocyanin content relative to the control in both seasons. The obtained results are in line with those obtained by kadir [20] who indicated that apple fruits skin color was improved by spraying the trees with calcium. Also, the found results were in line with those found by Hafez and Haggag [36] and Ashour [37].

Carotene Content: Change in carotene content in "Anna" apple fruit skin was assessed at harvest as reported in Table 2. It was evident that carotene responded in a similar manner to that found with the formation of anthocyanin since $CaSO_4$ treatment had the highest carotene content among all other treatment. Moreover, the combination of $MgSO_4$ plus $CaSO_4$ was able to enhance carotene in the fruits skin as compared with control. When $MgSO_4$ was combined with $CaSO_4$ the significant increase in carotene content was obtained as compared with using $MgSO_4$ as individual treatment, although, both treatments were similar in the first season in their effect on the content of carotene of "Anna" apple skin. In the same trend, $CaCl_2$ treatment was able to gain higher content of carotene than the combination of $MgCl_2$ plus $CaCl_2$ only in the second season since these treatments were similar in their effect on carotene content of "Anna" apple skins in the first season. $MgCl_2$-treated fruits had significantly the lowest content of carotene when compared with those of treatments, although, this treatment was equal in its effect with $CaCl_2$ whether alone or combined with $MgCl_2$ in the first season. Finally, the presented data revealed that all-used treatments were, in general, able to increase carotene content of "Anna" apple skin when compared with control treatment in both seasons.

Chlorophyll a: Assessment of chlorophyll a content in "Anna" apples skin as influenced by preharvest treatments with calcium and magnesium and their combinations was reported in Table 2. The data indicated that the highest content of chlorophyll a was found in un-treated fruits and $MgCl_2$- treated fruits in both seasons of the study. Meanwhile, $CaSO_4$-treated apples had significantly the lowest content of chlorophyll a. Moreover, $MgSO_4$, $CaCl_2$ and the combination of $MgCl_2$ plus $CaCl_2$ did not vary in their influence on the content of chlorophyll a but these treatments were significantly able to decrease this content of chlorophyll a as compared with control treatment. Furthermore, $MgSO_4$ plus $CaSO_4$ treatment gain apples with lower content of chlorophyll a, although, this treatment was equal in its influence with $CaCl_2$ when used alone or plus $MgCl_2$ in the second season.

Chlorophyll b: The data shown in Table 2 showed the influence of various treatments on chlorophyll b in the skin of “Anna” apples. It was apparent that $MgCl_2$ and control treatments resulted in the highest contents of chlorophyll b of “Anna” apple skins. However, $CaSO_4$ treated fruits had significantly the lowest content of chlorophyll b. Using $MgSO_4$ alone or combined with $CaSO_4$ and also, using $CaCl_2$ alone or plus $MgCl_2$ did not cause significant differences of the concentrations of chlorophyll b but these treatments were equal in their effect with $CaSO_4$ and control treatments in the first season. In the second season, nearly trend was found with the exception of the two combinations whether were $MgSO_4$ plus $CaSO_4$ or $MgCl_2$ plus $CaCl_2$ did not add advantage of the concentration of chlorophyll b of “Anna” apples skin. The data established in Table 2 which included the contents of anthocyanin, carotene, chlorophyll a and chlorophyll b of apples fruit peel revealed that all the used treatments in this study increased the percentage of anthocyanin and carotene and decreased the percentage of both chlorophyll a and b. Consequently, there was a negative relation between anthocyanin and carotene contents and chlorophyll a and b contents. These results may be due to that the applied treatments enhanced the activity of chlorophyllase enzyme which turned both kinds of chlorophyll from green pigments to colorless pigments, therefore, the main pigments which included anthocyanin and carotenes appeared. It could be also explained on the same bases, as ascribed above and might be due to the mild stress causing by Ca^{2+} and Mg^{2+} treatments leading to more production of ethylene then consequently carotene and anthocyanin formations in fruit skin. On the other side, the higher content chlorophyll b which were caused by Mg^{2+} treatments might be due to that magnesium is a main constituent of chlorophyll molecule bound by four pyroll groups consequently using concentration of Mg^{2+} in this study could be critical in influencing more synthesis of chlorophyll b in "Anna" apples skin. The presented data agreed with that indicated by Hafez and Haggag [36]; Verela *et al*. [22]; Picchioni *et al*. [38]; Picchioni *et al*. [39] ; Raese and Drake [2] ; Rocha *et al*. [40] ; Saftner *et al*. [24] and Wang *et al*. [41]. In the same trend, the results in this study were in line with those found by Sharples [42] who indicated that there was a negative relation among Mg^{2+} content and anthocyanin value of apple fruit.

The Second Study: Postharvest Treatments of "Anna" Apple Fruits

The Treatment Factor

Physical Characteristics

Weight Loss: Weight loss data of “Anna” apple cultivar as influenced by dipping after harvest in magnesium or calcium in the form of chloride salt as compared with Ethrel treatment was reported in Table 3. The results revealed that $CaCl_2$-treated apples either at 1% or 2% led to a significant reduction in weight loss as compared with the control in both seasons. However, the increase of $CaCl_2$ concentration resulted in a considerable reduction in weight loss in a consistent manner. Meanwhile, $MgCl_2$-treated apples also caused a significant reduction in weight loss relative to the control. There was no added advantage in the aspect when the concentration of $MgCl_2$ was increased from 1% to 2%. On the other hand, Ethrel-treated fruits after harvest had remarkably the greatest weight loss in both seasons as compared with the control or all other used treatments. The reduction in weight loss by either $CaCl_2$ or $MgCl_2$ might be attributed to their influence on maintaining the integrity of the plasma membrane since they bind to the polar head group of the phospholipid units of that membrane which reflect on retarding the cell deterioration and delaying senescence. Furthermore, calcium and magnesium play a vital role in preserving the cell wall structure which lead again to delaying fruit senescence and reflect on water loss. This explanation could be further supported by the finding that Ethrel, which accelerate fruit deterioration and senescence, causes a parallel increase in weight loss. Several other studies supported the findings of this research such as Raese and Drake [2] ; Hafez and Haggag [36] and Ashour [37] where they demonstrated that $CaCl_2$ treatment reduced the percentages of fruit weight losses in “Anna” apples.

Table 3: Some physical characteristics of "Anna" apple fruits as influenced by postharvest treatments with calcium, magnesium and chlorides or their combination and the commercial application of Ethrel during the two seasons 2009 and 2010

	Physical Characteristics					
	Weight loss (%)		Firmness of Red Cheek (Newton)		Firmness of Green Cheek (Newton)	
Factors	2009	2010	2009	2010	2009	2010
Time*						
Zero	0.00 b***	0.00 b	41.97 a	44.44 a	44.28 a	46.90 a
5 days	3.67 a	3.89 a	29.79 b	31.56 b	32.96 b	34.92 b
Treatments **						
Control	2.07 b	2.19 b	32.86 d	34.81 e	36.88 d	39.07 d
$CaCl_2$ 1%	1.82 c	1.92 c	36.83 b	39.01 b	39.38 b	41.71 b
$MgCl_2$ 1%	1.90 c	2.02 c	35.03 c	37.08 d	38.13 c	40.40 c
$CaCl_2$ 2%	1.65 d	1.76 d	37.21 b	39.41 b	39.38 b	41.70 b
$MgCl_2$ 2%	1.90 c	2.00 c	35.82 c	37.95 c	38.60 bc	40.91 bc
$CaCl_2$ + $MgCl_2$ 1%	1.64 d	1.75 d	38.69 a	40.98 a	42.02 a	44.53 a
$CaCl_2$ + $MgCl_2$ 2%	1.27 e	1.35 e	39.31 a	41.66 a	42.30 a	44.82 a
Ethrel 100 ppm	2.44 a	2.59 a	31.29 e	33.13 f	32.24 e	34.15 e

* The influence of the time factor.

** The influence of the treatment factor.

*** Values of similar letters, within the same column, were not significantly different according to the LSD values at 0.05 level.

Fruit Firmness (Red Cheek): Tissue firmness at the red cheek of "Anna" apple fruit was determined after postharvest treatments and the data was tabulated and shown in Table 3. The data indicated that $CaCl_2$-treated apple fruit whether at 1% or 2% caused a significant retardation in the loss of tissue firmness as compared with the control in both seasons. The increase in $CaCl_2$ concentration from 1% to 2% did not result in a proportional increase in fruit firmness. Meanwhile, a significant increase in tissue firmness at the red cheek was found with $MgCl_2$ treatment at both used concentrations (1% or 2%) relative to the control. It was more beneficial to combine both salts of calcium and magnesium to gain better tissue firmness. Moreover, treating "Anna" apples after harvest with Ethrel resulted in the lowest tissue firmness at the red cheek as compared with all other used treatments and the control.

Fruit Firmness (The Green Cheek): "Anna" apple firmness at the green cheek as influenced by postharvest treatments was recorded in Table 3. The data revealed that $CaCl_2$ or $MgCl_2$-treated fruits at 1% had significantly greater firmness at the green cheek than that of the control. It was beneficial to combine both calcium and magnesium salts to gain more advantage in tissue firmness at the green cheek. Meanwhile, Ethrel-treated fruits had the lowest tissue firmness at the green cheek as compared with the control and all other treated fruits in both seasons. These results were in agreement with those found by Dierend and Rieken [7] and Lee *et al*. [23].

Chemical Characteristics

Total Soluble Solids (TSS): Changes in TSS in "Anna" apple fruits in response to various postharvest treatments were recorded in Table 4. The data revealed that $CaCl_2$ treated fruit at 1% had significantly higher TSS than that of the control but did not vary from that treated with $CaCl_2$ at 2% in both seasons. However, $MgCl_2$ at 1% or 2% did not result in any significant change in TSS as compared with the control. However, $CaCl_2$-treated apples at 1% had superior TSS when compared with Ethrel at 100 ppm in both seasons. The application of Ethrel led to similar TSS to that found in the control. These results were in agreement with those indicated by Drake *et al*. [43]; kadir [20] and Hafez and Haggag [36].

Juice Acidity: Response of juice acidity of "Anna" apple fruit to various applied treatments after harvest was recorded in Table 4. The data indicated that juice acidity was greater in $CaCl_2$-treated fruits whether at 1% or 2% than that formed in the control fruits. Similar trend of results was found with the individual treatments of $MgCl_2$ at 1% or 2% relative to the control. Meanwhile, there was no significant difference in the acidity of $CaCl_2$ or $MgCl_2$ treated "Anna" apples at all used concentrations. In a similar manner, the combinations of $CaCl_2$ and $MgCl_2$ resulted in a similar acidity to that found by the individual application of each in both seasons. On the other hand, Ethrel-treated apples had juice acidity similar to that found in the control fruits. Moreover, treating fruits with Ethrel caused a significant reduction in juice acidity relative to

Table 4: Some chemical characteristics of "Anna" apple fruits as influenced by postharvest treatments with calcium, magnesium chlorides or their combinations and the commercial application of Ethrel during the two seasons of 2009 and 2010

	Chemical Characteristics															
	TSS (%)		Acidity (%)		Vitamin C (mg/100ml)		Electrolyte leakage (%)		Anthocyanin content (mg/100ml)		Carotene content (mg/L)		Chlorophyll a content (mg/L)		Chlorophyll b content (mg/L)	
Factors	2009	2010	2009	2010	2009	2010	2009	2010	2009	2010	2009	2010	2009	2010	2009	2010
Time*																
Zero	12.87 b***	13.13 b	0.473 a	0.493 a	7.47 a	7.47 a	22.32 b	23.68 b	3.70 b	3.94 b	0.630 b	0.672 b	0.178 a	0.190 a	0.111 a	0.119 a
5 days	13.85 a	14.12 a	0.378 b	0.398 b	4.98 b	5.19 b	34.57 a	36.69 a	6.10 a	6.52 a	0.891 a	0.591 a	0.125 b	0.133 b	0.083 b	0.089 b
Treatments**																
Control	13.13 b	13.40 b	0.392 b	0.410 b	5.83 de	5.88 de	30.86 b	32.74 b	3.97 e	4.23 e	0.669 e	0.715 e	0.168 a	0.180 a	0.106 a	0.113 a
$CaCl_2$ 1%	13.77 a	14.03 a	0.447 a	0.470 a	6.15 cd	6.23 cd	27.11 d	28.77 c	5.24 b	5.60 b	0.797 abc	0.851 b	0.145 c	0.154 d	0.095 bc	0.102 bc
$MgCl_2$ 1%	13.20 b	13.50 ab	0.425 a	0.447 a	6.13 cd	6.30 bcd	27.83 c	28.49 c	4.46 d	4.76 d	0.715 d	0.764 d	0.160 b	0.171 b	0.105 a	0.113 a
$CaCl_2$ 2%	13.33 ab	13.57 ab	0.447 a	0.463 a	6.37 bc	6.50 bc	27.11 d	28.74 c	5.30 b	5.65 b	0.813 ab	0.868 ab	0.144 c	0.154 d	0.093 cd	0.100 cd
$MgCl_2$ 2%	13.50 ab	13.83 ab	0.432 a	0.450 a	6.15 cd	6.30 bcd	27.14 d	28.78 c	4.57 d	4.87 d	0.715 d	0.764 d	0.156 b	0.169 bc	0.101 ab	0.109 ab
$CaCl_2$+$MgCl_2$ 1%	13.37 ab	13.63 ab	0.438 a	0.463 a	6.65 ab	6.70 ab	25.35 e	27.03 d	4.96 c	5.29 c	0.760 c	0.801 cd	0.148 c	0.159 cd	0.096 bc	0.104 bc
$CaCl_2$+$MgCl_2$ 2%	13.43 ab	13.70 ab	0.455 a	0.480 a	6.85 a	7.02 a	25.46 e	27.00 d	5.14 bc	5.47 bc	0.782 bc	0.835 bc	0.147 c	0.157 cd	0.096 bc	0.102 bc
Ethrel 100 ppm	13.13 b	13.33 b	0.370 b	0.382 b	5.63 e	5.68 e	36.70 a	38.93 a	5.58 a	5.95 a	0.833 a	0.893 a	0.141 c	0.150 d	0.087 d	0.094 d

* The influence of the time factor.

** The influence of the treatment factor.

*** Values of similar letters, within the same column, were not significantly different according to the LSD values at 0.05 level.

all $CaCl_2$ or $MgCl_2$ treated fruits. The reduction in juice acidity by Ethrel treatment could be ascribed to its influence on increasing the tissue respiration and increasing ripening-associated activities. The obtained results were in the same line with those found by Drake *et al*. [43] ; kadir [20] and Hafez and Haggag [36].

Vitamin C: Postharvest applications of various used treatments were recorded in Table 4 in terms of their influence on vitamin C. The results revealed that $CaCl_2$-treated fruits were equally effective in terms of vitamin C content whether at 1% or 2%. However, $CaCl_2$ at 2% led to more vitamin C content in the fruit than that found in the control fruits. Moreover, fruit treatments with $MgCl_2$ did not cause any significant change in vitamin C whether at 1% or 2% when compared with the control in both seasons. On the other hand, the combinations of both $CaCl_2$ and $MgCl_2$ (at 1% or 2%) caused a significant increase in vitamin C content in "Anna" apple juice as compared with the control. Meanwhile, the combinations did not vary from each other in their influence on vitamin C in both seasons. Ethrel-treated fruits had consistently lower vitamin C in the juice than all other treated fruits but equal to what was found in the control fruits. The highest vitamin C content was obtained with the combination of $CaCl_2$ plus $MgCl_2$ at 2% followed by the same combination at 1% in both seasons. These results could be ascribed to increasing the soluble matter in the juice by the penetrated calcium and magnesium chlorides.

Electrolyte Leakage: Response of electrolyte leakage to various postharvest treatments of “Anna” apples was reported in Table 4. The data revealed that $CaCl_2$-treated apple fruits at 1% significantly had lower percentage of electrolyte leakage than that of the control in both seasons. Typical trend of results was found when the concentration of $CaCl_2$ was raised to 2% since both concentrations did not vary in their influence on the leakage of electrolytes. On the other hand, $MgCl_2$ treatment at 1% or 2% resulted in lower electrolyte leakage than that of the control in both seasons. The combination of $CaCl_2$ plus $MgCl_2$ at 1% or 2% of each caused a significant reduction in electrolyte leakage relative to the control in a consistent manner. Meanwhile, these combinations (at 1% or 2%) were considerably more effective than the individual treatment. The remarkable increase in electrolyte leakage was found with postharvest treatment with Ethrel. This increase was superior to all other treatments and the control. The release of ethylene from Ethrel inside the tissue accelerated their senescence. Leakage of electrolytes through the cell plasma membrane is one of the main alterations as the tissue progresses toward senescence. On the contrary, magnesium and calcium maintain the integrity of the plasma membrane and the integrity of the cell wall which reflected on enhancing fruit quality and retarded the changes that lead to their deterioration.

Anthocyanin Content: The data in Table 4 showed the influence of various used treatments after harvest on anthocyanin content in the skin of “Anna” apples. It was apparent that the greatest anthocyanin content was obtained with Ethrel treatments at 100 ppm. Moreover, $CaCl_2$-treated apples whether at 1% or 2% still was able to enhance the formation of anthocyanin in fruit skin after harvest without any added advantage in this respect, when the concentration of $CaCl_2$ was increased from 1% to 2%. In a similar manner, $MgCl_2$-treated apples caused a significant increase in anthocyanin content relative to the control without any significant change by increasing its concentration from 1% to 2% (w/v). The increase in anthocyanin content whether by $MgCl_2$ or $CaCl_2$ could be attributed their influence on causing a mild chemical stress on the fruit at room temperature which reflected on enhancing ethylene production and increased this pigment formation. The results of this study were similar to those obtained by several investigators such as kadir [20]; Hafez and Haggag [36]; Wei Wei *et al*. [22] and Trentham *et al*. [44].

Carotene Content: Changes in carotene content in “Anna” apple fruit skins was assessed by the end of the shelf life test after 5 days at ambient temperature then reported in Table 4. It was evident that carotenes responded in a similar manner to that found with the formation of anthocyanin since Ethrel treatment (100 ppm) resulted in the highest carotene content among all other treatments. Moreover, calcium and magnesium salts at 1% or 2% (w/v) were able to enhance carotene formation in the fruit skin as compared with the control in both seasons. Thus, the use of only 1% with both nutrients (Mg^{2+} or Ca^{2+}) was sufficient to obtain a better enhancement in carotene formation in these fruits after harvest. It could be also explained on the same bases as ascribed above and might be due to the mild stress imposed by $CaCl_2$ or $MgCl_2$ treatments leading to more production of anthocyanin then consequently carotene formation in fruit skin.

Chlorophyll a: Assessment of chlorophyll a content in "Anna" apples skin as influenced by postharvest treatments with calcium or magnesium and their combinations was reported in Table 4. The data indicated that there was a reduced concentration of chlorophyll a by all treatments after harvest. Furthermore, no further reduction in chlorophyll a was induced by increasing the concentration from 1% to 2% whether in case of $CaCl_2$ or $MgCl_2$ salts after harvest when compared with each other in both seasons. Even the combination of both elements at 1% or 2% had similar influence on chlorophyll a content in the skin of "Anna" apples after harvest and five days on the shelf of ambient temperature in both seasons. Meanwhile, the influence of Ethrel treatment on chlorophyll a concentration was similar to that found by the effect of $CaCl_2$ alone or its combination with $MgCl_2$ (at 1% or 2%). The reduction in chlorophyll a content by the end of the shelf life period at ambient temperature might explain again an increase in chlorophyllase enzyme activity due to a mild increase in ethylene production in fruit skin.

Chlorophyll b: Data of chlorophylle b in fruit skin of "Anna" apples at the end of the shelf life test was reported in Table 4. The data revealed that the lowest chlorophyll b content was found with Ethrel treated apples at 100 ppm. However, chlorophyll b concentration did not significantly vary from that obtained with $CaCl_2$ treatment at 2% (w/v). Moreover, $CaCl_2$ at 1% was also effective as $CaCl_2$ at 2% in terms of chlorophyll b in fruit skin of "Anna" apples. On the other hand, the dipping treatment in $MgCl_2$ solutions whether at 1% or 2% resulted in similar chlorophyll b to that found in the control fruit in both seasons. The above results could be explained on the bases that magnesium is a main constituent of chlorophyll molecule bound by four pyroll groups. Thus used concentration of Mg^{2+} could be critical in influencing more synthesis of chlorophyll b in "Anna" apples skin. Meanwhile, the release of ethylene in fruit tissue as a result of Ethrel treatment could explain the considerable reduction of chlorophylls a and b in fruit skin in a consistent manner by the end of the shelf life duration.

The Time Factor: The influence of time factor on fruit characteristics of "Anna" apples at ambient temperature was reported in Tables 3 and 4. The data in Table 3 revealed that over 5 days of incubation, these fruits lost from 3– 4% of their weight which considered as a significant loss, regardless of the treatments, while firmness of the red cheek was also considerably reduced in both seasons. In a similar manner, the green cheek (unexposed) of the fruit lost some firmness but the degree of tissue firmness loss was similar in both cheeks. These results were in agreement with that documented by other researchers such as Hafez and Haggag [36]; Picchioni *et al*. [38] and Verela *et al*. [22].

Chemical characteristics of "Anna" apples were also affected as a result of incubation at ambient temperature for 5 days. It was evident that total soluble solids were significantly increased due to concentrating the apple juice as s result of water loss, while acidity was reduced in both seasons which could be ascribed to the conversion of some acids to sugars by the gluconeogenesis reactions or utilizing some organic acids to produce energy through the aerobic respiration. Moreover, vitamin C was reduced and that was expected since ascorbic acid is one of the main organic acids in the fruits. On the other hand, leakage of electrolytes was consistently increased in both seasons, but did not reach 50% that has been considered the concentration indicating to the cell membrane injury. Thus, used treatments did not cause a membrane damage which meant that tissues were still healthy and did not reach the senescence stage. Pigments of the skin such as anthocyanin and carotenes, the main quality attributes of apples, were increased in a highly significant manner, while chlorophyll a and b were considerably reduced as a result of exposure to light at 22±2° C and the activation of ethylene production by some of the used treatments such as Ethrel. The obtained results were in parallel with that found by Hafez and Haggag [36]. They emphasized that TSS and anthocyanin contents increased while total acidity content of “Anna” apple fruits decreased with the progress of storage period.

The Effect of the Interaction Between Treatments and Time: Weight loss of "Anna" apples as influenced by the interaction between postharvest treatments and the time factor (Table 5) revealed that the highest increase in weight loss over the five days after treatments was obtained with Ethrel at 100 ppm followed by the control in both seasons. Meanwhile, the application of calcium or magnesium resulted in drastic reduction in weight loss especially dipping in $CaCl_2$ at 2% (w/v). The least percentage of weight loss was found with the combination of $CaCl_2$ plus $MgCl_2$ at 2%. The reduction in weight loss could be due to maintaining the integrity of the cell membrane by applying calcium or magnesium. These elements bound to the polar head groups of this membrane phospholipids which retard the changes leading to cell senescence and leakage of electrolytes. These two important elements can also bind to the polysaccharides forming the cell wall of apple tissue which might increase the formation of calcium or magnesium pectate, thus leading to less deterioration of fruit tissues.

With regard to apple tissue firmness at the red (exposed) cheek of the fruit as influenced by the interaction between treatments and the time factor, the date, in Table 5, showed that the greatest fruit firmness was found at the initiation of the experiment then the tissue started to lose its firmness as expected, but the rate of such loss was remarkable with Ethrel treatment (to about half of the initial firmness of the red cheek). Moreover, all treatments were able to maintain a better fruit firmness as compared with the control. As the two nutrients were combined together at 1% or 2%, a greater firmness was found in both seasons when compared with using only one of them. Thus, in this case, an added advantage was obtained when two elements were used together.

Table 5: Some physical characteristics of "Anna" apple fruits as influenced by the interaction between the applied treatments after harvest and the time factor during the two successive seasons 2009 and 2010

	Physical Characteristics					
	Weight loss (%)		Firmness of red cheek (Newton)		Firmness of green cheek (Newton)	
Treatments	2009	2010	2009	2010	2009	2010
Control	4.14 b*	4.38 b	23.76 f	25.18 f	29.47 e	31.24 e
$CaCl_2$ 1%	3.63 c	3.83 d	31.70 c	33.58 d	34.48 c	36.52 c
$MgCl_2$ 1%	3.80 c	4.04 c	28.08 e	29.72 f	31.98 d	33.90 d
$CaCl_2$ 2%	3.30 d	3.51 e	32.44 c	34.38 d	34.49 c	36.50 c
$MgCl_2$ 2%	3.79 c	4.00 c	29.66 d	31.45 e	32.94 d	34.92 d
$CaCl_2$ + $MgCl_2$ 1%	3.28 d	3.49 e	35.41 b	37.51 c	39.76 b	42.16 b
$CaCl_2$ + $MgCl_2$ 2%	2.54 e	2.69 f	36.66 b	38.87 b	40.32 b	42.75 b
Ethrel 100 ppm	4.88 a	5.17 a	20.61 g	21.82 g	20.20 f	21.40 f
Initial Values	0.00 f	0.00 g	41.97 a	44.44 a	44.28 a	46.90 a

* Values of similar letters, within the same column, were not significantly different according to the LSD values at 0.05 level.

Table 6: Chemical characteristics of "Anna" apple fruits as influenced by the interaction between the applied treatments after harvest and the time factor during the two successive seasons 2009 and 2010.

	Chemical Characteristics															
	TSS (%)		Acidity (%)		Vitamin C (mg/100ml)		Electrolyte leakage (%)		Anthocyanin content (mg/100ml)		Carotene content (mg/L)		Chlorophyll a content (mg/L)		Chlorophyll b content (mg/L)	
Factors	2009	2010	2009	2010	2009	2010	2009	2010	2009	2010	2009	2010	2009	2010	2009	2010
Control	13.40 cd*	13.67 cde	0.310 d	0.327 e	4.20 e	4.30 e	39.40 b	41.80 b	4.23 e	4.54 f	0.708 f	0.758 f	0.159 b	0.170 b	0.100 b	0.106 b
$CaCl_2$ 1%	14.67 a	14.93 a	0.420 bc	0.447 bc	4.83 d	5.00 d	31.91 d	33.85 d	6.78 b	7.26 bc	0.963 bc	1.031 bc	0.112 de	0.118 de	0.79 d	0.084 d
$MgCl_2$ 1%	13.53 bcd	13.87 bcd	0.377 c	0.400 d	4.80 d	5.13 d	33.33 c	35.30 c	5.21 d	5.58 e	0.800 e	0.856 e	0.142 c	0.151 c	0.099 bc	0.106 b
$CaCl_2$ 2%	13.80 bc	14.00 bcd	0.420 bc	0.433 bcd	5.27 cd	5.53 cd	31.90 d	33.80 d	6.90 b	7.36 b	0.995 ab	1.065 ab	0.110 de	0.117 de	0.075 d	0.080 d
$MgCl_2$ 2%	14.13 ab	14.53 ab	0.390 c	0.407 cd	4.83 d	5.13 d	31.95 d	33.89 d	5.43 d	5.80 e	0.800 e	0.855 e	0.134 c	0.144 c	0.091 c	0.096 bc
$CaCl_2$ + $MgCl_2$ 1%	13.87 bc	14.13 bcd	0.403 bc	0.433 bcd	5.83 bc	5.93 c	28.37 e	30.39 e	6.21 c	6.64 d	0.890 d	0.930 d	0.119 d	0.128 d	0.081 d	0.088 cd
$CaCl_2$ + $MgCl_2$ 2%	14.00 abc	14.27 abc	0.437 ab	0.467 ab	6.23 b	6.57 b	28.59 e	30.31 e	6.57 bc	7.00 c	0.934 cd	0.998 c	0.116 d	0.124 d	0.080 d	0.085 d
Ethrel 100 ppm	13.40 cd	13.53 de	0.267 d	0.270 f	3.80 e	3.90 e	51.07 a	54.18 a	7.45 a	7.96 a	1.037 a	1.115 a	0.103 e	0.109 e	0.063 e	0.068 e
Initial Values	12.87 d	13.13 e	0.473 a	0.493 a	7.47 a	7.47 a	22.32 f	23.68 f	3.70 f	3.94 g	0.630 g	0.672 g	0.178 a	0.190 a	0.111 a	0.119 a

* Values of similar letters, within the same column, were not significantly different according to the LSD values at 0.05 level.

Firmness of "Anna" apples at the green cheek as influenced by the interaction between treatments and time factor was also reported in Table 5. A trend almost similar to that found with the red cheek, was also obtained in this side of the fruit. The data emphasized again that the highest rate of fruit tissue deterioration was caused by Ethrel at 100 ppm, while calcium and magnesium chlorides at 1% or 2% were capable of retarding the rate of tissue firmness loss even at ambient temperature which simulates the situation during marketing these fruits in many countries.

Chemical characteristics of "Anna" apples were also examined in relation to the influence of interaction between treatments and the time factor (Table 6). The data proved that there was no significant change in TSS of the control fruits over 5 days on the shelf as compared with the initial values. The only considerable increase in TSS was found with $CaCl_2$ 1% followed by $MgCl_2$ at 2% (w/v). Even Ethrel treatment did not result in a significant change in TSS over the five days of incubation at room temperature when compared with the initial TSS or the control fruits over time.

Changes in juice acidity of "Anna" apple fruits as affected by the interaction between treatments and the time factor (Table 6) were also maintained and indicated that there was a marked reduction in acidity as compared with the initial values in both seasons except with the treatment of combined $CaCl_2$ + $MgCl_2$ at 2%. Even the reduction in juice acidity by Ethrel dipping was greater than all other single or combined treatment of $CaCl_2$ or $MgCl_2$ at either 1% or 2% in a consistent manner in both seasons.

Since ascorbic acid (vitamin C) is one of the major organic acids in the juice, the data in Table 6 showed almost a typical trend to that found in titratable acidity of the juice since the highest acidity was found with the initial value while the lowest acidity was detected over the incubation time in the fruit treated with Ethrel (100 ppm) and the control fruits. However, it was found that vitamin C in the juice was significantly decreased by each of $CaCl_2$ and $MgCl_2$ at 2% than their combination in both seasons. Thus, calcium and magnesium chloride treatments after harvest were successful in preserving a desired juice acidity as requested by the consumers, whereas Ethrel-treated fruit or the control had a drastic reduction in fruit acidity and vitamin C.

Leakage of electrolytes as a very important parameter of membrane integrity and the progress toward senescence, the interaction between treatments and the time factor in Table 6, revealed that fruits of both seasons were similar in terms of their initial percentage of electrolyte leakage. The highest leakage occurred by Ethrel treatment over the incubation period (5 days), followed by the leakage from the control fruits. On the other hand, the lowest electrolyte leakage over time was evident by the dipping in the combination of $CaCl_2$ plus $MgCl_2$ at 1% or 2% in both seasons. Thus, the individual treatment of Ca^{2+} or Mg^{2+} solutions were also effective in delaying tissue senescence over time but less than when both elements were combined.

With regard to the anthocyanin content in the skin of "Anna" apple fruits as influenced by the interaction between postharvest treatments and the time factor, the data indicated that there was a remarkable increase in this pigment by Ethrel treatment as compared with the initial value of zero time. It was also very beneficial to gain an increase in anthocyanin content in the skin of "Anna" apples resulting from using various calcium or magnesium salts and their combinations since they also caused an increase in fruit firmness and considerably reduced membrane damage and cell wall hydrolysis. Thus, it is very important outcome in this study to show to apple growers retailers that there is a possibility to enhance important quality attributes of apple fruits such as firmness and skin color with postharvest dipping in $CaCl_2$ or $MgCl_2$ and retard tissue senescence. It was also found that by increasing tissue ethylene through the application of Ethrel, the physiological disorder of soft scaled dramatic increased in "Anna" apple fruits [45].

Carotene contents in the skin of treated "Anna" apples as affected by the interaction between treatments and the factor of time tended to be almost typical to results found with anthocyanin content in both seasons. Consumers have been encouraged to eat apple fruits not only for the sake of nutrients and vitamins, but also for the great beneficial content of antioxidants in their skins such as carotenes. As reported in Table 6, various used treatments in this study, were able to enhance carotene content in the fruit skin over time with undesired effects only obtained with dipping in Ethrel after harvest in both seasons.

The changes in chlorophyll a and b were also assessed in terms of their response to the interaction between the treatments and the factor of time. There were obvious trends in similar responses since the highest chlorophylls (a and b) were recorded at the initial time followed by the control, then calcium and magnesium chlorides (at both 1% or 2% w/v) were able to cause a significant reduction in chlorophylls in both seasons except with dipping in $MgCl_2$ (1%) that did not considerably influence chlorophyll b as compared with the control.

REFERENCES

1. DeEll, R.J., F. Saad and S. Khanizadeh, 1999. Factors influencing apple fruit firmness. The 42nd Annual IDFTA Conference, February 20-24, Hamilton, Ontario.
2. Raese, J. T. and S. R. Drake, 2002. Ca spray materials and fruit Ca concentrations influence on apple quality. J. Amer. Pomol. Soc., 56: 136-143.
3. Watkins, C.B., E.W. Hewett, C. Bateup, A. Gunson and C.M. Triggs, 1989. Relationships between maturity and storage disorders in "Cox's Orange Pippin " apples as influenced by preharvest calcium or ethephon sprays. New Zealand J. Crop Hort. Sci., 326: 231-235.
4. Mason, J.L., B.G. Drought and J.M. McDouglad, 1974. Effect of a calcium chloride dip on senescent breakdown, firmness and calcium concentration in "Spartan" apple. Hort Science, 9: 596.
5. Sams, C.E. and W.S. Conway, 1984. Effect of calcium infiltration on ethylene production, respiration rate, soluble polyuronide content and quality of "Golden Delicious" apple fruit. J. Amer. Soc. Hort. Sci., 109: 53-57.
6. Beavers, W.B., C.E. Sams, W.S. Conway and G.A. Brown, 1994. Calcium source affects calcium content, firmness and degree of injury of apples during storage. Hort Science, 29: 1520-1523.
7. Dierend, W. and S. Rieken, 2007. Postharvest treatment of apples with Ca chloride. Erwerbs-Obstbau, 49: 51-56.
8. Conway, W.S., 1989. Altering nutritional factors after harvest to enhance resistence to postharvest disease. Phytopathology, 79: 1384-1387.
9. Ghoname, M.I., 1992. Effect of nitrogen, potassium and their interactions on quantitative, qualitative characteristics of some strawberries cultivars. Ph.D. Thesis Fac. of Agric., Alexandria Univ., Egypt.
10. Spayed, S.E. and J.R. Morris, 1981. Effects of immature fruit and holding on strawberry puree quality and colour stability. J. Amer. Soc. Hort. Sci., 106: 211-216.
11. Egan, H., R.S. Kird and R. Sawyer, 1987. Pearson's Chemical Analysis of Foods. Eight Edition-Longman Scientific and Technical. Essex CM_{20}.2 TE, England.
12. Gary, B.C.M., 1970. Effect of Ca^{2+} on the water and electrolyte permeability of phospholipd membranes. Nature., 228: 1101-1102.
13. Ahrenes, M.J. and D.L. Ingram, 1988. Heat tolerance of citrus leaves. Hort. Science, 23: 747-748.
14. Fuleki, T. and F.J. Francis, 1968. Quantitive methods for anthocyanin. 1. Extraction and determination of total anthocyanin in cranberries. J. Food Sci., 33: 72-77.
15. Wintermans, J.F.M. and D.E. Mots, 1965. Spectrophotometric characteristics of chlorophylls and their pheophytins in ethanol. Biochem. Biophys. Acta., pp: 448-453.

16. Steel, R.G.D. and T.H. Torrie, 1980. Principles and procedures of statistics. (Mc. Grow-Hill, Co., U.S.A.).
17. Bramlage, W.J., M. Drake and J.H. Baker, 1979. Changes in calcium level in apple cortex tissue shortly before harvest and during postharvest storage. Commum. Soil Sci. Plant Anal., 10: 417-426.
18. Bramlage, W.J., M. Drake and S.A. Weis, 1985. Comparisons of calcium chloride, calcium phosphate and calcium chelate as foliar sprays for "McIntosh" apple trees. J.Amer. Soc. Hort. Sci., 110: 786-789.
19. Davenport, J.R. and F.J. Peryea, 1990. Whole fruit mineral element composition and quality of harvested "Delicious" apples. J. Plant Nutr., 13: 701-711.
20. Kadir, S.A., 2005. Influence of preharvest Ca application on storage quality of "Jonathan" apples in Kansas. Trans. Aca. Sci., 108: 129-138.
21. Wei Wei, C. Feng, X. Yuehua, Z. Jing and Z. Heng, 2007. Interactive effects of organic fertilizers, $CaSO_4$ and amino acid Ca on Fuji apple in Burozem soil in China. Front. Agric. China, 1: 460-467.
22. Verela, P., A. Salvador and S.M. Fiszman, 2007. The use of calcium chloride in minimally processed apples: A sensory approach. Eur. Food. Res. Technol., 224: 461-467.
23. Lee, J.Y., H.J. Park, C.Y. Lee and W.y. Choi, 2003. Extending shelf life of minimally processed apples in edible coatings and antibrowning agents. Lebensm–Wiss U– Technol., 36: 323-329.
24. Saftner, R., B.J Abbott and J.Y Lee, 2003. Sanitary dips with calcium propionate, chloride, or a calcium amino acid chelate maintain quality and shelf stability of fresh-cut honeydew chunks. Postharvest Biol. Technol., 29: 257-269.
25. Betts, H.A. and W.J. Bramlage, 1977. Uptake of Ca by apples from postharvest dips in Ca chloride solutions. J. Amer. Soc. Hort. Sci., 102: 785-788.
26. Haker, F.R. and I.B. Ferguson, 1988. Transport of Ca across cuticles isolated from apple fruit. Scientia Hort., 36: 205-217.
27. Tobias, R.B., W.S. Conway and C.E. Sams, 1993. Cell wall composition and calcium-treated apples moculated with *Botrytis cinerea*. Phytochemistry, 32: 35-39.
28. Glenn, G.M. and B.W. Poovaiah, 1985. Cuticular permeability to Ca compounds in "Golden Delicious" apples. J. Amer. Soc. Hort. Sci., 110: 116-171.
29. Paliyath, G., B.W. Poovaiah, G.R. Munske and J.A. Magnuson, 1984. Membrane fluidity in senescing apples: Effects of temperature and calcium. Plant Cell Physiol., 25: 1083-1087.
30. Glenn, G.M., A.S.N. Reddy and B.W. Poovaiah, 1988. Effect of calcium on cell wall structure, protein phosphorylation and protein profiles in senesceing apples. Plant Cell Physiol., 29: 565-572.
31. Van Goor, B.J., 1971. The effect of frequent spraying with calcium nitrate solutions on the mineral composition and the occurrence of bitter pit of the apple Cox's Orange Pippin. J. Hort. Sci., 46: 347-364.
32. Cooper, T. and F. Bangerth, 1976. The effect of Ca and Mg treatments on the physiology, chemical composition and bitter-pit development of 'Cox's Orange' apples. Sci. Hort., 5: 49-57.
33. Poovaiah, B.W., G.M. Glenn and A.S.N. Reddy, 1988. Calcium and fruit softening, physiology and biochemistry. Hort. Res., 10: 107-152.
34. Lurie, S. and R. Ben-Arie, 1983. Microsomal membrane changes during ripening of apple fruit. Plant physiol., 73: 636-638.
35. Lurie, S., L. Sonego and R. Ben-Arie, 1987. Permeability, microviscosity and chemical changes in the plasma membrane during storage of apple fruit. Sci. Hort., 32: 73-83.
36. Hafez, O.M. and K.H.E. Haggag, 2007. Quality improvements and storability of apple cv. Anna by preharvest applications of boric and calcium chloride. Research Journal of Agriculture and Biological Sciences, 3: 176-183.
37. Ashour, N.N., 2000. Effect of environmental factors, calcium and potassium fertilization on yield and quality of apple. Ph.D. Thesis, Fac. Agric., Mansoura Univ., Egypt.
38. Picchioni, G.A., A.E. Watada, W.S. Conway, B.D. Whitaker and C.E. Sams, 1995. Phospholipid, galactolipid and sterylipid composition of apple fruit cortical tissue following postharvest $CaCl_2$ infiltration. Phytochemistry, 39: 763-769.
39. Picchioni, G.A., A.E. Watada, W.S. Conway, B.D. Whitaker and C.E. Sams, 1998. Postharvest calcium infiltration delays membrane lipid catabolism in apple fruit. J. Agric. Food chem., 46: 2452-2457.
40. Rocha, A., C. Brochads and A. Morais, 1998. Influence of chemical treatment on quality of cut apple. (cv Jonagored). J. Food Qual., 21: 13-28.
41. Wang, C.Y., W.S. Conway, J.A. Abbott, G.F. Kramer and C.E. Sams, 1993. Postharvest infiltration of polyamines and calcium influences ethylene production and texture changes in "Golden Delicious" apples. J. Amer. Soc. Hort. Sci., 118: 801-806.

42. Sharples, R.O., 1968. The structure and composition of apples in relation storage quality. Rpt. E. Malling Res. Sta., pp: 182-189.
43. Drake, S.R., T.A. Eisele, D.C. Elfving, M.A. Drake and S.L. Drake, 2005. Effects of the bioregulators aminoethoxy-vinylglycine and ethephon on brix, carbohydrate, acid and mineral concentrations in "Scarletspur Delicious" apple juice. Hort. Science, 40: 1421-1424.
44. Trentham, R.W., C.E. Sams and W.S. Conway, 2008. Histological effects of calcium chloride in stored apples. J. Amer. Soc. Hort. Sci., 133: 487-491.
45. Farag, K.M., 2006. Effect of Ethrel pH on the shelf life and incidence of scalding of "Anna" apple fruits. J. Agric. & Environ. Sci., 5: 21-31.