Postharvest Application of Ozone and Calcium Chloride to Control of "Anna" Apple Fruit Mold during Cold Storage

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ABSTRACT

The present study were carried out on"Anna" Apple fruits in two successive seasons 2008 and 2009 to study the effect of postharvest treatments $CaCl_2$ dip for 2 min, O_3 application in storage conditions of 4°C and 85-90% RH on fruit decay.

It was found that ozone treated fruits had higher weight loss than that of the control. However, but the differences were not significant. Skin color improved as the anthocyanin pigment concentration increased, O_3 treatment significantly changes fruit skin color. $CaCl_2$ a treated fruit was the firmest and had the highest soluble solids content. Generally, fruit acidity decreased during storage period. O_3 and $CaCl_2$ treatments reduced decay in apple fruits.

INTRODUCTION

Apples must be harvested when mature not when fully ripe. If harvest too early fruits often small, poor in flavor and color, may fail to fully ripen and more susceptible to scald and bitter-pit disorders. Conversely, apples harvested when over-mature are vulnerable to mechanical injury and disease, develop off-flavors, and often have a higher occurrence of water core and senescent breakdown (Meheriuk *et al.*, 1994). The importance of calcium in apple fruit is its role is maintaining the optimum fruit firmness quality during postharvest storage and fruit ripening. This role is seen directly in the prevention of specific disorders such as bitter pit, and in relationships between calcium and more general quality properties such as flesh firmness (Ian Ferguson, 2001).

Ozone has been evaluated for postharvest disease control and other storage uses for many years. Some commercial use has occurred with some commodities such as apples, cherries, kiwi, peaches, plums and table grapes. There is increasing interest and a great deal of empirical activity in the evaluation of ozone for a diversity of water treatment and air treatment (fumigation) uses in postharvest quality management. The benefits of ozone treatment in cold storage rooms or facilities may be direct and indirect (Trevor, 2003). Accordingly, the objective of this study was to obtain high quality fruits of "Anna" apple and free of mold during cold storage by post harvest application of ozone and calcium chloride.

MATERIALS AND METHODS

The present investigation was carried out during 2009 and 2010 seasons on fresh Anna apple fruits. The fruits were harvested at maturation stage with red color from orchard located in Nobaria-Behera governorate.

Harvested fruits were packed in field boxes and immediately transported to the Postharvest Center, Faculty of Agriculture, Alexandria University. Undamaged fruits were selected on the basis of size uniformity and color.

Selected fruits were washed with running water to element the spines and them dried by an electric fan. Another sorting was done to recheck the fruits for any defects. The initial quality of apple fruits was done and tabulated in Table (1).

Sound selected apple fruits were divided to 4 groups each one contained 21 fruits. Each group was placed in 3 open carton boxes each one contained 7 fruits and every group received one of the following treatments:

1. Fruits treated with ozone (150 ppb) and dipped in $CaCl_2$ for 2 min. and stored at 4°C, (T_1).
2. Fruits dipped in $CaCl_2$ for 2 min and stored at 4°C, (T_2).
3. Fruits treated with ozone (150 ppb) and stored at 4°C, (T_3).
4. Fruits stored at 4°C, as the control (T_4).

Physical characteristics:

1- Weight loss

In the present experiment 3 fruits of each replicate, each treatment were initially weighed and labeled.

The initial weight of each labeled 3 fruits was recorded to calculate fruit weight loss percent at 14 days intervals throughout the course of storage at 4°C. The average weight loss percent was calculated for each treatment according to the following equation:

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$$\text{Weight loss}\% = \frac{\text{Initial weight - Sample weight}}{\text{Initial weight}} \times 100$$

2- Firmness:

Fruit firmness was measured on 3 fruits sets with manually controlled digital set. Firmness was measured at two opposite points on the equator of the fruit after removal of a 2 mm slice with a fixed blade slicer at the end of an 1-d period at 20°C (Chen and Mellenthin, 1981).

Chemical characteristics:

Changes of chemical parameters were determined using three fruits for every replicate during storage period at the previously mentioned intervals as before above described.

1- Soluble Solids Content (SSC):

The Soluble solids content (SSC) of fruit juice was determined using a hand refrectometer (ATAGO, mod. N-1E, Japan). A.O.A.C. (1990).

The Soluble solids content values were expressed as percent (%).

2- Titratable acidity:

The titratable acidity of fruit juice was determined using five milliliters form the obtained juice for the titration; 0.1N sodium hydroxide was used in the presence of phenolphethalein as an indicator according to A.O.A.C. (1990). The titratable acidity was expressed as gms of citric acid per 100 ml of apple fruit juice.

3- Peel anthocyanin content (mg/ 100 g peel):

Anthocyanin of peel was determined following the method of Fuleki and Francis (1968). Three samples of 50 gm peel of each treatments were extracted in 100 ml mixture of ethanol and HCl (85:15).

Five milliliters of the above extraction were centrifugated for 10 min, then were measured spectro-photometrically at 535 nm and peel anthocyanin content was calculated as mg/ 100 g sample.

The termination of the experiment was done by the occurrence of peel disorders (Shrinkage, drying and off shining). All data were statistically analyzed according to Snedecor and Cochran (1980). The individual comparisons were carried out by using the least significant differences (LSD) according to SAS Institute (1985).

The experimental design was factorial (RCBD) with three replicates and the data obtained were statistically analyzed according to Snedecor and Cochran (1973).

RESULTS AND DISCUSSION

A- Physical characteristics:

1- Weight loss (%):

The presented in Table (2), showed that weight loss percentages were gradually increased, as an average, for all treatments in both seasons, as the storage period increased. The differences among all storage periods were not significant in both seasons. These results in the same line of Palou *et al.* (2002) and Softner (1999) on peach.

Moreover, the data of the present study pointed out that high significant differences were noticed between $CaCl_2$ all other treatments in the first season. In the second season, one there were significant difference was found between fruits dipped in $CaCl_2$ for 2 min and fruits, treated with ozone or $CaCl_2$ only.

These results were in agreement with those observed by EL-Saedy *et al.* (2010) on mango.

2- Firmness:

The data in Table (3) showed that the highest significant fruit firmness was noticed for O_3+ $CaCl_2$ treatment, the lowest value was for control treatment in the first season. On the other hand, there was no significant differences among the remaining treatments in 2009 and among all treatments in 2010. Conway *et al.* (2003) reported that calcium in adequate amounts helps to maintain apple fruit firmness.

Table1. The initial quality of Anna apple fruits

Replicates	Firmness 16/ cm^2	SSC (%)	Acidity %	Anthocyanin mg/ 100 gm peel
			Season 1	
1	18.4	12.2	0.72	18.07
2	17.8	10.0	0.75	24.09
3	19.0	10.6	0.97	19.92
Average	18.4	10.93	0.81	20.69
			Season 2	
1	19.55	13.0	0.82	28.16
2	18.0	12.4	0.73	28.96
3	18.5	12.2	0.79	32.73
Average	18.68	12.53	0.78	29.95

Table 2. Effect of ozone and calcium chloride treatments on weight loss (%) of "Anna" apple fruits in 2009 and 2010 seasons

	Storage period (days interval)					
Treatments	**1**	**2**	**3**	**4**	**5**	**Means**
			Season 2009			
Ozone + $CaCl_2$	0	1.06	1.97	2.61	3.87	1.91
$CaCl_2$	0	1.32	2.86	3.38	3.93	2.20
Ozone	0	1.09	1.99	3.05	3.37	1.89
Control	0	0.97	1.55	2.68	3.31	1.70
Means	0	1.11	1.97	2.93	3.62	
			Season 2010			
Ozone + $CaCl_2$	0	0.88	1.71	2.53	3.26	1.67
$CaCl_2$	0	1.18	2.04	3.08	4.02	2.07
Ozone	0	1.17	2.04	3.32	3.48	2.00
Control	0	0.84	1.38	1.81	2.16	1.23
Means	0	1.02	1.79	2.68	3.23	

$L.S.D_{0.05}$	A	B
Season 1	0.28	0.32
Season 2	0.317	0.35

Table 3. Effect of ozone and calcium chloride treatments on firmness (Ib/ cm^2) of "Anna" apple fruits in 2009 and 2010 seasons

	Storage period (days interval)					
Treatments	**1**	**2**	**3**	**4**	**5**	**Means**
			Season 2009			
Ozone + $CaCl_2$	18.40	16.16	15.63	15.31	11.95	15.49
$CaCl_2$	18.40	14.83	14.93	11.08	12.35	14.32
Ozone	18.40	16.10	14.90	11.15	10.36	14.18
Control	18.40	13.5	15.63	11.16	9.71	13.68
Means	18.40	15.15	15.27	12.18	11.09	
			Season 2010			
Ozone + $CaCl_2$	18.68	15.08	14.18	13.73	11.97	14.73
$CaCl_2$	18.68	15.38	16.22	13.50	12.03	15.16
Ozone	18.68	15.40	13.18	12.81	11.86	14.39
Control	18.68	15.53	15.83	11.45	10.23	14.34
Means	18.68	15.35	14.85	12.88	11.53	

$L.S.D_{0.05}$	A	B
Season 1	1.03	1.16
Season 2	1.16	1.129

The trend of firmness was observed to decrease with the progress of the storage period in both seasons. This result was in agreement with those previously reported by Saftener (1999) on apple and EL-Saedy (2010) on mango. The decrease in the rate of softening due to O_3+ $CaCl_2$ in 2009 may be due to inhibition of the synthesis of cell wall hydrolytic enzymes such as polygalacturonase and a-and b-galactosidase, maintaining, membrane stability and reducing the solubility of polyuronide.

B- Chemical characteristics:

1- Acidity:

The data of Table (4) represent the acidity values of "Anna" apple fruits. In the first season, no significant difference were found among treatment. In second season, there were significant differences between fruits dipped in $CaCl_2$ for 2 min and treated with ozone and stored at 4°C and those treated with ozone alone and stored at 4°C and there were non significant differences between the fruits which dipped it $CaCl_2$ for 2 min and stored at 4°C and fruits treated with ozone alone and stored at 4°C in the first season. Control fruits had the

lowest acidity in both seasons. Pirmoradian and Babalar (1995) indicated that $CaCl_2$ did not affected acidity.

With the progress of the storage period, in most dates of sampling no significant decrease in titratable acidity of apple fruits was observed. Dnake (1998) reported that acidity decreased during storage and also Argenta and Mondardo (1994) found the same trend on Gala apple.

2- SSC:

The data presented in Table (5) indicted that, fruits treated with $CaCl_2$ + O_3 and control fruits had the highest SSC compared with other treatments in the first season.

However, in the second one, fruits treated with $CaCl_2$ and control had significantly higher values compared with other treatments. Pirmoradian and Babalar (1995) on Red delicious apple and Argenta and Mondardo (1994) on Gala apples found the similar trend.

SSC decrease of the storage period that could be due to the degradation of complex in soluble compounds like starch to simple soluble compounds like sugars that are the major component of SSC content in the fruits (Chen and Mellenthin, 1981).

Table 4. Effect of ozone and calcium chloride treatments on titratable acidity (%) of "Anna" apple fruits in 2009 and 2010 seasons

	Storage period (days interval)					
Treatments	**1**	**2**	**3**	**4**	**5**	**Means**
			Season 2009			
Ozone + $CaCl_2$	0.81	0.54	0.38	0.59	0.39	0.54
$CaCl_2$	0.81	0.65	0.36	0.53	0.42	0.55
Ozone	0.81	0.77	0.30	0.44	0.39	0.55
Control	0.81	0.5	0.29	0.46	0.45	0.51
Means	0.81	0.62	0.34	0.51	0.41	
			Season 2010			
Ozone + $CaCl_2$	0.78	0.48	0.37	0.72	0.58	0.59
$CaCl_2$	0.78	0.53	0.34	0.51	0.41	0.51
Ozone	0.78	0.58	0.31	0.47	0.37	0.50
Control	0.78	0.50	0.30	0.42	0.37	0.43
Means	0.78	0.53	0.33	0.53	0.43	

$L.S.D_{0.05}$	A	B
Season 1	0.069	0.077
Season 2	0.091	0.102

Table 5. Effect of ozone and calcium chloride treatments on SSC (%) of "Anna" apple fruits in 2009 and 2010 seasons

	Storage period (days interval)					
Treatments	**1**	**2**	**3**	**4**	**5**	**Means**
			Season 2009			
Ozone + $CaCl_2$	10.93	13.33	8.80	10.66	10.66	10.87
$CaCl_2$	10.93	12.00	9.73	9.53	9.33	10.30
Ozone	10.93	12.00	9.87	8.67	9.60	10.21
Control	10.93	10.67	10.00	10.67	10.00	10.45
Means	10.93	12.00	9.60	9.88	9.90	
			Season 2010			
Ozone + $CaCl_2$	12.53	11.33	9.47	9.67	11.47	10.89
$CaCl_2$	12.53	12.0	9.73	9.13	10.70	10.81
Ozone	12.53	12.0	9.87	10.67	9.3	10.87
Control	12.53	13.33	10.00	10.00	9.00	10.97
Means	12.53	12.16	9.76	9.87	10.11	

$L.S.D_{0.05}$	A	B
Season 1	0.95	1.065
Season 2	0.82	0.93

3- Anthocyanin content:

From the tabulated data (Table 6) it was noticed that there were no significant differences for total anthocyanin content of apple peel in the first season between all treatments. In the second season, there was no significant difference among fruits dipped in $CaCl_2$ for 2 min and stored at 4°C in those treated with ozone alone and stored at 4°C. Zimmer *et al.* (1996) on Gala apple recorded that calcium treated fruits improved color which was not noticed in the present study.

4- Decay %:

Ozone treatment affected decay percentage of fruits with significant difference than control fruits (Figure 1). Also, $CaCl_2$ treatment and $CaCl_2$ plus ozone treatment had lower significant decay percentages during the storage period compared with control fruits. Shog and Chu (1999 and 2001) observed similar trend.

Conway *et al.* (2003) recorded that reduced decay in apples due to $CaCl_2$ is associated with maintaining cell wall structure by delaying or modifying chemical changes in cell wall composition.

Table 6. Effect of ozone and calcium chloride treatments on anthocyanin content (mg/ 100 g peel) of "Anna" apple fruits in 2009 and 2010 seasons

	Storage period (days interval)					
Treatments	**1**	**2**	**3**	**4**	**5**	**Means**
			Season 2009			
Ozone + $CaCl_2$	20.69	25.44	21.14	27.47	24.88	23.93
$CaCl_2$	20.69	23.05	23.28	30.08	27.62	24.95
Ozone	20.69	23.39	22.39	30.25	26.82	24.71
Control	20.69	28.78	23.05	27.97	29.28	25.95
Means	20.69	25.166	22.467	28.95	27.15	
			Season 2010			
Ozone + $CaCl_2$	29.95	29.18	26.70	28.79	29.49	28.81
$CaCl_2$	29.95	21.69	25.15	29.15	28.00	26.83
Ozone	29.95	25.75	18.90	30.20	29.21	26.80
Control	29.95	28.29	22.78	29.44	30.32	28.15
Means	29.95	26.23	23.38	29.45	29.45	

$L.S.D_{0.05}$	A	B
Season 1	2.73	3.05
Season 2	1.91	2.13

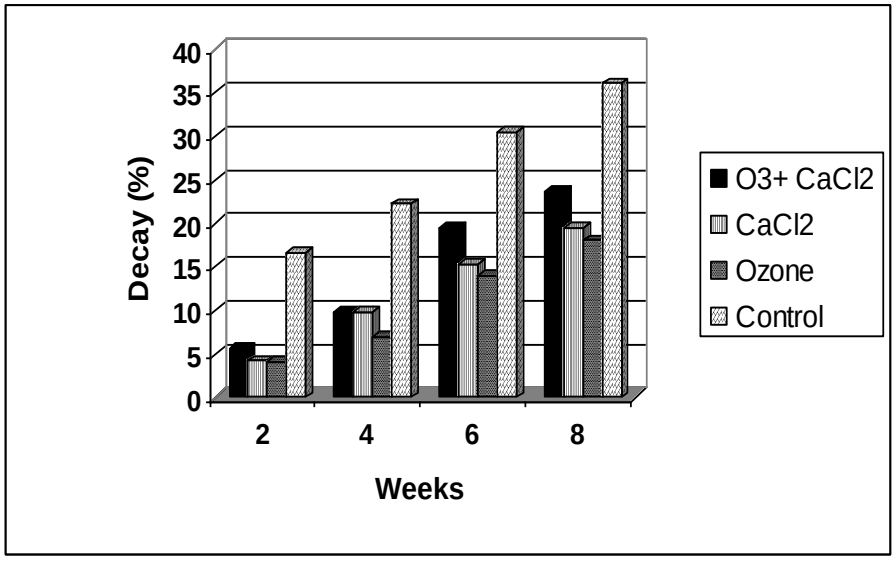

Figure 1. Effect of ozone and calcium chloride treatments on decay (%) of "Anna" apple fruits in 2009 seasons

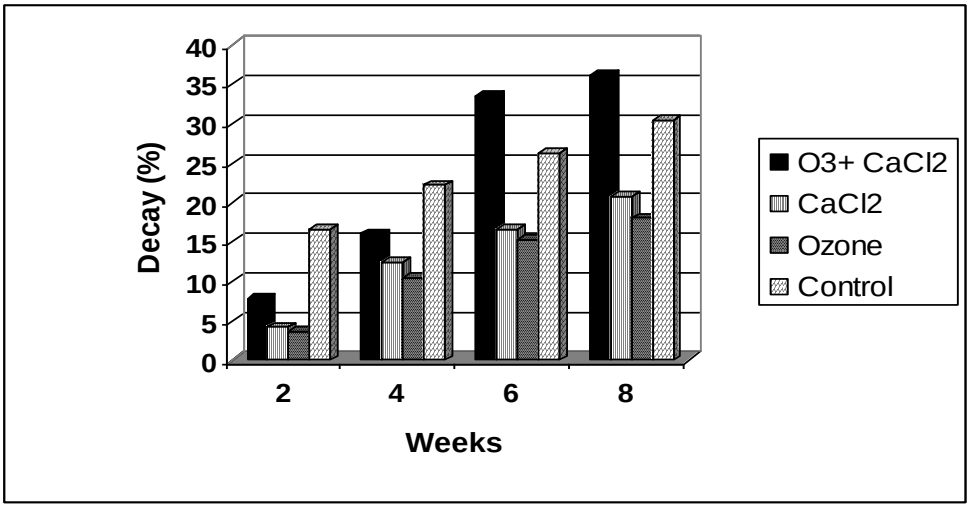

Figure 2. Effect of ozone and calcium chloride treatments on decay (%) of "Anna" apple fruits in 2010 seasons

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الملخص العربي

معاملات ما بعد الحصاد بالأوزون وكلوريد الكالسيوم للتحكم فى عفن ثمار التفاح "أنا" أثناء التخزين المبرد

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أجريت هذه الدراسة خلال موسمى 2008، 2009 على ثمار التفاح وذلك لدراسة تأثير الأوزون وكلوريد الكالسيوم على درجة 4°م لمدة 2 دقيقة وذلك لتقليل العفن وتحسين الجودة والقدرة التخزينية لثمار التفاح صنف "أنا" وذلك خلال التخزين على 4°م.

وأظهرت النتائج أن الثمار المعاملة بالأوزون أعطت أكبر نسبة فى الفقد وزن بالمقارنة بالثمار غير المعاملة (الكنترول). وقد أعطت المعاملة بكلوريد الكالسيوم أعلى قيم صلابة للثمار وزادت نسبة المواد الصلبة الذائبة بينما لم يوجد أى تأثير لهذه المعاملة على حموضة الثمار خلال فترة التخزين. كذلك فإن المعاملة بالأوزون أثرت على لون الثمار ومحتواها من الأنثوسيانين كما قللت من نسبة العفن فى الثمار.