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Pre-harvest application of calcium chloride and chitosan on fruit quality and storability of 'Early Swelling' peach during cold storage

Efeito da aplicação pré-colheita de cloreto de cálcio e quitosano na qualidade e no armazenamento de frutos de pessegueiro da variedade 'Early Swelling' durante o armazenamento a frio

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ABSTRACT
Studies related with the storage of peach fruits have received great relevance in Egypt. In this study, the effect of pre-harvest sprays of calcium chloride and chitosan, separately and in combination, on quality attributes and storability of peach fruits stored at 0±1 °C was studied. 'Early Swelling' peach trees were sprayed twice with 1% or 2% calcium chloride. The first spraying was at pea stage, while the second one was performed at 10 days before harvesting. Chitosan sprays were performed at 0.5 or 1%, alone or in combination with 1 and 2% calcium chloride, at 10 days before harvesting. Untreated trees served as control. Fruits were harvested at maturity stage, then packaged and stored at 0±1 °C and 85-90% of relative humidity. Fruit physical and chemical properties were evaluated at 7-day intervals. Results showed that pre-harvest application with 2% $CaCl_2$+1% chitosan was most effective in minimizing weight loss (%) and decay (%), as well as in maintaining maximum firmness and lengthening shelf life. Fruit color was not affected by any of the treatments, while untreated fruits and calcium chloride treatment alone, at both applied concentrations, maintained higher total soluble solids (TSS, %), total phenolic content, and lower titratable acidity percentage.

Index terms: Alternative chemicals; $CaCl_2$; quality attributes.

RESUMO
Estudos relacionados ao armazenamento de pêssego tem sido considerado de grande relevância no Egito. Neste estudo, o efeito da pulverização pré-colheita com cloereto de calico e quitosano, isoladamente ou em conjunto, na qualidade e na capacidade de armazenamento de frutos de pessegueiro armazenados a 0±1 °C foi avaliada. Frutos da variedade 'Early Swelling' foram tratados com 1,0 ou 2,0% de cloreto de cálcio. O primeiro tratamento foi aplicado num estado precoce de formação do fruto enquanto o segundo foi aplicado 10 dias antes da colheita. Quitosano foi aplicado nas concentrações de 0,5 ou 1,0%, isoladamente ou em combinação com 1,0% ou 2,0% de cloreto de cálcio também 10 dias antes da colheita. Árvores não tratadas serviram como controle. Os frutos foram colhidos quando maduros, empacotados e mantidos a 0±1 °C e sob condições de humidade relativa de 85-90%. Propriedades físicas e químicas dos frutos foram avaliadas em intervalos de 7 dias. Os resultados mostraram que a aplicação de 2% $CaCl_2$ + 1% quitosano foi a mais eficaz em minimizar a perda de peso (%) e a senescência (%), bem como na manutenção da rigidez e aumento do período de vida em prateleira. A cor dos frutos não foi afectada por nenhum dos tratamentos enquanto frutos não tratados e cloreto de cálcio isoladamente, em ambas as concentrações, mantiveram um elevado teor de sólidos solúveis totais (TSS, %), elevado teor de fenóis e reduzida percentagem de acidez de titulação.

Termos para indexação: Químicos alternativos; $CaCl_2$; atributos qualitativos.

INTRODUCTION

Peach (*Prunus persica* (L.) Batsch) belongs to the family Rosaceae and is one of the most popular fruits in the world because of its high nutrient value and pleasant flavor. Peach is considered as one of the most important deciduous fruits in Egypt, where it has great success and is widespread in the newly reclaimed areas. The storage of peach fruits is problematic due to a short post-harvest life, as fruits quickly pass from ideal maturity to over ripening phase, readily lose water and shrivel, can be attacked and destroyed by fruit rotting organisms and become unmarketable as a result of the internal breakdown. An increased concern among consumers about food safety and the potentially harmful health effects of chemical residues encouraged research to find safe alternative chemicals which can maintain the marketable quality throughout prolonged storage period of fruits, including peach.

In this sense, pre- and post-harvest application of calcium may delay senescence in fruits with no detrimental effect on consumer acceptance. Calcium is a key plant nutrient that has a significant role in cell functions, including reducing softening and senescence of fruits (Barker; Pilbeam, 2015), and it is also considered the most important mineral element determining fruit quality (El-Badawy, 2012). The role of calcium in stabilizing cellular membranes and delaying senescence in horticultural crops, (Poovaiah; Glenn; Reddy, 1988), as well as its contribution to the linkages between pectic substances within the cell-wall are well known. Pre-harvest calcium treatments to increase calcium content of the cell wall were effective in delaying senescence, resulting in firmer and higher fruit quality (Serrano et al., 2004). The mobility of calcium in trees is low, and the root uptake from fertilized soils is poorly effective in increasing the calcium content in fruits. In this context, Ghani, Awang and Sijam (2011), reported that the direct application of liquid source of calcium on leaves and fruits may offer an alternative solution. Calcium may have also a potential use as an alternative method in integrated disease management (Biggs; Hogmire; Collins, 2000).

Chitosan is an N-acetylated derivative of the polysaccharide chitin. It is a natural polymer with a polycationic nature, which has numerous applications in agriculture (e.g., as soil modifier, films, fungicide, elicitor) and agroindustry, as well as in cosmetics, biomedicine, environmental protection, wastewater management (Deepmala et al., 2014). Chitosan is edible and safe for humans (Rhoades; Roller, 2000), and it is used in human medicine, advised in slimming diets (Maezaki et al., 1993). Due to these properties, chitosan could be applied near harvest time. Many studies have shown the high potential of chitosan for preserving fresh fruits and vegetables. Pre-harvest spraying with chitosan is highly feasible and has a beneficial effect on fruit quality attributes (Reddy et al., 2000). Chitosan has been also considered as a valid alternative to synthetic fungicides (El Ghaouth, 1997). Romanazzi (2010) reported that chitosan has a double mechanism of actions, i.e., it inhibits the development of decay-causing fungi, and induces resistance responses of host tissues. It is considered as an ideal preservative coating because it has a disease suppressive effect, resulting from both physical and biochemical mechanisms. The physical properties of the polymer allow it to produce a film on the surface of treated fruit (Du; Gemma; Iwahori, 1998), and has also the potential to prolong the storage life of many fruits, such as peach, Japanese pear, kiwifruit, strawberry and sweet cherry (Du; Gemma; Iwahori, 1997; EL Ghaouth; Ponnampalam; Boulet, 1991; Romanazzi; Nigro; Ippolito, 2003). Chitosan also induces chitinase activity, and elicits phytoalexins and defense barriers in the host tissues (El Ghaouth et al.,1992), as well as the defense responses in several plant systems.

Calcium has a general application in either pre-harvest or post-harvest treatments, while reports on pre-harvest application of chitosan are limited (El-Badawy, 2012; Reddy et al., 2000). In this study, the effect of pre-harvest application of calcium chloride, chitosan and their combination on storability and quality attributes of peach fruits, cv Early Swelling, has been investigated.

MATERIAL AND METHODS

The present study was conducted through two successive seasons (2014 and 2015) on peach (*Prunus persica*), cv Early Swelling. Trees were grown in a private orchard at El- Khatatba City, in El-Menufiya Governorate, Egypt, in a sandy soil. Trees were 7 years old, budded on 'Nemaguard' peach rootstock, cultivated at 4×4 m distance, open-vase shape trained, drip irrigated, and received normal cultural practices adopted in the orchard.

Experimental design

Each treatment consisted of three trees in a randomized complete block design where a single tree represented the experimental unit. The experiment consisted of a total of nine treatments (including the control) for each season. The treatments were: (i) 1 or 2% calcium chloride, with two sprayings of trees, the 1st at "pea stage", the 2nd 10 days before harvesting, (ii) 0.5 or 1% chitosan, alone or in combination with 1 or 2% calcium chloride, with only one spraying, 10 days before harvesting. A set of trees were left untreated and served as control. Trees were sprayed with hand-sprayer, till fruits were completely wet to run off. Samples of fruits at full maturation from each treatment were hand-harvested during the first two weeks of May. In the laboratory of the Faculty of Agriculture, Cairo University, fruits of each treatment were then sorted and selected for uniformity of weight, size, and absence of mechanical damage or visible pathological infections. The selected fruits were packed in carton boxes (2 kg capacity). Each treatment was triplicated. Fruits were then stored for 35 days at 0±1 °C with a relative humidity (RH) of 85-90%. Fruit physical and chemical quality attributes were periodically assessed at intervals of 7 days throughout all the storage period.

Quality assessments of fruits by physical characteristics

The following physical characteristics were evaluated:

Weight loss: Samples of each treatment were weighed at weekly intervals until the end of experiment. Weight loss (%) was calculated as follows:

Fruit weight loss (%) = [(initial weight – weight at sampling date / initial weight)] x 100.

Fruit decay percentage: evaluated by type, as skin appearance, shriveling, chilling injury and pathogenic rots. In every inspection date, decayed fruits were discarded and the relative amount expressed as decay percentage.

Fruit firmness (kg/cm^2): fruits from each replicate were taken at weekly intervals to determine the changes in fruit firmness using stationary firmness tester. Fruit firmness was measured in two opposite sides of the equatorial fruit zone after removing the peel, and expressed in kg/cm^2.

Shelf life (days): after 35 days of cold storage, fruit samples of each treatment were placed at ambient temperature, and shelf life was determined as a number of days of which fruits maintained acceptable eating quality and appearance.

Color assessment: hue angle (h^o) values of fruits were assessed using a Minolta colorimeter CR-40 (Konica Minolta Sensing Inc, Sakai, Japan).

Quality assessments of fruits by chemical characteristics

The following chemical characteristics were evaluated:

Fruit total soluble solids: total soluble solids (TSS) were measured using digital pocket refractometer (model PAL 1, ATAGO™, Tokyo Tech.) and expressed as percentage.

Fruit titratable acidity: titratable acidity (TA) was measured in the juice of fruit sample for each replicate by titration against calibrated 0.1N NaOH solution in the presence of phenolphthalein as an indicator. Acidity was calculated as percentage of malic acid according to A.O.A.C. (*https://archive.org/details/gov.law.aoac.methods.1.1990*).

Total phenols: total phenolics were analyzed spectrophotometrically using the method described by Swain and Hillis (1959). Results were expressed as g of gallic acid /100 g fresh weight (FW).

Statistical analysis

The experiment was conducted using a completely randomized design with three replicates. Data from the analytical determinations were subjected to analysis of variance (ANOVA). Mean comparisons were performed by Duncan's Multiple range test at 5% level (Snedecor; Cochran, 1982).

RESULTS AND DISCUSSION

Weight loss (%)

The percentage of weight loss increased all through the storage period (Table 1). With regard to the effect of the tested pre-harvest treatments, considering mean values at the end of the analyzed period of time, in both seasons the highest significant weight loss percentage was obtained by untreated fruits (control), while treatments with chitosan and $CaCl_2$ (at different concentrations, depending on the season) recorded the lowest significant weight loss. Other treatments did not show clear differences among them, especially in the first season, although mean values resulted always significantly higher than the control fruits. The lowest fruit weight loss (%) was obtained in fruits treated with 1% $CaCl_2$ + 1% chitosan after 7 days of storage.

Weight loss of fresh fruits is mainly due to water loss as a result of evaporation and transpiration, while the amount of dry matter is lost by respiration. Results of this study on weight loss reduction are in accordance with what observed on 'Florida prince' peach (El-Badawy, 2012) and plums (Kirmani et al., 2013), where a progressive and significant increase in physiological loss in weight (PLW) of fruits with the increase in storage duration was recorded. However, the increase in PLW of calcium chloride-treated fruits was relatively slower and, consequently, these fruits exhibited significantly lower overall losses as compared to other treatments and control fruits. The reduction in weight loss in $CaCl_2$ treated fruits might be due to the maintenance of fruit firmness and tissue rigidity by decreasing the enzyme activity responsible for disintegration of cellular structure, which decreases the gaseous exchange (Levy; Poovaiah, 1979).

In this study, chitosan proved to be an effective coating reducing weight loss, alone as reported on longan fruit (Jiang; Li, 2001), or in combination with calcium chloride on peach (El-Badawy, 2012). Chitosan coatings act as barriers, thereby restricting water transfer and protecting fruit skin from mechanical injuries, as well as sealing small wounds and thus delaying dehydration (Ribeiro et al., 2007).

Table 1: Effect of some pre-harvest treatments on weight loss (%) of 'Early Swelling' peach fruits stored at 0±1 ºC during 2014 and 2015 seasons. Different letters indicate significantly different values by ANOVA followed by Duncan test at P≤0.05 (small letters refer to values recorded in each season, capital letters refer to mean values).

Treatment	2014 season						2015 season					
	Storage period (days)					Mean	Storage period (days)					Mean
	7	14	21	28	35		7	14	21	28	35	
Control	8.67nop	13.70hij	18.72def	22.44b	26.69a	18.04A	5.33w	10.01qrs	16.47i-l	25.11b	33.73a	18.13A
1% $CaCl_2$	7.00pqr	9.15m-p	13.98hi	17.23ef	21.32bc	13.74B	7.00t-w	11.19o-r	16.51i-l	20.48efg	24.50bc	15.94B
2% $CaCl_2$	7.00pqr	11.38j-m	13.80hi	16.54fg	20.39bcd	13.82B	6.00vw	9.27q-t	14.82klm	19.11f-i	24.13bc	14.67BC
0.5% Chitosan	5.00rs	9.20m-p	13.34h-k	17.75ef	22.45b	13.55B	5.67vw	10.48p-s	14.64klm	18.12g-j	23.41bcd	14.46C
1% Chitosan	5.67qrs	9.12m-p	14.23hi	18.11def	19.60cde	13.35B	5.00w	8.94q-u	14.36k-n	19.50e-h	22.02cde	13.96CD
1% $CaCl_2$ + 0.5% chitosan	4.67rs	9.74mno	12.26i-l	17.32ef	21.06bc	13.01BC	6.00vw	7.63s-w	13.05m-p	16.00jkl	20.48efg	12.63DE
1% $CaCl_2$ + 1% chitosan	4.33s	6.78pqr	10.82lmn	16.78fg	21.13bc	11.97CD	6.00vw	8.46r-v	14.38k-n	16.80h-k	18.95f-i	12.92D
2% $CaCl_2$+ 0.5% chitosan	5.33qrs	6.85pqr	13.36hijk	17.04f	22.26b	12.97BC	6.33uvw	8.36r-v	13.83l-o	16.59i-l	21.14def	13.25CD
2% $CaCl_2$ +1% chitosan	5.33qrs	7.52opq	11.09klm	14.69gh	18.45def	11.42D	5.00w	7.82s-w	11.54n-q	14.25k-n	18.34f-j	11.39E
Mean	5.89E	9.27D	13.51C	17.54B	21.48A		5.82E	9.13D	14.40 C	18.44B	22.97A	

Decay (%)

In both seasons, decay percentages had progressive increment as the storage period increased (Table 2). Decay incidence started at the second week of storage, and gradually increased up to day 35 of storage. Pre-harvest treatments of 2% $CaCl_2$+1% chitosan, 1% $CaCl_2$+1% chitosan, and 2% $CaCl_2$+0.5 % chitosan, in this order, recorded the lowest significant decay percentages. On the whole, the lowest fruit decay (%) was obtained when the fruits were treated with 2% $CaCl_2$+1% chitosan and stored for 14 days; on the contrary, the highest significant decay percentage was gained following 35 days of storage period of untreated fruits (control).

Results here may be attributed to the role of calcium ions in reducing fruit softening by strengthening the cell walls, as well as of chitosan covering cuticle and lenticels. Moreover, their high antifungal activity reduces respiration, ripening processes and infection during storage. Results of this study are in agreement with previous reports which indicated that calcium chloride spray reduces physiological disorders of fruits and increases their resistance to infection than untreated ones (Kirmani et al., 2013).

Chitosan modifies gas exchange of fruit with the atmosphere, and its internal gas composition by producing a film coating on the surface. The suppressive effect on decay by chitosan can be in part attributed to delaying the senescence process. Chitosan, as a natural polycation compound, c limit fungal decay of fruits by its direct antifungal activity (Bautista-Banos et al., 2006), induction of host resistance to pathogens (Trotel-Aziz et al., 2006), and its self-polymerisation that covers the fruit surface (Gonzalez-Aguilar, 2009). It offers a protection against deterioration by slowing decay and ripening; moreover, its protective effect is very high against infection (Reddy et al., 2000). For all these properties, chitosan has a potential to prolong storage life and control decay of fruits. As for the combination of chitosan and calcium chloride, its potential as fungal decay inhibitors has been reported also by Muñoz et al. (2008).

The results of pre-harvest chitosan efficiency in reducing decay percentages are in accordance with those obtained by Reddy et al. (2000) that evidenced the protection effect against infection due to pre-harvest chitosan spray of *Botrytis cinerea*. Similar findings have been also reported on grape (Meng et al., 2008), citrus (Chien; Sheu; Yang, 2007), 'Florida prince' peach (El-Badawy, 2012).

Table 2: Effect of some pre-harvest treatments on decay (%) of 'Early Swelling' peach fruits stored at 0±1 °C during 2014 and 2015 seasons. Different letters indicate significantly different values by ANOVA followed by Duncan test at P≤0.05 (small letters refer to values recorded in each season, capital letters refer to mean values).

Treatment	2014 season						2015 season					
	Storage period (days)					Mean	Storage period (days)					Mean
	7	14	21	28	35		7	14	21	28	35	
Control	0.00w	8.33r	18.67m	31.84fg	51.66a	22.10A	0.00r	9.14o	17.20ij	31.73f	52.27a	22.07A
1% $CaCl_2$	0.00w	3.54t	13.56n	23.66h	41.66b	16.48B	0.00r	4.43pq	13.86klm	22.35g	42.36b	16.60B
2% $CaCl_2$	0.00w	5.32s	11.60o	21.75ij	39.66c	15.67C	0.00r	4.19pq	13.25klm	22.11g	41.53b	16.22BC
0.5% Chitosan	0.00w	3.14tu	11.34o	22.74hi	37.00de	14.84D	0.00r	4.11pq	14.21kl	21.31gh	40.31bc	15.99BCD
1% Chitosan	0.00w	3.56t	10.75op	21.54ijk	36.33e	14.44D	0.00r	5.23p	14.94jk	14.15kl	40.25bc	14.91CDE
1% $CaCl_2$ + 0.5% chitosan	0.00w	2.67tuv	10.44opq	21.33ijk	37.88d	14.46D	0.00r	3.24pq	12.57k-n	19.33hi	38.47cd	14.72DEF
1% $CaCl_2$ + 1% chitosan	0.00w	1.43vw	9.32pqr	20.44jkl	32.33f	12.70EF	0.00r	2.15qr	11.35mno	17.44ij	36.44de	13.48FG
2% $CaCl_2$ + 0.5% chitosan	0.00w	1.87uv	10.11opq	20.21kl	33.33f	13.10E	0.00r	3.77pq	12.26lmn	18.83hi	37.56d	14.48EF
2% $CaCl_2$ + 1% chitosan	0.00w	1.21vw	9.23qr	19.33lm	30.33g	12.02F	0.00r	2.13qr	10.23no	17.12ij	34.15ef	12.73G
Mean	0.00E	3.45D	11.67C	22.54B	37.80A		0.00E	4.27D	13.32C	20.48B	40.37A	

Fruit firmness (kg/cm²)

Fruit firmness is one of the most crucial factors in determining the post-harvest quality and physiology of fruits (Kirmani et al., 2013). Data presented in Table 3 show that fruit firmness has significantly decreased as the storage period increased, reaching its lowest values at the end of storage period, regardless of the pre-harvest treatments. All treatments recorded significantly high firmness than control fruits; however, in both seasons, 2% $CaCal_2$+1% chitosan showed, at each period, with few exceptions, the highest values of peach firmness. It should be noted that all the treatments had an effect in preserving fruit firmness as, at the end of the storage period, the lowest firmness value was obtained by the untreated fruits.

Fruit softening is caused either by breakdown of insoluble proto-pectins into soluble pectin or by hydrolysis of starch (Matto et al., 1975), or by increased membrane permeability caused by cellular disintegration (Oogaki; Wang; Gemma*,* 1990). The loss of pectic substances in the middle lamellae of the cell wall is perhaps the key step in ripening process that leads to the loss of cell integrity or firmness (Solomes; Latics, 1973). The desired effect of calcium in maintaining fruit firmness may be due to the calcium binding to free carboxyl groups of polygalacturonate polymer, stabilizing and strengthening the cell wall, which in turn may strengthen the tissue thus becoming more resistant to hydrolytic enzyme activity, where calcium inhibits the polygalacturonase activity in cell walls (Buescher; Hobson, 1982). Results here regarding the role of $CaCl_2$ in the reduction of fruit softening are in correlation with those obtained on plums (Kirmani et al., 2013) and strawberry (Muñoz et al., 2008). All these reports evidenced a similar reduction in the firmness loss following the pre-harvest application of $CaCl_2$.

Pre-harvest beneficial effect of chitosan on fruit firmness could be due to the formation of a chitosan film on fruit which can act as a barrier for O_2 uptake, thereby slowing the metabolic activity, and consequently the ripening process (Reddy et al., 2000). Higher firmness during the storage period of 'Early swelling' peach fruits following pre-harvest chitosan treatments are in harmony with those observed in previous studies on peach fruits (Brar; Simnani; Kaundal, 1997; Li; Yu, 2000), as they reported that chitosan treatments delayed the loss of firmness. In addition, Reddy et al. (2000) reported the beneficial effect of pre-harvest chitosan sprays on flesh firmness

Table 3: Effect of some pre-harvest treatments on firmness (kg/cm²) of 'Early Swelling' peach fruits stored at 0±1 °C during 2014 and 2015 seasons. Different letters indicate significantly different values by ANOVA followed by Duncan test at P≤0.05 (small letters refer to values recorded in each season, capital letters refer to mean values).

Treatment	2014 season							2015 season						
	Storage period (days)						Mean	Storage period (days)						Mean
	0	7	14	21	28	35		0	7	14	21	28	35	
Control	1.10b-h	1.03f-l	0.87o-v	0.72x	0.59y	0.49y	0.80C	1.15abc	1.00d-j	0.89i-q	0.84l-u	0.75r-v	0.57w	0.87B
1% $CaCl_2$	1.23a	1.07c-j	0.96j-q	0.91l-t	0.86p-w	0.82s-x	0.98B	1.10a-e	1.03b-i	0.95f-m	0.90i-q	0.84m-u	0.79p-v	0.94A
2% $CaCl_2$	1.20ab	1.04e-k	1.02g-m	0.97i-p	0.90n-t	0.75wx	0.98B	1.16ab	1.10a-e	1.00e-k	0.88j-s	0.79o-v	0.69vw	0.94A
0.5% Chitosan	1.18abc	1.09b-i	1.00h-n	0.93k-s	0.85r-w	0.71x	0.96B	1.18a	1.08a-f	0.94f-o	0.85k-t	0.78q-v	0.71uv	0.92AB
1% Chitosan	1.15a-e	1.06d-j	1.01h-n	0.94k-r	0.88o-u	0.80t-x	0.97B	1.14a-d	1.05a-h	0.94f-n	0.89i-r	0.80n-v	0.72tuv	0.92AB
1% $CaCl_2$ + 0.5% chitosan	1.20ab	1.14a-f	1.06d-j	0.98i-o	0.91m-t	0.77u-x	1.01AB	1.13a-e	1.05a-g	0.98e-l	0.91h-q	0.84m-u	0.75s-v	0.94A
1% $CaCl_2$ + 1% chitosan	1.23a	1.13a-g	1.03f-k	0.94k-r	0.87o-v	0.84r-w	1.01AB	1.16ab	1.10a-e	1.01c-j	0.91i-q	0.81n-v	0.72tuv	0.95A
2% $CaCl_2$ + 0.5% chitosan	1.24a	1.18abc	1.06d-j	0.98i-p	0.86q-w	0.76vwx	1.01AB	1.17ab	1.06a-g	1.00d-j	0.93g-p	0.83m-v	0.71uv	0.95A
2% $CaCl_2$ + 1% chitosan	1.25a	1.16a-d	1.08c-i	1.02g-m	0.94k-r	0.87o-v	1.05A	1.18a	1.10a-e	1.01c-j	0.90i-q	0.82m-v	0.74tuv	0.96A
Mean	1.20AB	1.10B	1.01C	0.93D	0.85E	0.76F		1.15A	1.06B	0.97C	0.89D	0.81E	0.71F	

of strawberries; moreover, they found that fruit firmness correlated with the increasing of chitosan concentration.

Hue angle (h°)

Hue angle (h°) values showed a significant decline along the storage periods, irrespective of the applied treatments (Table 4). Indeed, all the treatments exerted a gradual decrease of h° along the storage periods, with the highest mean value in the first season (2014) recorded by 0.5% chitosan. In the second season (2015), no significant differences between treatments were recorded. At the end of the storage period (35 days), highest h° values were shown with fruits treated with 0.5 and 1% chitosan, in the first and second season, respectively. Muñoz et al. (2008) reported that the h° of un-treated strawberry fruits began to decrease after the second day of storage and, at the end of the storage period (7 days) the decline was 32%. Moreover, calcium and chitosan treated fruits failed to show significant change during the storage period. They suggested that the control of moisture loss by chitosan coating contributes to minimizing external color changes in fully ripe strawberries. Wang et al. (2007) reported that post-harvest treatments with 'Chito-care' significantly reduced the color change rate of mango fruits during their storage, compared to untreated fruits.

Shelf life (days)

The application of chitosan or $CaCl_2$ at different concentrations had a high effect on post-harvest shelf-life of 'Early swelling' peach fruits (Table 5). In both seasons, maximum effect was achieved by the treatment with 2% $CaCl_2$+1% chitosan, and the recorded value (4.33) was significantly greater than all other treatments in the second season (2015).

Calcium chloride has been extensively used for improving fruit quality, delaying deterioration in storage and thereby increasing the shelf life of various fruits (Kirmani et al., 2013). For instance, El-Badawy (2012), Mahajan and Sharma (2000) on peaches, and Kirmani et al. (2013) on plums, reported that shelf life was longer in pre-harvest Ca-treated fruits. Calcium treatment has been shown to decrease respiration, reduce ethylene production and slow down the onset of ripening in apple (Ferguson, 1984) and avocado (Wills; Sirivatanapa, 1988).

Chitosan is a well-known natural and edible fruit coating material, because of its potential to

Table 4: Effect of some pre-harvest treatments on hue angle (h°) of 'Early Swelling' peach fruits stored at 0±1 °C during 2014 and 2015 seasons. Different letters indicate significantly different values by ANOVA followed by Duncan test at P≤0.05 (small letters refer to values recorded in each season, capital letters refer to mean values).

Treatment	2014 season							2015 season						
	Storage period (days)						Mean	Storage period (days)						Mean
	0	7	14	21	28	35		0	7	14	21	28	35	
Control	59.88a-e	56.64a-f	51.26g-l	48.52h-n	46.09m-q	42.56pq	50.83C	66.25abc	62.28a-e	58.51e-h	54.04hij	48.61j-o	44.06no	55.63A
1% $CaCl_2$	61.21ab	55.47d-g	51.48g-k	49.54h-m	47.70i-o	43.93n-q	51.55AB	67.29ab	63.15a-e	59.21e-h	55.24f-i	50.99i-l	45.62k-o	56.92A
2% $CaCl_2$	60.55a-d	55.67c-g	52.62f-j	49.98h-m	47.44k-p	42.89opq	51.53AB	66.31abc	62.69a-e	59.49d-h	55.23f-i	50.86i-l	45.08k-o	56.61A
0.5% Chitosan	59.04a-e	56.19b-g	52.60f-j	48.98h-n	47.59j-p	45.51m-q	51.65A	65.52a-d	61.79a-e	58.87e-h	55.25f-i	50.11i-n	45.18k-o	56.12A
1% Chitosan	59.22a-e	55.07efg	53.14fgh	49.05h-m	46.36l-q	42.76opq	50.93C	66.48ab	61.17b-f	57.86e-h	54.59g-j	50.77i-l	46.23k-o	56.18A
1% $CaCl_2$ + 0.5% chitosan	61.42a	56.64a-f	51.51g-k	48.38h-n	45.21m-q	43.09opq	51.04BC	66.03abc	61.49b-e	58.54e-h	54.87ghi	49.51i-n	45.40k-o	55.97A
1% $CaCl_2$ + 1% chitosan	60.40a-d	56.12c-g	52.65f-j	48.80h-n	46.07m-q	42.85opq	51.15ABC	67.79a	61.48b-e	58.04e-h	54.60g-j	50.24i-m	42.71o	55.81A
2% $CaCl_2$ + 0.5% chitosan	59.15a-e	56.79a-f	52.77f-i	49.89h-m	45.47m-q	42.17q	51.04BC	67.75a	61.74a-e	57.86e-h	55.07f-i	50.13i-n	44.91l-o	56.24A
2% $CaCl_2$ + 1% chitosan	60.66abc	56.26b-g	53.13fgh	49.13h-m	45.62m-q	43.08opq	51.31ABC	66.68ab	61.83a-e	60.34c-g	54.74g-j	51.13ijk	44.33mno	56.51A
Mean	60.17A	56.09B	52.35C	49.14D	46.39E	43.20F		66.68A	61.96B	58.75C	54.85D	50.26E	44.84F	

inhibit fungal growth and extend the shelf life of fruits (Han et al., 2004). These beneficial effects may due to the reduction of fruit respiration metabolism (El Ghaouth; Ponnampalam; Boulet, 1991; Hagenmaier, 2005), respiration rate and creation of a modified atmosphere within fruits, thus leading to an increase of CO_2 concentrations inside fruit (Sivakumar et al., 2005) and reducing color changes of skin and flesh and increased the shelf life of fruits (Maftoonazad; Ramaswamy, 2005). In addition, chitosan acts as an inhibitor of various enzymes, leading to delay fruit senescence (Dutta et al., 2009). Here, the positive effect of chitosan on shelf life extension of peach fruits was consistent with what was reported for various other fruits, such as papaya (Chutichudet; Prasit, 2014), longan (Jiang; Li, 2001), and fresh-cut strawberry (Campaniello et al., 2008). Moreover, chitosan and calcium chloride, separately and in combination, were an effective preservative for the increase of shelf life of peach (El-Badawy, 2012), and mango (Chauhan; Agrawal; Raje, 2014).

Table 5: Effect of some pre-harvest treatments on shelf life (days) of 'Early Swelling' peach fruits after 35 days of cold storage at 0±1 °C during 2014 and 2015 seasons. Different letters indicate significantly different values by ANOVA followed by Duncan test at P≤0.05.

Treatments	Shelf life (days)	
	2014 season	2015 season
Control	2.00D	2.33 C
1% $CaCl_2$	2.33CD	3.00 BC
2% $CaCl_2$	2.33CD	3.33 BC
0.5% Chitosan	2.67BCD	3.33 BC
1% Chitosan	2.67BCD	3.67 B
1% $CaCl_2$ + 0.5% chitosan	3.00ABC	3.67 B
1% $CaCl_2$ + 1% chitosan	3.33AB	3.67 B
2% $CaCl_2$ + 0.5% chitosan	3.67A	3.67 B
2% $CaCl_2$ + 1% chitosan	3.67A	4.33 A

Total soluble solid (TSS)

TSS percentage showed a steady increase in firmness, commensurate with the advance in the storage period (Table 6), this was regardless of treatments. In both seasons, untreated fruits recorded

Table 6: Effect of some pre-harvest treatments on total soluble solid (%) of 'Early Swelling' peach fruits stored at 0±1 °C during 2014 and 2015 seasons. Different letters indicate significantly different values by ANOVA followed by Duncan test at P≤0.05 (small letters refer to values recorded in each season, capital letters refer to mean values).

Treatment	2014 season							2015 season						
	Storage period (days)						Mean	Storage period (days)						Mean
	0	7	14	21	28	35		0	7	14	21	28	35	
Control	12.63p-u	13.17n-t	14.10h-q	15.53b-i	16.57abc	17.73a	14.96A	13.20uvw	14.47o-t	15.47j-m	16.67f-i	17.50cde	18.67a	15.99A
1% $CaCl_2$	12.93o-t	13.53k-s	14.30g-o	15.00d-l	15.87b-f	16.70ab	14.72AB	12.70wx	13.67tuv	15.13k-o	16.20hij	17.27def	18.77a	15.62AB
2% $CaCl_2$	12.77o-u	13.33m-t	14.03i-q	15.07c-k	16.00b-f	16.73ab	14.66AB	13.13vw	14.00stu	14.97m-q	15.90i-l	17.10d-g	18.23abc	15.56B
0.5% Chitosan	12.77o-u	13.60k-r	14.00i-r	14.83e-m	15.77b-g	16.40a-d	14.56AB	12.93vwx	14.00stu	15.10l-o	16.23hij	17.20def	18.13abc	15.60B
1% Chitosan	12.80o-u	13.30m-t	13.90j-r	14.60f-n	15.60b-h	16.70ab	14.48AB	12.17xy	14.03st	14.90m-r	16.20hij	17.23def	18.43ab	15.49B
1% $CaCl_2$ + 0.5% chitosan	12.00stu	13.47l-s	14.10h-q	14.77e-m	15.30b-j	15.87b-f	14.25BC	13.07vw	13.97stu	15.00m-p	15.93ijk	16.80e-h	17.73bcd	15.42B
1% $CaCl_2$ + 1% chitosan	12.57q-u	13.07n-t	13.57k-r	14.27g-o	15.40b-j	16.50a-d	14.23BC	11.77y	12.93vwx	14.10rst	15.30k-n	17.10d-g	18.63a	14.97CD
2% $CaCl_2$ + 0.5% chitosan	11.87tu	12.47r-u	13.70k-r	14.83e-m	15.47b-i	16.23b-e	14.09BC	12.83wx	14.17q-t	14.63n-s	15.10l-o	16.67f-i	18.70a	15.35BC
2% $CaCl_2$ + 1% chitosan	11.40u	13.07n-t	13.57k-r	14.17h-p	14.83e-m	15.90b-f	13.82C	12.17xy	12.90vwx	14.20p-t	15.00m-p	16.30ghi	17.43c-f	14.67D
Mean	12.41F	13.22E	13.92D	14.79C	15.64B	16.53A		12.66F	13.79E	14.83D	15.84C	17.02B	18.30A	

the highest TSS mean percentage, compared to all the other treatments, while the lowest mean percentage was obtained with the treatment of 2% $CaCl_2$+1% chitosan.

Increased TSS percentages throughout the storage period are presumably due to increased activity of enzymes responsible for starch hydrolysis to soluble sugars and can be caused by the decline in the amount of carbohydrates, pectines, partial hydrolysis of protein and decomposition of glycosides into subunits during respiration (Abbasi et al., 2009). Wongmetha and Ke (2012) reported that chitosan had no effect on TSS of mango fruits. A similar response was also showed in strawberry (Muñoz et al., 2008).

Results of calcium as a pre-harvest treatment are in harmony with those mentioned by Montanaro et al. (2006) on kiwifruit, Bhat et al. (2012) on pear and El-Badawy (2012) on 'Florida Prince' peach.

Titratable acid (TA)

TA content decreased with the prolonging of the storage period (Table 7). As for treatments, in both seasons highest means of TA contents were obtained with all the combinations of $CaCl_2$ and chitosan, while the lowest TA contents were recorded with 2% and 1% $CaCl_2$ treatments in the first and second season, respectively. These results can be due to the reduction in metabolic changes of organic acids into carbon dioxide and water, as a result of reducing respiration rate, and therefore maintaining higher rates of acids. The results reported in this study are in agreement with those found by Goutam, Dhaliwal and Mahajan (2010) on guava, and El-Shazly et al. (2013) on peach.

Total phenolic content (%)

It is worthy to note that total phenolic content increased with the advance in the storage period (Table 8). This was observed in both the studied seasons, irrespective of the treatments. As for tested treatments, in both seasons the highest total phenolic content was obtained by untreated fruits followed by 1% $CaCl_2$, while the lowest total phenolic content was recorded by the treatment with 2% $CaCl_2$+1% chitosan.

As for the effect of chitosan treatments, it has been reported that pre-harvest chitosan spray enhanced total phenol content of table grapes during further storage at 0 °C (Meng et al., 2008).

Table 7: Effect of some pre-harvest treatments on titratable acidity (%) of 'Early Swelling' peach fruits stored at 0±1 ºC during 2014 and 2015 seasons. Different letters indicate significantly different values by ANOVA followed by Duncan test at P≤0.05 (small letters refer to values recorded in each season, capital letters refer to mean values).

Treatment	2014 season						Mean	2015 season						Mean
	Storage period (days)							Storage period (days)						
	0	7	14	21	28	35		0	7	14	21	28	35	
Control	0.360a-g	0.330d-l	0.290h-q	0.283i-r	0.260 n-s	0.223rs	0.291C	0.360ef	0.330j	0.320k	0.293o	0.270r	0.260st	0.306E
1% $CaCl_2$	0.380a-e	0.333c-k	0.306g-p	0.286h-q	0.250o-s	0.210s	0.294C	0.370d	0.353g	0.323k	0.293o	0.273r	0.243v	0.309D
2% $CaCl_2$	0.380a-e	0.316e-n	0.306g-p	0.266l-s	0.243p-s	0.210s	0.287C	0.390b	0.347h	0.330j	0.307m	0.280q	0.250u	0.317C
0.5% Chitosan	0.380a-e	0.326d-m	0.303g-p	0.286h-q	0.266l-s	0.246p-s	0.302BC	0.360ef	0.347h	0.323k	0.300n	0.273r	0.260st	0.311D
1% Chitosan	0.380a-e	0.350a-h	0.323d-n	0.280j-r	0.260n-s	0.236qrs	0.305BC	0.390b	0.347h	0.330j	0.307m	0.280q	0.250u	0.317C
1% $CaCl_2$ + 0.5% chitosan	0.380a-e	0.356a-g	0.330d-l	0.326d-m	0.286h-q	0.263m-s	0.324AB	0.380c	0.363e	0.330j	0.310lm	0.287p	0.270r	0.323B
1% $CaCl_2$ + 1% chitosan	0.393abc	0.380a-e	0.356a-g	0.333c-k	0.313f-o	0.283i-r	0.343A	0.380c	0.357fg	0.340i	0.313l	0.290op	0.273r	0.326AB
2% $CaCl_2$+ 0.5% chitosan	0.406a	0.370a-f	0.346a-i	0.326d-m	0.300g-p	0.276k-r	0.338A	0.400a	0.357fg	0.340i	0.313l	0.280q	0.257t	0.324AB
2% $CaCl_2$+ 1% chitosan	0.400ab	0.387a-d	0.343b-j	0.323d-n	0.307g-p	0.273k-r	0.340A	0.390b	0.360ef	0.337i	0.320k	0.290op	0.263s	0.327A
Mean	0.384A	0.350B	0.323C	0.300C	0.276D	0.247E		0.380A	0.351B	0.330C	0.31D	0.280E	0.259F	

Table 8: Effect of some pre-harvest treatments on total phenols (%) of 'Early Swelling' peach fruits stored at 0±1 ºC during 2014 and 2015 seasons. Different letters indicate significantly different values by ANOVA followed by Duncan test at P≤0.05 (small letters refer to values recorded in each season, capital letters refer to mean values).

Treatment	2014 season				Mean	2015 season				Mean
	Storage period (days)					Storage period (days)				
	0	7	21	35		0	7	21	35	
Control	0.044b-g	0.051a-e	0.058ab	0.068a	0.055A	0.027h-k	0.032e-h	0.036de	0.048a	0.036A
1% $CaCl_2$	0.040c-g	0.043b-g	0.047b-g	0.054a-d	0.046AB	0.028h-l	0.031f-i	0.034efg	0.048a	0.035AB
2% $CaCl_2$	0.039c-g	0.043b-g	0.046b-g	0.051a-e	0.045B	0.024klm	0.029g-j	0.035def	0.045ab	0.033ABC
0.5% Chitosan	0.039c-g	0.043b-g	0.046b-g	0.050b-f	0.045B	0.028i-m	0.030g-j	0.032e-h	0.041bc	0.032BC
1% Chitosan	0.037d-g	0.041c-g	0.045b-g	0.052a-d	0.044B	0.026i-m	0.028h-k	0.031f-i	0.039cd	0.031CD
1% $CaCl_2$+ 0.5% chitosan	0.030g	0.038d-g	0.047b-g	0.056abc	0.043B	0.026i-m	0.027h-l	0.029g-j	0.039cd	0.030CDE
1% $CaCl_2$+ 1% chitosan	0.032g	0.039d-g	0.046b-g	0.052a-d	0.042B	0.025j-m	0.025j-m	0.026i-m	0.035def	0.028EF
2% $CaCl_2$+ 0.5% chitosan	0.035efg	0.039c-g	0.044b-g	0.053a-d	0.043B	0.025j-m	0.026i-m	0.027h-l	0.035def	0.029DE
2% $CaCl_2$+ 1% chitosan	0.033fg	0.037d-g	0.040c-g	0.050b-f	0.040B	0.022m	0.023lm	0.024klm	0.033efg	0.025F
Mean	0.037C	0.042BC	0.047AB	0.054A		0.025C	0.028BC	0.030B	0.040A	

CONCLUSIONS

The effect of pre-harvest application of calcium chloride, chitosan and their combinations on storability and quality attributes of 'Early Swelling' peach fruits was investigated in this study. $CaCl_2$ at 2% and chitosan at 1% concentrations proved to be effective in reducing weight loss (%), decay (%), and maintaining maximum firmness and lengthening shelf life. Fruit color (h°) was not affected by any of the treatments, while untreated fruits and calcium chloride alone with both the applied concentrations, were found to maintain higher total soluble solids (%), total phenolic content, and lower titratable acidity percentage (%).

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