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RESEARCH ARTICLE

EFFECT OF CALCIUM CHLORIDE APPLICATION ON QUALITY CHARACTERISTICS AND POST HARVEST PERFORMANCE OF LOQUAT FRUIT DURING STORAGE

Irrum Babu*, Muhammad Azhar Ali , Farah Shamim , Zarina Yasmin , Muhammad Asghar , Abdul Rahim Khan

Post Harvest Research Centre, Ayub Agricultural Research Institute Faisalabad, Pakistan

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Abstract

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*Corresponding Author

Irrum Babu

..... Loquat (Eriobotrya japonica Lindl.) is an important sub-tropical fruit. In Pakistan, Khyber Pakhtunkhwa Province (KPK) and Northern Punjab are the loquat growing provinces and its fruit comes to the market when no other fresh fruit is available. It is an important but little studied fruit for which no research work has been reported inside the country. The objective of this study was to examine the effectiveness of different Ca⁺² treatments on the post-harvest physiology and quality of loquat fruit. Freshly harvested loquat fruit was treated with different concentrations of calcium chloride (1%, 2% and 3%), stored at 4 °C, RH 85-90%, and evaluated regarding various quality parameters. Results showed significant (p = 0.05) retention of firmness and ascorbic acid content in samples dipped in 3% calcium chloride. Total soluble solids content was inversely correlated with acidity and throughout the 24-day storage period was significantly (p = 0.05) lower in samples treated with 3% CaCl₂ than the untreated samples. Results of the weight loss %, firmness and vitamin C assessments suggested that 3% CaCl₂ was helpful in extending shelf-life.

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INTRODUCTION

Loquat (*Eriobotrya japonica* Lindl.) is a popular fruit in Pakistan. Two local cultivars viz., "Surkh" and "Sufaid" are widely grown in the North Western Frontier Province (NWFP) and Punjab province. The "Surkh" cultivar is nearly pear shaped with orange colored skin and flesh while "Sufaid" cultivar has light yellow skin with creamy white flesh and is less acidic (Hussain et al., 2007).

This fruit originated in China and spread to many countries including Japan, Pakistan, India, Italy, Spain, Brazil, Turkey, USA and Australia. Presently, a number of loquat genotypes are being grown in the loquat growing areas of Punjab and Khyber Pakhtunkhwa Province of Pakistan (Hussain et al., 2009). Loquat is grown on an area of 1534 ha in Pakistan, producing 8731 tonnes annually (GOP 2011-12). Loquat fruit has a short shelf life and its quality deteriorates rapidly after harvest (Akhtar et al., 2010). Decay and mechanical damage leading to browning are the prime problems of loquat after harvest (Ding et al., 2002).

Loquat is usually consumed as fresh fruit, but recently a significant amount of loquat fruits is being used in Japan as the major ingredient in various processed food products such as jellies and jams (Kazunori et al., 2007). The fruit contains nearly all the essential nutrients, particularly minerals and carotenoids (vitamin A) (Shaw, 1980). Consumption of loquat has been shown to have a protective effect against many chronic diseases such as cardiovascular disease; the positive health effects are mainly attributable to its high flavonoid contents (Middleton and Kandaswani, 1992; Cook and Samman, 1996).

Loquat fruit are susceptible to decay, mechanical damage, moisture and nutritional losses during their postharvest life. Loquat fruit is very susceptible to mechanical damage during harvesting and handling, and can easily develop brown spots on the fruit surface and in the pulp (Lin et al., 1999). Various experiments have been conducted to identify treatments and techniques to maintain fruit quality and extend postharvest life (Shaw, 1980). While lower temperatures can extend loquat storage periods, this does not completely inhibit the decline in organic acid levels or water loss during prolonged storage (Ding et al., 1998).

Storage at low temperature has been shown to extend the postharvest life in fruit of the Mogi cultivar. Fruit were stored at 1 or 5°C for up to 30 days (Ding et al., 1998). However, fruit weight and acidity was progressively lost, adversely affecting fruit quality. Increased respiration and in ethylene production were not observed during storage at 20°C, which led to the conclusion that Mogi loquat behaved as a non-climacteric fruit (Hamauzu et al., 1997). Recently, loquat fruit has been stored at 5°C for 2 months in the modified atmosphere created by low density polyethylene bags (Ding et al., 2002). Due to the relatively high respiration rate of loquat fruit, low temperature storage was required to maintain the internal carbon dioxide and oxygen concentration below 5 kPa and 4 kPa, respectively, in these bags.

Surface treatments can delay physiological decay in fruit tissues, stabilize the fruit surface and prevent degradation that affects the quality of the product. These treatments can also rinse the enzymes and substrates released by injured cells away from the surfaces of pre-cut fruit (Glenn and Poovaiah, 1990). Pre and postharvest application of calcium may delay senescence in fruits with no detrimental effect on consumer acceptance (Lester and Grusak, 2004). Postharvest applications place the Ca^{2+} solutions in direct contact with the surface of the fruit (Conway et al., 1992) Exogenously applied calcium stabilizes the plant cell wall and protects it from cell wall degrading enzymes (White and Broadley, 2003). Infiltrated calcium in fresh apples has been shown to bind to the cell wall and middle lamellae of cells, where major influences on firmness are expected (Glenn and Poovaiah, 1990).

Calcium has an important role in the ripening process (Ferguson et al., 1995). Studies have shown that the rate of senescence often depends on the calcium status of the tissue; increasing calcium levels alter various measures of senescence such as respiration, protein, chlorophyll content and membrane fluidity (Poovaiah, 1986). Calcium (Ca² ⁺) has been extensively reviewed as both an essential element and in regard to its potential role in maintaining postharvest quality of fruit and vegetable crops (Bangarth, 1979; Kirkby and Pilbeam, 1984). The presence of Ca²⁺ ions contributes to the linkages among pectic substances within the cell-wall (Demarty et al., 1984). It also reduces the rate of senescence and fruit ripening (Ferguson, 1984). A 1% solution of CaCl₂ delayed fruit ripening, improved resistance to fungal attack and maintained the structural integrity of cell walls of strawberry fruit during a 10 day storage period at 3°C (Lara et al., 2004). Softening was delayed and storage life was increased by 10–12 weeks in Kiwi fruits stored at 0°C by application of 1% CaCl₂, compared with untreated fruit (Dimitrios and Pavlina, 2005). A 1% CaCl₂ dip reduced softening and browning rates of 'Bartlett' pear slices (Rosen and Kader 1989). High calcium concentrations decreased calcium-associated flesh browning symptoms in fruits (Hewajulige et al., 2003).

Loquat fruit is non-climacteric and has a short postharvest life (Blumenfeld, 1980). As the fruits ripen and senesce, titratable acidity (TA) declines, accompanied by a loss of flavor and taste, browning and hardening of the fruit flesh, and decreased juiciness (Lin et al., 1999; Zheng and Xi, 1999; Zheng et al., 2000). There is a need for more information on the physiology of loquat fruit ripening and response to storage conditions, to identify methods to increase storage and shelf life (Chong et al., 2006).

In order to expand the fresh produce market for loquat, exploration of possible methods to extend storage-life of perishable commodities is required. Although a reduction in quality is inevitable when the level of Ca2+ drops below the optimal concentration, application to Ca^{+2} to produce commodities as a fungicidal, senescence-delaying treatment has potential to achieve this goal (Esmel, 2005). Therefore, the present study was designed to evaluate the effectiveness of postharvest immersion of fruit into CaCl₂ solutions of various concentrations on the postharvest physiology and quality attributes of loquat fruit in refrigerated stores.

MATERIAL AND METHOD

Fruit of "Surkh" cultivar of loquat were harvested at mature ripe stage from the orchard of Hill Fruit Research Station, Tret, Murree, Pakistan (73° 17' 00"E longitude and 33° 50' 00"N latitude) and transported on the same day to the Post Harvest Research Centre Laboratory at the Ayub Agricultural research Institute, Faisalabad, Pakistan in a refrigerated shipping container. The fruit were clipped with the help of clipper (harvesting tool) to separate individual fruit from the bunch and washed with distilled water to remove any dirt and dipped for two minutes in 0%, 1%, 2% and 3% concentration of calcium chloride (CaCl₂) solutions:

Each treatment was applied to one hundred fruits and was replicated three times. The treated fruit were placed in corrugated cardboard cartons in three layers separated by cardboard sheets and stored at 4°C in the cold store at 85-

90% relative humidity for 24 days. A sample of 10 fruits was randomly selected at day one and again at three-day intervals from each replication in a treatment during the storage period. Several measurements were recorded for each sample at each evaluation date.

Weight Loss

Weight loss was determined by the following formula:

Weight loss (%) = $[(A-B)/A] \times 100$

Where A indicates the fruit weight at the time of harvest and B indicates the fruit weight after storage.

Firmness

Fruit firmness was determined by means of a digital fruit-firmness tester (model 53205, TR di Turoni, Forli, Italy), equipped with an 8mm plunger tip. Firmness was measured as the maximum penetration force reached during fruit penetration and values were expressed in kilogram force (kgf). Each of the 10 fruits in a sample was tested.

Total Soluble Solid

Total soluble solid (TSS) was measured in Brix%, using a digital refractometer (Hanna HI 95801 10450, USA Romania). One wedge shaped slice of uniform size was cut from each of the ten fruits in a sample and combined for further testing. The slices were ground and juice was extracted for the determination of TSS and titratable acidity. Acidity

Titratable acidity was measured with a digital fruit-acidity meter (GMK-835F, Germany) after calibration.

Ascorbic Acid

The ascorbic acid content of loguat fruit was determined using the AOAC method (AOAC, 2000). A 0.1% solution of 2,6 dichlorophenol indophenol (DCPIP) was standardized against a 0.1% (w/v) ascorbic acid solution, and this dye solution was used to titrate the loquat juice. 10 ml juice sample was diluted with 0.4% oxalic acid to make up the volume 100 ml and filtered. Take 10 ml sample from the filtrate, add 15 ml 0.4% oxalic acid solution, shake and titrate against dye solution till the light pink color end point reached. The ascorbic acid was computed according to the expression given below:

Ascorbic acid (mg/ 100mL) = $\frac{R_{1x} V \times 100}{R x W x V_{1}}$

Browning Index

Browning index was assessed visually on weekly basis by measuring the extent of the surface area of a fruit which was affected by browning as described by Wang et al., (2005), using 30 fruits that were selected at random from the treated group of fruit, and were returned to the group after assessment. Fruits were assessed on the following scale: 0 = no browning; 1=less than ¹/₄ browning; 2= ¹/₄ to ¹/₂ browning; 3= ¹/₂ to ³/₄ browning; 4= more than ³/₄ browning. The browning index was calculated using the following formula:

Browning Index = $[(1 \times N1 + 2 \times N2 + 3 \times N3 + 4 \times N4) / (4 \times N)] \times 100$

where N = total number of fruits observed and N1, N2, N3 and N4 were the number of fruits which were scored in each degrees of browning.

The experiment was a completely randomized design (CRD) with a factorial arrangement. Comparison between means was evaluated by Duncan's Multiple Range Test at the 5% level of significance. All storage treatments had three replications.

RESULTS AND DISCUSSION

Weight Loss

Maximum weight loss was observed in the control and 1% CaCl₂ treatments, while the minimum weight loss (1.64%) was observed in the 3% CaCl₂ treatment (Table 1). Weight loss was highest during the 21st and 24th day of storage. Overall, the highest weight loss occurred in the control samples after 17 days of storage (Fig. 1). The reported effects of calcium on membrane functionality and integrity maintenance (Lester & Grusak, 1999) may explain the lower weight loss observed in calcium treated fruits. Mahajan and Dhatt (2004) reported that pear fruit treated with CaCl₂ had reduced weight loss compared to non-treated fruit during 75 days of storage. It is possible that calcium delayed the senescence and reduced the rate of respiration and transpiration in the stored loquat fruit treated with calcium.

Firmness

The maximum firmness was observed in loquats treated with 2% & 3% CaCl₂ as compared to the control and 1% CaCl₂ treatment (Fig. 2, Table 1). Maximum firmness was observed in the 3% CaCl₂ treatment during last five days. The retention of firmness in calcium treated fruits might be due to its accumulation in the cell walls leading to facilitation in the cross linking of the pectic polymers which increases wall strength and cell cohesion (White and

Broadly, 2003). Shuiliang et al (2002) have reported that postharvest dips with $CaCl_2$ maintained firmness and eating quality of loquat.

Total Soluble Solids

The minimum Total Soluble Solids (TSS) content was observed in the 3% CaCl₂ treatment (11.76 Brix %), followed by fruit which received the 2% CaCl₂ treatment. The highest TSS content was observed in the control. The low TSS content in fruit treated with 3% CaCl₂ may be associated with the fact that more concentration of CaCl₂ (3%) formed a thin layer on the surface of fruit which delayed degradation process. The increase in TSS from the second week up to the end of storage (Fig. 3) may be attributable to hydrolysis of polysaccharides and concentration of juice solutes as a result of partial dehydration of the fruit. Increased TSS contents is associated with hydrolytic changes in starch and conversion of starch to sugar. These are important features used to index the ripening process in fruits (Arthey and Philip, 2005). The decrease in respiration rate and conversion of sugars to carbon dioxide and H₂O at later stages of storage can also affect the TSS content (Arthey and Philip, 2005).

Acidity

Acidity is a measure of the concentration of organic acids present in the fruit, which are important in maintaining quality. Titratable acidity decreased gradually in all treatments (Fig. 4) and was not influenced by the postharvest calcium dips. This is similar to the findings of Manganaris et al (2005), who showed that postharvest calcium chloride dips did not affect TA % in peaches during four weeks of storage. In loquat malic acid is the principal acid, contributing 90% of the total organic acid content (Ding et al., 1998). Acidity decreases due to conversion of acids to sugars in fruits during respiration (Ball, 1997). However,In the present study calcium treatments did not significantly affect the TA content. The decrease in TA in all treatments at the end of storage may be attributable to the metabolic changes in the fruits or consumption of organic acid in respiratory process (Echeverria and Valich, 1989).

Ascorbic acid

Ascorbic acid (Vit C) levels decreased gradually in all treatments during 24 days storage period. Fruit from the 1%, 2% and 3% CaCl₂ treatment had ascorbic acid losses of 10.9%, 8.4% and 2.5%, respectively, compared to the initial content, whereas Vit C content decreased 19% in the control treatment (Fig. 5). Ascorbic acid is an important nutrient quality parameter and is very sensitive to degradation due to its oxidation (Veltman et al., 2000). The retention of Vit C observed here may be because the high concentrations of CaCl₂ delayed the rapid oxidation of ascorbic acid. Ruoyi et al (2005) found that ascorbic acid content of peaches was maintained during fifty days of storage in response to a postharvest application of 0.5% CaCl₂.

Browning Index

Overall browning index increased during storage (Fig. 6). Data in Table 1 shows a significant difference in browning percent as a result of $CaCl_2$ treatments. The maximum browning (8.46%) was observed in the control fruit, while the least browning (5.34%) was observed in fruit which received the 3% $CaCl_2$ treatment. This could be attributable to better membrane stability in the cacium-treated fruit (Poovaiah, 1988; Picchioni et al., 1995).

This could be due to the fact that calcium helps to maintain membrane stability Membrane deterioration is a consequence of senescence related peroxidation of membrane lipids and is associated with tissue browning (Thompson et al., 1987). Oxidative membrane injury also allows the mixing of the enzyme poly phenol oxidase (PPO) and oxidizable substrates (polyphenols), which are normally separated by cellular membranes. This can lead to browning (Hodges, 2003). High calcium concentrations result in decreased flesh browning symptoms which are directly associated with calcium content in fruits (Hewajulige et al., 2003). Calcium dips could reduce browning in loquat fruits, which are less susceptible to flesh browning symptoms than those studied by Hewajulige et al (2003). 1% CaCl₂ dip reduced softening and browning rates of 'Bartlett' pear slices (Rosen and Kader, 1989).

Table 1.Effect of different CaCl ₂ treatments on the Post harvest quality of loquat fruit during 24 days storage at 4^{0} C						
and 85-90% relative humidity (average data)						
Treatments	Weight loss (%)	Browning index (%)	Acidity (%)	TSS (%)	Vitamin C	Firmness (kg)
T0	2.82a	8.46a	0.42b	13.71a	1.8c	1.84d
T1	2.24b	6.83b	0.45c	13.22b	2.20b	2.5c
T2	1.73c	5.92c	0.51a	12.87c	2.60a	2.65b
T3	1.38d	5.34d	0.52a	11.76d	2.60a	2.70a
$T_0 = 0\%$ CaCl ₂ (control)						
$T_1 = 1$ % CaCl ₂ solution						
$T_2 = 2 \%$ CaCl ₂ solution						

$T_3 = 3 \%$ CaCl₂ solution

Values in a column which are followed by the same letters are not significantly different at p<0.05













CONCLUSION

This study shows that $CaCl_2$ treatment had a significant effect on the keeping quality of loquat fruit. 1% $CaCl_2$ treatment did not show significant effect on quality parameters and was similar to the control, while 2% $CaCl_2$ had higher firmness and least browning. Dipping fruit in 3% CaCl2 retained maximum TSS, firmness and reduced browning index and weight loss up to 24 days.

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