

# Quality changes in fresh-cut pear slices as affected by controlled atmospheres and chemical preservatives

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Received 7 November 2000; accepted 7 June 2001

## Abstract

Low O<sub>2</sub> (0.25 or 0.5 kPa) elevated CO<sub>2</sub> (air enriched with 5, 10 or 20 kPa CO<sub>2</sub>), or superatmospheric O<sub>2</sub> (40, 60, or 80 kPa) atmospheres alone did not effectively prevent cut surface browning or softening of fresh-cut pear slices. A post-cutting dip of 2% (w/v) ascorbic acid, 1% (w/v) calcium lactate and 0.5% (w/v) cysteine adjusted to pH 7.0 did significantly extend shelf-life of 'Bartlett' pear slices, by inhibiting loss of slice flesh firmness and preventing cut surface browning. Participants in a quality evaluation could not distinguish between pear slices treated with this preservative solution and stored overnight at 0 °C and freshly prepared control pear slices. After 10 days storage in air at 0 °C, 82% of participants judged treated pear slices to be acceptable in appearance and 70% judged flavor to be acceptable. © 2002 Elsevier Science B.V. All rights reserved.

**Keywords:** Ascorbic acid; Browning; Calcium lactate; Controlled atmosphere; Cysteine; Minimal processing Pear

## 1. Introduction

The greatest hurdles to the commercial marketing of fresh-cut fruit products, including fresh-cut pear slices, are limited shelf-life due to excessive tissue softening and cut surface browning. Cut surface browning in sliced pears is caused by the action of polyphenol oxidase (PPO) on phenolic compounds released during the process of cutting (Amiot et al., 1995). Fruit tissue softening during

ripening and senescence is triggered by ethylene and has been demonstrated to be a consequence of alterations in cell wall metabolism. There are numerous chemical and physical preservation strategies that can be used to reduce enzymatic browning (McEvily and Iyengar, 1992) and fruit tissue softening after cutting. However, many of these treatments impart off flavors or the compounds used may not be generally recognized as safe by the FDA.

Many factors affect the shelf-life of fresh-cut pear slices including cultivar, stage of ripeness at cutting and storage regime before processing (Sapers and Miller, 1998; Gorny et al., 2000). The

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optimal pear fruit ripeness stage for fresh-cut processing based on flesh firmness is between 45–58 N firmness (penetration force); if softer fruit are used, shelf-life is less due to increased cut surface browning (Gorny et al., 1998). Rosen and Kader (1989) indicated that a combination of a 1%  $\text{CaCl}_2$  dip and 0.5%  $\text{O}_2$  atmosphere reduced softening and browning rates of 'Bartlett' pear slices. It has also been demonstrated that a combination of 1% calcium lactate and 2% ascorbic acid applied as a post-cutting dip can reduce the incidence of cut surface browning and firmness loss in fresh-cut pears (Gorny et al., 1998). Unfortunately, this treatment alone does not completely control post-cutting enzymatic browning of fresh-cut pear slices, if the pears are not peeled, since discoloration occurs on the cut surface at the pear flesh/skin interface directly beneath the pear fruit epidermis (peel).

Recently, Dong et al. (2000) reported that a 2-min dip in 0.01% 4-hexylresorcinol + 0.5% ascorbic acid + 1% calcium lactate can provide 15–30 days shelf life at 2–3 °C for 'Anjou', 'Bartlett', and 'Bosc' pears that were sliced at 43, 49, and 38 N flesh firmness, respectively: panelists could not detect a flavor difference between 0.01% 4-hexylresorcinol treated pears and controls.

The thiol containing amino acid cysteine has been reported to effectively inhibit PPO-mediated enzymatic browning of fruits and vegetables (Joslyn and Ponting, 1951) and has been reported to completely inhibit enzymatic browning in fresh-cut potatoes (Gunes and Lee, 1997). Three mechanisms have been proposed to explain how thiol compounds inhibit enzymatic browning; (1) reduction of the o-quinone back to o-dihydroxyphenol (Kahn, 1985); (2) direct inhibition of PPO (Dudley and Hotchkiss, 1989; Robert et al., 1996); and (3) the formation of a colorless cys-quinone adduct (Richard et al., 1991). When cysteine is used as an inhibitor of enzymatic browning on sliced apples (Walker and Reddish, 1964) or pears (Sapers and Miller, 1998), pinkish-red colored compounds are formed due to phenol regeneration with deep color formation (Richard-Forget et al., 1992). If this off-color formation can be prevented by for

example altering the treatment solution pH, cysteine may prove to be an effective replacement of bisulfites, since cysteine is a naturally occurring amino acid that has GRAS status for use as a dough conditioner (Code of Federal Regulations 21:184.1271 and 21:184.1272).

Atmospheres reduced in  $\text{O}_2$  and elevated in  $\text{CO}_2$  can also extend the postharvest life of many whole commodities (Kader, 1986). Low  $\text{O}_2$  and/or elevated  $\text{CO}_2$  environments generated by modified atmosphere packaging of fresh-cut produce can extend produce shelf-life by (1) slowing browning reactions at cut surfaces; (2) reducing the rates of product transpiration (water loss) and respiration; and (3) reducing  $\text{C}_2\text{H}_4$  biosynthesis and action (Gorny, 1997). Fresh-cut iceberg lettuce products are routinely commercially packaged in plastic bags that have a low  $\text{O}_2$  (< 0.5%) and elevated  $\text{CO}_2$  (> 10%) atmosphere to reduce cut surface browning. The efficacy of modified atmospheres to extend the shelf-life of fresh-cut fruit products has not been studied extensively and warrants investigation.

The objectives of our research were to determine the affects that: (1) atmospheric modification; and (2) post-cutting chemical treatments with cysteine in combination with calcium lactate and ascorbic acid, have on the subsequent shelf-life of pear slices, based on appearance and eating quality.

## 2. Materials and methods

### 2.1. Fruit storage and ripening conditions

'Bartlett' pears (110 count, mean fruit mass = 166 g) were obtained on the day of harvest from commercial grower/shippers in Lake and Sacramento Counties, transported to Davis, CA, and stored at -1 °C in air for up to 4 months, until they were used in various studies. Fruit were ripened at 20 °C and 90–95% relative humidity in air + 10 Pa ( $100 \mu\text{l l}^{-1}$ )  $\text{C}_2\text{H}_4$ . Ripeness stage was based on measuring the force required for an 8 mm probe to penetrate the cheek of 20 pears per lot, with the skin removed, to a depth of 10 mm using a University of California firm-

ness tester (Western Industrial Supply Co., San Francisco, CA). After ripening, fruit were cooled to  $\approx 0$  °C in 3–4 h using a laboratory scale forced-air cooling unit and held overnight at 0 °C until cut. Before cutting, the firmness of each individual fruit was determined, to select pears with a flesh firmness of 45–58 N. These pears were each cut into eight slices (wedges) with a sharp stainless steel knife, and the wedge used to determine firmness was discarded. Fruit wedges were then dipped in distilled water with 2.7 mM sodium hypochlorite or treatment solutions for 5 min, gently dried by hand with cheesecloth, and then placed (20 slices per replicate) into 2-l jars ventilated with a continuous flow ( $100 \text{ ml min}^{-1}$ ) of humidified air at 0 °C. Each jar of 20 slices was considered a replicate and three replicates were used per treatment.

## 2.2. Controlled atmosphere treatments

'Bartlett' pear slices were dipped in distilled water containing 2.7 mM sodium hypochlorite (pH 7.0) and placed into jars (20 slices per jar) at 5 °C ventilated with humidified air, 0.25 kPa O<sub>2</sub>, 0.5 kPa O<sub>2</sub>, 19.9 kPa O<sub>2</sub> + 5 kPa CO<sub>2</sub>, 18.8 kPa O<sub>2</sub> + 10 kPa CO<sub>2</sub>, or 16.7 kPa O<sub>2</sub> + 20 kPa CO<sub>2</sub> (balance N<sub>2</sub> in all gas mixtures) at a flow rate of  $100 \text{ ml min}^{-1}$ . Additionally, atmospheres of air, 40, 60 or 80 kPa O<sub>2</sub> (balance N<sub>2</sub>) were also tested at 10 °C.

## 2.3. Cysteine, ascorbic acid and calcium treatments

'Bartlett' pear slices were immersed for 5 min at 20 °C in a solution containing: (1) distilled water; (2) 0.032 M (1% w/v) calcium lactate; (3) 0.114 M (2% w/v) ascorbic acid; (4) 0.041 M (0.5% w/v) cysteine; (5) 0.032 M calcium lactate + 0.114 M ascorbic acid + 0.041 M cysteine (pH 3.7); or (6) 0.032 M calcium lactate + 0.114 M ascorbic acid + 0.041 M cysteine + enough NaOH to reach pH 7.0. After treatment, slices were gently dried by hand with cheesecloth, and then placed into jars (20 slices per jar) and held at 0 °C in a continuous flow ( $100 \text{ ml min}^{-1}$ ) of humidified air.

## 2.4. Residual cysteine, ascorbic acid and calcium analysis

Slices were cut from pear fruit that were 45–58 N firmness, then soaked for 5 min at 20 °C in a solution of: (1) distilled water; or (2) 0.114 M (2% w/v) ascorbic acid + 0.032 M (1% w/v) calcium lactate + 0.041 M (0.5% w/v) cysteine + enough NaOH to reach pH 7.0, blotted dry with cheesecloth and then placed in air at 0 °C. Pear slices were analyzed to determine the amount of ascorbic acid, calcium and cysteine present after storage at 0 °C in air for 0, 3, 7 or 10 days. Ascorbic acid was determined by HPLC using the method of Perez et al. (1997). Cysteine concentrations were determined by the method of Buwalda et al. (1988) and modified by Pesis et al. (1998). Total flesh calcium concentrations were determined by flame ionization atomic adsorption spectrophotometry of ashed samples.

## 2.5. Quality evaluations

The visual quality of each replicate was determined based on the following visual hedonic scale: 9, excellent, just sliced; 7, very good; 5, good, limit of marketability; 3, fair, limit of usability; and 1, poor, inedible. A color photograph of slices rated via this scale was used by two or three of the researchers (authors of this paper) to score individual slices based on color, visible structural integrity and general visual appeal. A weighted average of individual fruit slice quality scores was calculated as the mean visual quality score for each replicate. CIE  $L^*a^*b^*$  values were determined (midpoint between endocarp and skin) with a Minolta chromameter (Model CR-200, Minolta, Ramsey, N.J.) calibrated to a white porcelain reference plate ( $L^* = 97.95$ ,  $a^* = -0.39$ ,  $b^* = 2.00$ ). The  $L^*$  color value or hue angle ( $h^\circ$ ) was used as an indicator of cut surface browning intensity (Sapers and Douglas, 1987). Pear slice firmness (penetration force) was determined by measuring the force required for a 3 mm probe to penetrate the slice surface (midpoint between endocarp and skin), held perpendicular to the probe,

to a depth of 10 mm using a University of California firmness tester.

## 2.6. Sensory-based shelf-life analysis

Quality evaluations were done using human subjects, because it is important that not just quantifiable physical and chemical measurements be used to determine if a shelf-life extending treatment has merit. Quality evaluation was done by presenting participants with treated and untreated pear slices and asking a series of simple questions to determine: (1) if the control and treated pear slices could be obviously distinguished from each other by untrained persons; and (2) what is the perceived acceptability of pear slices that have been treated and held in cold storage for various lengths of time. Slices were cut from pear fruit that were 45–58 N firmness, then soaked for 5 min at 20 °C in a solution of: (1) distilled water; or (2) 0.114 M (2% w/v) ascorbic acid + 0.032 M (1% w/v) calcium lactate + 0.041

M (0.5% w/v) cysteine + enough NaOH to reach pH 7.0, blotted dry with cheesecloth and then placed in air at 0 °C overnight. Control fruit of the same lot were held overnight at 0 °C and then sliced the next day and rinsed in distilled water. Ninety-seven randomly-selected consumers (university students, staff and faculty) were each given two sets of control and treated samples and asked to taste each slice and then answer the question, “Which slice do you prefer, a or b?”

In another experiment, 50 randomly-selected consumers were asked, “Is this pear slice acceptable in appearance?” and “Is this pear slice acceptable in flavor?”, after being treated as mentioned above and stored for 3, 5, 7 or 10 days in air at 0 °C.

## 2.7. Statistical analysis

Data were treated for multiple comparisons by analysis of variance with least significant difference (LSD) between means determined at 5% level.

## 3. Results and discussion

### 3.1. Effects of controlled atmospheres on shelf-life and quality

Pear slices kept in air, 0.25 kPa O<sub>2</sub>, 0.5 kPa O<sub>2</sub>, air + 5 kPa CO<sub>2</sub>, air + 10 kPa CO<sub>2</sub>, or air + 20 kPa CO<sub>2</sub> all softened at similar rates (data not shown) and exhibited significant cut surface discoloration as measured by *L\** value changes during storage at 5 °C. (Fig. 1A and B). Atmospheres of air + 10 kPa CO<sub>2</sub> and air + 20 kPa CO<sub>2</sub> accelerated tissue browning and necrosis compared to the air control. Tissue necrosis and severe cut surface browning first occurred in the flesh tissue closest to the core and spread radially outward over time. High CO<sub>2</sub> injury occurred in a dose responsive manner with damage occurring earlier and more severely in air + 20 kPa CO<sub>2</sub>-treated slices than in air + 10 kPa CO<sub>2</sub>-treated slices. These symptoms are similar to those observed in whole fruit (Meheriuk et al., 1994). Controlled atmospheres of 1–2 kPa O<sub>2</sub> and < 1

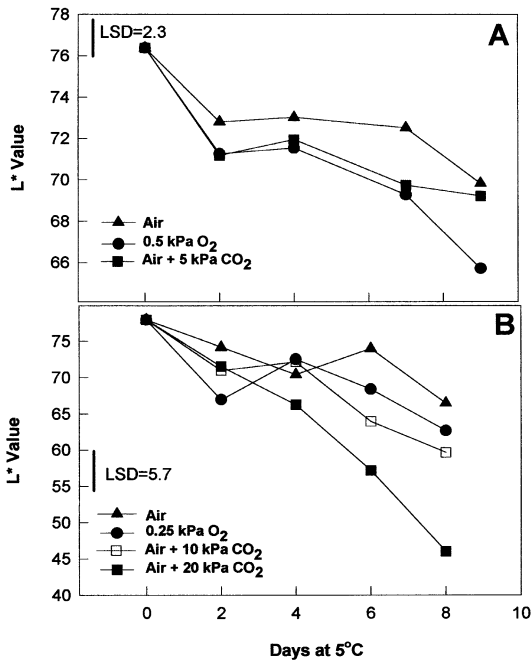


Fig. 1. Effects of elevated CO<sub>2</sub> or reduced O<sub>2</sub> atmospheres on the cut surface color of pear slices kept at 5 °C. Data shown are the means of three replicates of 20 slices each. Vertical bar = pooled LSD at the 5% level.

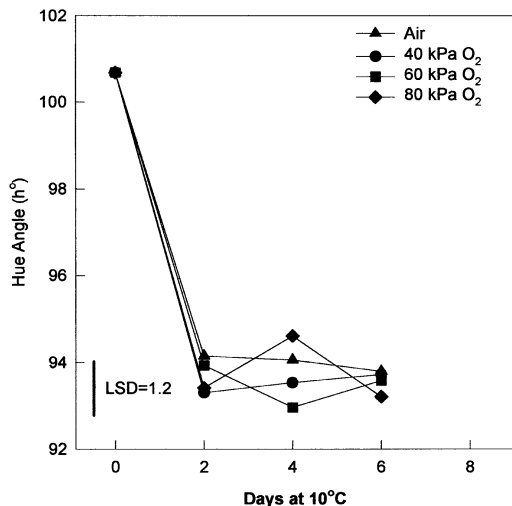


Fig. 2. Effects of elevated O<sub>2</sub> atmospheres on the cut surface color of pear slices kept at 10 °C. Data shown are the means of three replicates of 20 slices each. Vertical bar = pooled LSD at the 5% level.

kPa CO<sub>2</sub>, which are commercially used to extend the storage life of whole, unripened 'Bartlett' pears, do not provide enough benefits to recommend their commercial use on fresh-cut pears. Senesi et al. (1999) found that increased CO<sub>2</sub> levels within packages did not protect fresh-cut pears from browning. 'Abate Fetel' cultivar was more suitable than 'Kaiser' for fresh-cut products. They concluded that pears can be processed into fresh-cut products with an expected shelf-life of 8–12 days at 3 + 1 °C, but the highest level of quality only lasts until 4–8 days post-cutting.

Pear slices kept in air, 40, 60 or 80 kPa O<sub>2</sub> (balance N<sub>2</sub>) all softened at similar rates (data not shown) and exhibited similar severities of cut surface browning during storage at 10 °C (Fig. 2.). Superatmospheric O<sub>2</sub> atmospheres have been suggested to inhibit enzymatic browning of fresh-cut produce (Day, 1996), however our results do not support these claims.

### 3.2. Effects of chemical treatments on the shelf-life and quality

Cut surface darkening as measured by (*h*°) of pear slices was completely inhibited for up to 8 days at 0 °C by treatment with an aqueous solu-

tion of 0.114 M (2%w/v) ascorbic acid + 0.032 M (1% w/v) calcium lactate + 0.041 M (0.5% w/v) cysteine adjusted with NaOH to a pH 7.0 (Fig. 3.). The mixture of 2% ascorbic acid + 1% calcium lactate + 0.5% cysteine at pH 3.7 also significantly reduced cut surface flesh darkening of pear slices compared to a control of distilled water for up to 4 days at 0 °C. However, after 4 days at 0 °C the mixture of 2% ascorbic acid + 1% calcium lactate + 0.5% cysteine at pH 3.7 and the 0.5% cysteine treatment alone resulted in the formation of undesirable pinkish-red pigments. The 2% ascorbic acid or 1% calcium lactate treatment alone did not prevent cut surface darkening at the flesh/skin interface just beneath the fruit epider-

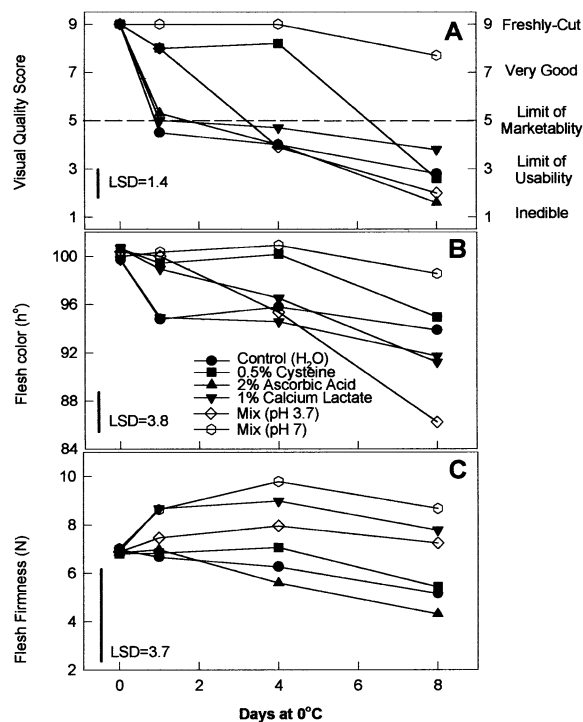


Fig. 3. Effects of post-cutting chemical dips on pear slice (A) visual quality, (B) flesh color (hue angle) and (C) firmness. Slices were dipped for 5 min at 20 °C in: (1) distilled water (control); (2) 0.5% (0.041 M) cysteine; (3) 2% (0.114 M) ascorbic acid; (4) 1% (0.032 M) calcium lactate; (5) mix (pH3.7) 0.041 M cysteine + 0.114 M ascorbic acid + 0.032 M calcium lactate; or (6) mix (pH7.0) 0.041 M cysteine + 0.114 M ascorbic acid + 0.032 M calcium lactate (+ enough NaOH to reach pH 7.0). Data shown are the means of 20 slices. Vertical bar = pooled LSD at the 5% level.

mis where a thin line of discoloration developed. In previous work we determined that a minimum of 2% ascorbic acid and 1% calcium lactate were necessary to inhibit flesh browning and prevent flesh softening of fresh-cut 'Bartlett' pear discs, (Gorny et al., 1998). Addition of 0.5% cysteine and adjusting the pH to 7 increased the efficacy of the mixture by controlling the subepidermal browning.

Sulfites have been demonstrated to act as a reducing agent at a pH below 4 (Cheynier et al., 1989). At a pH above 4, quinones will form colorless adduct products with sulfites, cysteine or glutathione (Richard et al., 1991). Nucleophilic attack of quinones by cysteine may be more effective at a neutral pH since the thiol group of cysteine has a  $pK_a$  of 8.33. Because fruit tissue is acidic, use of cysteine alone as an inhibitor of enzymatic browning is not effective. However, when cysteine is used in combination with ascorbic acid, which is a reducing agent and weak buffer at pH 7.0, PPO-mediated enzymatic browning can be completely inhibited in pear slices for up to 8 days at 0 °C.

### 3.3. Cysteine, ascorbic acid and calcium analysis

Immediately after treatment, ascorbic acid and cysteine levels in slices were significantly greater in treated slices compared to controls (Fig. 4). However, after 3 days at 0 °C ascorbic acid and cysteine residues on fruit slices dropped to endogenous control levels in the treated slices. Ascorbic acid was most likely converted to dehydroascorbic acid and then further degraded to 2,3 diketo-gluconic acid. Cysteine may have formed adduct products with phenolic compounds, which is the proposed mechanism for inhibition of enzymatic browning. Calcium levels were not significantly higher in treated fruit slices compared to control slices except after 10 days of storage at 0 °C.

Only very small quantities of the treatment compounds were present in the fruit tissue even just after dipping. This may be due to the treatment solution penetrating only a short distance into the fruit tissue and that the samples were analyzed on a whole slice weight basis. This is

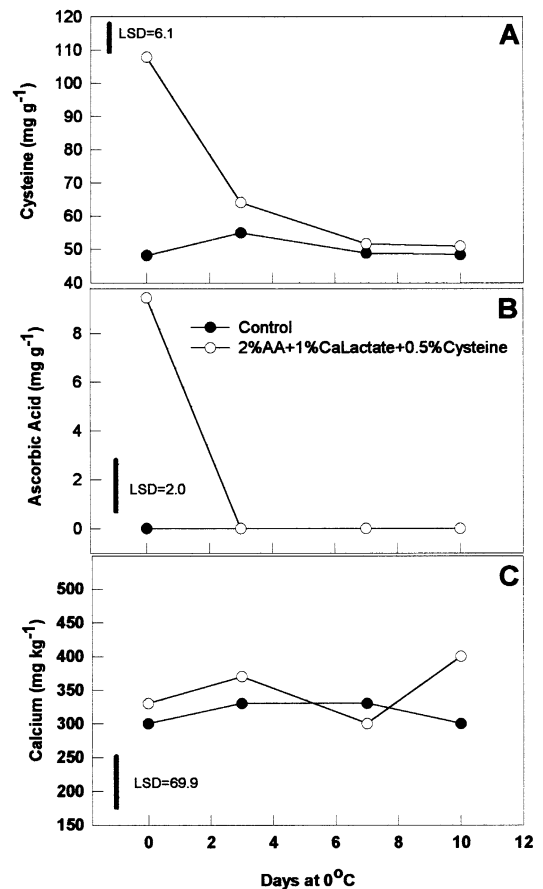


Fig. 4. Effects of a 2% (w/v) ascorbic acid + 1% (w/v) calcium lactate + 0.5% (w/v) cysteine (+ enough NaOH to reach pH7.0) dip on residual pear slice: (A) cysteine; (B) ascorbic acid; and (C) calcium contents. Data shown are the means of 20 slices. Vertical bar = pooled LSD at the 5% level.

beneficial since the treatment compounds are mainly present where they are needed at the cut surface of the slice. Because ascorbic acid, cysteine and calcium levels drop to or are near endogenous levels, these compounds might be considered processing aids since they are naturally present in pear tissue. However, the US Food and Drug Administration may consider these compounds to be functioning as preservatives, a direct food additive, in which case they must be labeled as a preservative and the treated product could not be labeled as a 'fresh' (21CFR101.95). The treatment solution should not be reused as it may serve as source of microbial cross contamination.

### 3.4. Sensory evaluation

Participants in the preference test could not distinguish between pear slices that had been treated with the aqueous solution of calcium lactate + ascorbic acid + cysteine adjusted with NaOH to a pH of 7.0 compared to fruit slices which had been just cut and rinsed in distilled water. In our consumer taste panel, 94 respondents preferred the treated pear slices and 100 preferred the untreated samples. Chi-square statistical analysis ( $P < 0.05$ ) indicated that consumer panelists had no strong preference (or objection) to either of the samples they were asked to evaluate. From this it can be concluded that use of this treatment to extend the shelf-life of fresh-cut pear products is a viable option.

The shelf-life of a food product can be defined as determining how many consumers one is willing to disappoint with a food product at any given point in time (Labuza and Schmidl, 1988). To this end we determined how long fresh-cut pear slices treated with ascorbic acid, calcium lactate and cysteine (pH 7.0) remained acceptable to consumers over a 10-day period of time. After

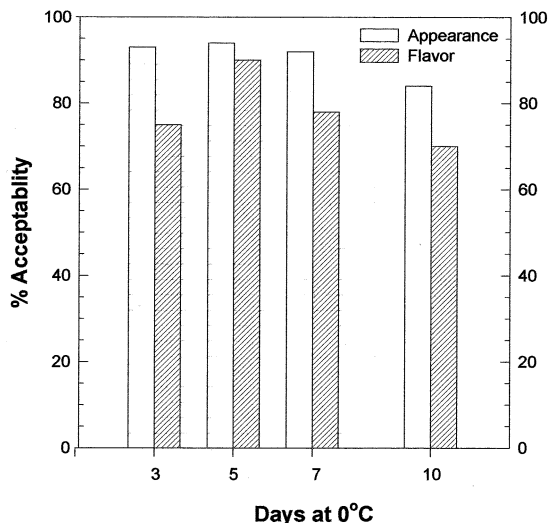


Fig. 5. Effects of post-cutting storage time on consumer acceptability of pear slices based on appearance and flavor. Slices were treated with 2% (w/v) ascorbic acid + 1% (w/v) calcium lactate + 0.5% (w/v) cysteine (+ enough NaOH to reach pH 7.0) and stored in air at 0 °C.

3, 5, 7 or 10 days in air at 0 °C, 50 randomly selected consumers were asked to determine if the pear slice they were presented with was acceptable in appearance and flavor. After 3 days of storage, 93% of the respondents rated the pear slices as acceptable in appearance and 75% of the respondents found the pear slices to be of acceptable flavor (Fig. 5). After 10 days of storage in air at 0 °C, 82% of consumers found appearance acceptable and 70% of consumers found flavor acceptable.

### 4. Conclusions

Low O<sub>2</sub> (0.25 or 0.5 kPa) or elevated CO<sub>2</sub> (air + 5, 10 or 20 kPa CO<sub>2</sub>) or superatmospheric O<sub>2</sub> (40, 60 or 80 kPa) atmospheres alone did not effectively prevent cut surface browning or firmness loss in fresh-cut pear slices. A post cutting dip of 2% (w/v) ascorbic acid, 1% (w/v) calcium lactate, and 0.5% (w/v) cysteine adjusted with NaOH to pH 7.0 significantly extended shelf-life of the slices by inhibiting loss of firmness and cut surface browning. Sensory analysis of the cysteine + ascorbic acid + calcium lactate (pH 7.0)-treated slices revealed no objectionable off-flavors. Use of this mixture to inhibit enzymatic browning may help fresh-cut processors overcome one of the significant hurdles that currently impedes the commercial development of fresh-cut fruit products.

### Acknowledgements

Research supported in part by US Department of Agriculture research agreement no. 95-37500-1910, NRI Competitive Grants Program, USDA and the California Pear Advisory Board. We thank Sarah Cathcart, Carolyn Menke and Erica Mandle for valuable assistance.

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