

EFFECTS OF CONTROLLED ATMOSPHERE ON QUALITY OF MINIMALLY PROCESSED APPLE (cv. JONAGORED)

ADA MARGARIDA CORREIA NUNES ROCHA and
ALCINA MARIA MIRANDA BERNARDO DE MORAIS¹

*Escola Superior de Biotecnologia da Universidade Católica Portuguesa
Rua Dr. António Bernardino de Almeida
4200-072 Porto, Portugal*

Accepted for Publication March 31, 2000

ABSTRACT

Controlled atmosphere (2% O₂ + 4% to 12% CO₂) storage was found to be advantageous over air storage in terms of overall preservation of fresh-cut apple (cv. Jonagored). After 3 days, CA-stored apple cubes were firmer, showed better color and higher content of fructose and soluble solids content than air-stored cubes. In relation to color preservation this benefit was increased when CO₂ in the atmosphere was increased to 12% CO₂. Controlled atmosphere storage showed no advantage over air storage in relation to sensory evaluation of firmness and flavor.

INTRODUCTION

To satisfy the increasing consumers' interest in ready-to-use (minimally processed) fruits, extensive research has been performed in order to extend the shelf-life of those products. Fruits are intended to be sold conveniently peeled, cored or sliced, providing high nutritional value, fresh in appearance and ready for consumption (Bolin and Huxsoll 1989). However, minimally processed (MP) products may suffer several degrading reactions, particularly severe color changes, which lead to the loss of 'fresh' quality. Control of browning of MP fruits is an essential step for their successful marketing.

Extensive information is available on the use of chemical additives to reduce browning (Pizzocaro *et al.* 1993; Janovitz-Klapp *et al.* 1990; Rouet-Mayer *et al.* 1989; Rocha *et al.* 1998). However, due to the consumers increasing concern about the use of chemical additives on food products, research has been carried out using alternatives, such as heat, irradiation and

¹ Corresponding author: E-mail: amorais@esb.ucp.pt; FAX: 351-22-5090351; TEL: 351-22-5580050.

moisture reduction (Wiley 1994), edible coatings (Baldwin *et al.* 1995), and also combined techniques that take advantage of the synergism of the various preservation techniques (Bolin and Huxsoll 1989; Huxsoll *et al.* 1989; Nicolas *et al.* 1994). Because controlled atmosphere (CA) is considered an innocuous treatment, it is a promising technique for preserving MP fruits (Huxsoll *et al.* 1989).

Use of CA for the commercial storage of whole apples is widely practiced (Ke *et al.* 1991; Lau and Yastremski 1991; Drake 1993; Drake *et al.* 1993) and several advantages over air storage have been described. With respect to firmness, Knee (1980) reported a reduction of apple flesh (cv. Cox's Orange Pippin) softening by storing under 2.5-4.0% O₂; Lau and Looney (1982) also obtained firmer apples and higher titratable acidity (TA) with a storage atmosphere of 2.5% O₂. Herregods and Goffings (1993) and Sfakiotakis *et al.* (1993) reported a reduction in the loss of firmness of 'Jonagold' and 'Golden Delicious' apples respectively, when stored in 1% O₂ + 1.5 to 2% CO₂ at 0.1 to 1°C. With respect to chemical composition, Chen *et al.* (1989) reported that low O₂ storage resulted in apples with higher titratable acidity (TA) and were crisper, more acid, and juicier to sensory panelists.

Recommended CA storage conditions for whole apple are well established and have been reported to be cultivar dependent. Kader (1989) suggested CA conditions of 2% O₂ minimum and 5% CO₂ maximum, concentrations tolerated by the fruit for the majority of apple varieties. Labuza and Breene (1989) recommended 3% O₂ and 5% CO₂ for 'McIntosh' and 'Newton' varieties, and 3% O₂ and 3% CO₂ for other apple varieties. No specific information exists concerning recommended storage conditions for 'Jonagored'. For 'Jonagold', the variety from which 'Jonagored' originated, the CA conditions recommended are variable: 1.5% O₂ + 1.5% CO₂ (Lau 1988), 1.25% O₂ + < 1% CO₂ (Stow 1987), 1% O₂ + 3% CO₂ (Drake 1993).

Limited information is available in the literature concerning tolerance limits of MP apple to CA, and little is known about the effects of reduced O₂ and elevated CO₂ on biochemical and physiological reactions that take place in apple tissue after harvest, and more precisely after minimal processing. Kader (1989) and Brecht (1995) suggested that MP fruits have fewer barriers to gas diffusion than whole fruits, resulting in much reduced gas concentration gradients within the tissue. Therefore MP fruit should be able to tolerate and benefit from higher concentrations of CO₂ and lower O₂ levels than intact commodities. On the other hand, abusive CA conditions (injurious O₂ or CO₂ levels) can affect cellular integrity and result in decompartmentalization and reaction of enzymes (e.g. polyphenoloxidase) and phenolic substrates (Barrett *et al.* 1991). Nicoli *et al.* (1994) stored 'Golden Delicious' apple slices in 20% CO₂ after dipping in ethanol solutions and reported prevention of browning reactions for at least 9 days of storage. Bolin and Huxsoll (1989) reported that peeled and halved peach

and apricot fruits can retain their light color and firm texture by dipping in 2% calcium chloride plus 1% zinc chloride followed by anaerobic packaging and storage at 0-2°C. However, the same treatment did not succeed with pears.

Data on the effects of CA storage on MP apple browning are limited, and since the response of apples to different storage conditions varies according to variety, no common CA conditions exist for application to all apple varieties. Therefore, new varieties necessitate research on specific tolerance limits.

The aim of this work was to evaluate the effects of elevated CO₂ levels on 'Jonagored' cut apple quality, particularly with respect to browning. CA conditions tested were 2% O₂ + 4% CO₂; 2% O₂ + 8% CO₂; 2% O₂ + 8% CO₂ after dipping in 42.5 mM ascorbic acid (AA) solution; 2% O₂ + 12% CO₂. Combined use of an ascorbic acid dip with CA storage was performed in order to use one more hurdle to the browning reactions. Chemicals were selected according to results obtained in previous work (Rocha *et al.* 1998) that focused on effects of chemical treatments on the quality of fresh-cut apple. Several authors have suggested that ascorbic acid effectiveness may be enhanced in conjunction with CA storage (Sapers and Hicks 1989).

MATERIALS AND METHODS

Plant Material

Apples, *Malus domestica*, cv. Jonagored, were grown at Estação Regional de Fruticultura e Vitivinicultura — Quinta de Sergude, Felgueiras, Portugal. 'Jonagored' is a new variety with promising dessert characteristics and good storage ability. It is a red mutant of 'Jonagold' with a more attractive red color. The harvest was on September 10th. Fruits were stored in air at 4°C for 1 to 3 months until used in the experiments.

Treatment and Storage Conditions

Apples stored under refrigeration (4°C) were transported weekly to the laboratory in Porto in bulks of about 10 kg randomly selected. Apples were initially washed with chlorinated water (150 ppm of active chlorine for 5 min) to remove surface contamination (Wardowski and Brown 1991). After peeling the apples were hand cut into cubes of ≈ 1.5 cm using a sharp cutter. Apples were submerged in distilled water or a chemical solution of AA (42.5 mM) for 5 min (Rocha *et al.* 1998). Three replicates of 40 cubes randomly selected from the whole bulk available were used for each storage time and atmosphere composition. Apple samples were stored in sealed glass jars of 1 L capacity, under the specific CA conditions at 4°C for 7 days in the dark. Lids of sealed glass jars were provided with three holes. The CA mixture was flushed into and

out of the jars through two of the holes equipped with barbed bulkhead polypropylene fittings with rubber tubings on both ends. Silicone gasket and nut assemblies provided leakproof service. The third hole was equipped with a rubber stopper with hollow plug to allow for gas sampling. Desired gas mixtures were established by the use of flowmeters and metered to the samples at a flow rate of 3.0 L / h after humidification by bubbling through water. Gas concentrations in the head space of the glass jars were measured by injection of 0.6 mL in a gas chromatograph (Shimadzu GC-14-A, Tokyo, Japan), equipped with a thermal conductivity detector (TCD) and connected to a 3 m × 0.32 cm column packed with 80/100 mesh Carbosieve S II (Supelco). Injector and detector temperatures were set at 120 and 210C, respectively. Temperature of the column oven was programmed for 40C for 6 min and subsequently for 15C/min to 170C and then held at 170C for 5 min. Flow rate of the carrier gas, helium, was 30 mL/min and the bridge-current of the TCD was 140 mA.

Three replicates of cut apple were stored in humidified air after immersion in distilled water, in parallel with each experimental series and used as control. After 3 and 7 days of storage under the specified conditions, samples were removed and evaluated in terms of several physicochemical quality attributes as detailed below. Cut apple samples were analyzed on day zero (after peeling and cutting) as a reference. All the experiments were repeated within approximately 1.5 months.

Samples of 10 cubes were weighed, then the color and firmness measured as described. The same 10-cube samples were homogenized for subsequent chemical measurements. The remaining 30 cubes were used for sensory analysis.

Weight loss was calculated from the weight of 40 apple cubes per replicate before and after storage. Weight loss was expressed as a percentage of the initial weight of the cubes. Concentrations of chemical constituents were expressed in terms of both fresh and dry weight in order to show the actual concentration of the chemical constituents in the product, as well as the differences in amounts of those compounds between treatments / replicates that tended to be hidden by differences in water content.

Color of the cut apple surface was measured on 10 apple cubes per replicate with a hand-held tristimulus reflectance colorimeter (Minolta CR-300, Minolta Corp., Ramsey, New Jersey). Three replicates of 10 apple cubes were used for each storage time. Color was recorded using a CIE-L*a*b* uniform color space. Numerical values of a* and b* were converted into hue angle and chroma (Francis 1980).

Fruit firmness was measured by compression of each of the 10 apple cubes used for color measurement with an Instron Universal Testing Instrument (model 4501, Instron Corp., Ohio) (Kader 1982). Results were expressed as the maximum force (Newton) required for a deformation of 5 mm.

Titrateable acidity was expressed as percentage of malic acid and results were expressed in terms of fresh and dry weight. pH was measured in the juice of the crushed apple using a Crison pH meter, model Micro pH 2002.

Sugar analyses (sucrose, D-glucose and D-fructose) were carried out using a high performance liquid chromatograph SP 8800 (Spectra Physics). Soluble solids content (SSC) of the nondiluted juice from crushed apple cubes was determined at 20C with a hand-held refractometer (model Atago - ATC1, Japan) and expressed in terms of fresh and dry weight (%).

Sensory Analysis

Screening and selection of the sensory panel was based on the ability to recognize and determine the intensity of basic tastes, to recognize odor and to rate texture (Stevens and Albright 1980). Fifteen judges were selected from graduate students between 24 and 34 years of age. Samples were presented randomly and were evaluated by these 15 judges at room temperature. Tasting was performed in a sensory testing room with individual booths and controlled lighting (white), with samples presented in plastic transparent boxes. Each panelist was asked to rate three main components of apple quality, color, firmness and flavor and also overall fruit quality, in terms of overall acceptability. A 5-point hedonic scale was used: 1 = dislike extremely; 3 = neither like or dislike and 5 = like extremely. Each sample was evaluated twice.

Statistical Analysis

In order to be able to compare the experiments performed at different dates, differences between the controls in those experiments were taken into account. Results from different treatments were corrected by those differences: Corrected Experimental Result 2 = Experimental Result 2 * Control 1/Control 2; Experimental Result 1 was compared to Corrected Experimental Result 2 and the control used was Control 1. The SAS statistical analysis system package (SAS Institute 1982) was used for analysis of the data. Statistical significance was assessed by two-way analysis of variance (the source of variation was chemical treatment). Significant differences between treatments were detected using Duncan's multiple range test. The overall least significant difference (LSD) ($P=0.05$) was calculated and used to detect significant differences among storage times. To study the relationship between sensory attributes and objective quality measurements, correlation analysis was conducted between both sets of variables. 'r' is the correlation coefficient.

RESULTS AND DISCUSSION

Weight Loss and Firmness

Overall, weight loss was greater in apple cubes stored in air when compared to CA-stored samples for both storage times (Table 1). In order to overcome the effect of weight loss differences, the data for TA, SSC and sugars were expressed in terms of fresh and dry weight in order to illustrate the actual losses that occurred in those constituents irrespective of the concentrating effect imposed by water loss.

TABLE 1.
WEIGHT LOSS (%)^z AND FIRMNESS OF MINIMALLY PROCESSED
APPLE STORED AT 4°C IN AIR AND DIFFERENT CA CONDITIONS

Experimental series	Storage time (days)				
	0	3	3	7	7
	Firmness (N)	Weight loss (%)	Firmness (N)	Weight loss (%)	Firmness (N)
Regular air	52.1a	1.39a ^y	44.6b	3.99a	33.9b
2% O ₂ + 4 % CO ₂	52.1a	0.71bc	45.7a	1.63bc	38.0ab
	52.1a	0.96b	45.9a	2.69b	37.6ab
2% O ₂ + 8% CO ₂	52.1a	0.70bc	46.7a	2.03b	42.7a
	52.1a	1.08b	44.5a	2.40b	40.5a
2% O ₂ + 8% CO ₂ + 42.5 mM Asc. Acid	52.1a	0.87bc	44.7b	1.33c	33.9b
	52.1a	0.54c	43.1b	1.98b	33.6b
2% O ₂ + 12 % CO ₂	52.1a	0.47c	46.1a	1.42c	42.3a
	52.1a	1.01b	45.2a	2.01b	43.0a

^zData are means of 3 replicates of 40 apple cubes each.

^yMeans separation in columns by Duncan's multiple range test, $P \leq 0.05$

When compared to air storage CA conditions reduced the loss of firmness after 3 and 7 days of storage (Table 1). Reduced oxygen and increased carbon dioxide levels were reported by Burton (1974) to affect plant cell breakdown. Bolin and Huxsoll (1989) found that reducing the oxygen level retarded texture loss of peach halves.

However, when 'Jonagored' apple cubes were previously dipped in ascorbic acid, the benefit of storage in 2% O₂ + 8% CO₂ was lost. Also at day 7, 2% O₂ + 4% CO₂ was the least effective of the CA treatments. Firmness of the tissue was not different from the control. It is accepted that CA storage helps to maintain the desirable texture of whole apples (Drake 1993).

Color Assessment

All CA conditions were effective in reducing adverse color changes of apple cubes when compared to air control samples. This benefit seemed to be higher with progressive increases in the concentration of CO₂ in the storage atmosphere. After 7 days of storage, the color of cut apple was preserved most effectively when the highest CO₂ concentration (12%) had been used ($P < 0.05$): higher L* values (light color), lower a* values and higher hue angle (more green color) (Table 2). Drake *et al.* (1993) also found that flesh color of 'Delicious' apples was influenced by the level of CO₂ in the storage atmosphere. Apples from 3% CO₂ were distinctly lighter (higher L* values) in color than apples from 1% CO₂ storage. Nevertheless, this internal color difference was not reflected on Hunter a* and b* values.

In spite of the observed inhibition of browning by CA storage, apple cubes still lost their initial color, regardless of the CO₂ concentration: they lost green pigmentation and lightness during storage, but this loss in color was greater for apple cubes stored in atmospheres with lower CO₂ levels (Table 2).

Experimental results confirm that fresh-cut apple (cv. Jonagored) tolerate and benefit from higher levels of CO₂ compared to those recommended for whole 'Jonagold': 1.5% CO₂ by Lau (1988), <1% CO₂ Stow (1987), 3% CO₂ (Drake 1993). This is in agreement with Kader (1989) and Brecht (1995) who stated that MP fruits have fewer barriers to gas diffusion, being able to tolerate higher concentrations of CO₂ than intact commodities.

Titrateable Acidity and pH

After 7 days of storage, apple cubes treated with ascorbic acid previous to CA storage showed the highest TA (dry weight basis) compared to other CA conditions except when 4% CO₂ was used (Table 3), which might be probably due to the acidic dip prior to storage (Chitarra and Chitarra 1990). After 7 days of storage, apple cubes treated with ascorbic acid previous to CA storage also showed lower pH than those in CA conditions, except when 4% CO₂ was used, and a similar pH of the control (air) samples (Table 3). Carbon dioxide levels higher than 4% CO₂ were associated with higher pH. All pH values were lower than 3.8 which is advantageous in terms of flavor preservation and microbiological contamination.

TABLE 2.
L*, a* AND HUE VALUES^z OF MINIMALLY PROCESSED APPLE STORED AT 4C IN AIR AND DIFFERENT CA CONDITIONS

Experimental series	Storage time (days)								
	0	0	0	3	3	3	7	7	7
	L*	a*	Hue	L*	a*	Hue	L*	a*	Hue
Regular air	77.16a ^y	-5.46ab	104.06de	73.29d	-3.23a	97.16d	71.61c	-2.85a	96.07cd
2% O ₂ + 4% CO ₂	77.16a	-5.48ab	104.73abc	75.75ab	-3.82bc	98.76bc	72.28c	-3.80b	98.11b
	77.16a	-5.18a	103.39de	72.71d	-3.78bc	98.15cd	71.20c	-2.79a	96.22cd
2% O ₂ + 8% CO ₂	77.16a	-5.46ab	104.06cd	74.38cd	-4.02d	98.53bc	73.74b	-3.81b	99.20a
	77.16a	-5.41ab	104.34bc	75.69ab	-3.49ab	97.38d	70.99cd	-2.79a	96.08cd
2% O ₂ + 8% CO ₂ + 42.5 mM AA	77.16a	-5.78b	104.96ab	74.00cd	-3.79bc	100.07a	69.83d	-2.99a	96.03cd
	77.16a	-5.58ab	104.19c	75.18ab	-3.58abc	98.22cd	69.85d	-2.72a	95.48d
2% O ₂ + 12% CO ₂	77.15a	-5.46ab	103.22e	74.40cd	-4.31d	97.30d	76.08a	-4.08b	99.78a
	77.56a	-5.56ab	105.17a	76.51a	-3.94bc	99.40ab	71.98c	-3.54b	96.51c

^z Data are means of 3 replicates of 10 apple cubes.

^y Means separation in columns by Duncan's multiple range test, $P \leq 0.05$

TABLE 3.
TITRATABLE ACIDITY (%)¹ AND pH² OF MINIMALLY PROCESSED APPLE STORED T 4C IN AIR AND DIFFERENT CA CONDITIONS

Experimental series / Storage time (days)	Fresh weight						Dry weight		
	0	0	3	3	7	7	0	3	7
	TTA	pH	TTA	pH	TTA	pH	TTA	TTA	TTA
Regular air	0.28a ^y	3.47a	0.28ab	3.32b	0.27a	3.61b	2.33a	2.30ab	2.16ab
2% O ₂ + 4% CO ₂	0.28a	3.46a	0.26a	3.36ab	0.27a	3.67ab	2.33a	2.15b	2.21ab
	0.26a	3.47a	0.26ab	3.28b	0.28a	3.66ab	2.17a	2.15b	2.16ab
2% O ₂ + 8% CO ₂	0.27a	3.47a	0.31a	3.36ab	0.20b	3.70a	2.25a	2.57a	1.65b
	0.26a	3.46a	0.32a	3.30b	0.22b	3.78a	2.17a	2.64a	1.79b
2% O ₂ + 8% CO ₂ + 42 mM AA	0.27a	3.47a	0.28ab	3.32b	0.30a	3.59b	2.25a	2.31ab	2.47a
	0.27a	3.47a	0.29ab	3.33b	0.29a	3.63b	2.25a	2.40ab	2.37a
2% O ₂ + 12% CO ₂	0.26a	3.48a	0.31a	3.37ab	0.22b	3.73a	2.17a	2.57a	1.81b
	0.27a	3.46a	0.29a	3.43a	0.24ab	3.78a	2.25a	2.39ab	1.96b

²Data are means of 3 replicates of 10 apple cubes.

^yMeans separation in columns by Duncan's multiple range test, $P \leq 0.05$

Results reported in the literature indicate that, in general, whole apples retain higher acidity in low-O₂ atmospheres. Ben-Arie *et al.* (1991) found that TA of apples (cv. Jonagold) was higher in fruit stored in 3% O₂ than in 15% O₂, irrespective of the CO₂ concentration. Ke *et al.* (1991) reported that pH of 'Yellow Newton' apples was not consistently influenced by the low O₂ levels (0.02 or 0.25%) at 0 or 5°C. However, these CA conditions decreased acidity and increased slightly the pH of 'Granny Smith' apples which is in agreement with the results obtained in this work (Table 3). Drake (1993) reported that TA of several apple varieties in CA storage was higher than air-stored apples. Herregods and Goffings (1993) reported that lowering the O₂ concentration to 1% produced more acid in apples in comparison to air storage (cv. Jonagold) after 6 months at 1°C.

Sugars and Soluble Solids

As fructose is the major sugar in apple, differences between treatments were noticed mainly for this sugar (Table 4). Overall, the atmosphere conditions did not affect either the sucrose or glucose contents of the apple cubes (data not shown).

After 3 days apple cubes stored in either 2% O₂ plus 4% CO₂ or 12% showed higher fructose content than air-stored samples and apples stored at 2% O₂ + 8% CO₂ (Table 4). However, after 7 days no differences were detected between apple cubes from different CA conditions. Drake and Eiselle (1994) reported that total carbohydrates were similar for apples stored in CO₂ concentrations of 5% or higher.

After 7 days all CA-stored samples, except those stored in 2% O₂ + 8% CO₂ had higher SSC than the control samples (Table 4) which is in agreement with CA effects on fructose content.

Sensory Analysis

After 3 days of storage, the beneficial effect of CA storage in higher CO₂ levels on color was observed by panelists (Table 5). Apple cubes from the 2% O₂ + 12% CO₂ or 8% CO₂ + AA treatments obtained the highest scores. Considering the limit of acceptability (score = 3), only these conditions were effective in preserving apple color for 3 days (Table 5). The scores obtained were lower than the initial value (day zero). After 7 days of storage, fruit from none of the CA conditions received a score above 3 with respect to color evaluation (Table 6). Nevertheless, apples from the 2% O₂ + 12% CO₂ atmosphere achieved the highest score, just close to the limit of acceptability.

TABLE 4.
FRUCTOSE CONTENT (%)^z AND SOLUBLE SOLIDS CONTENT (SSC)^z OF MINIMALLY PROCESSED APPLE STORED AT 4C IN AIR
AND DIFFERENT CA CONDITIONS

Experimental series / Storage time (days)	Fresh weight						Dry weight					
	0	0	3	3	7	7	0	0	3	3	7	7
	Fruct.	SSC	Fruct.	SSC	Fruct.	SSC	Fruct.	SSC	Fruct.	SSC	Fruct.	SSC
Regular air	5.28a ^y	10.1a	5.91b	9.9a	6.90b	10.4a	43.8a	83.81a	48.56b	81.36b	55.21b	83.21b
2% O ₂ + 4% CO ₂	5.78a	10.0a	6.96a	10.2a	7.11b	10.3a	47.90a	82.00a	57.59a	84.40a	58.28a	84.43ab
	5.78a	10.0a	6.57a	9.9a	7.23ab	10.3a	47.90a	82.00a	54.22a	81.71b	58.63a	83.52b
2% O ₂ + 8% CO ₂	5.78a	10.1a	5.69b	10.1a	6.73b	9.3a	47.90a	83.81a	47.83b	83.58a	55.52b	76.73b
	5.78a	10.1a	5.96b	10.3a	7.09b	9.0a	47.90a	83.81a	47.65b	84.91a	57.67ab	73.20b
2% O ₂ + 8% CO ₂ + 42.5 mM AA	5.52a	9.6a	4.58b	10.3a	7.26ab	10.5a	45.82 a	79.68a	37.83c	85.09a	59.70a	86.34a
	5.44a	11.0a	5.02b	10.2a	7.95a	10.7a	45.15a	83.81a	41.61bc	84.54a	64.94a	87.40a
2% O ₂ + 12% CO ₂	5.80 a	9.8a	6.80a	10.2a	7.63a	10.3a	48.14a	81.34a	56.40a	84.60a	62.68a	84.61ab
	5.78a	9.6a	6.10a	9.7a	7.18b	11.0a	47.90a	79.68a	50.32a	80.02b	58.63a	89.82a

^zData are means of 3 replicates of 10 apple cubes.

^yMeans separation in columns by Duncan's multiple range test, $P \leq 0.05$

TABLE 5.
SENSORY QUALITY RATING (1-5)^z OF MINIMALLY PROCESSED APPLE AFTER
3 DAYS AT 4C IN AIR AND DIFFERENT CA CONDITIONS

Sensory parameters	Color	Firmness	Flavor	Overall liking
Regular air	1.87b ^y	3.87a	3.47a	3.20a
2% O ₂ + 4% CO ₂	1.87b	4.03a	3.40a	3.47a
	2.00b	3.80a	3.17a	3.20a
2% O ₂ + 8% CO ₂	2.00b	4.20a	3.57a	3.07a
	2.00b	3.87a	3.60a	3.73a
2% O ₂ + 8% CO ₂ + 42.5 mM AA	3.07a	2.93b	2.73b	2.67b
	3.13a	2.80b	2.50b	3.33a
2% O ₂ + 12% CO ₂	3.20a	3.93a	3.33a	3.07a
	3.33a	3.27a	3.67a	3.40a
Day zero	4.60	4.67	4.27	4.67

^z Data are means of 15 judges' evaluation of 3 replicates of 30 apple cubes.

^y Mean separation in columns by Duncan's multiple range test, $P \leq 0.05$

TABLE 6.
SENSORY QUALITY RATING (1-5)^z OF MINIMALLY PROCESSED APPLE AFTER
7 DAYS AT 4C IN AIR AND DIFFERENT CA CONDITIONS

Sensory parameters	Color	Firmness	Flavor	Overall liking
Regular air	1.83b ^y	3.47a	3.07a	2.87ab
2% O ₂ + 4% CO ₂	1.80b	3.47a	3.20a	2.87ab
	1.87b	3.40a	3.00a	3.07a
2% O ₂ + 8% CO ₂	1.73b	3.87a	3.00a	3.13a
	1.60b	3.20a	3.20a	3.20a
2% O ₂ + 8% CO ₂ + 42.5 mM AA	2.07b	2.27b	3.07a	3.13a
	1.87b	2.73b	3.00a	3.13a
2% O ₂ + 12% CO ₂	2.73a	3.20a	2.67a	2.20b
	2.80a	3.13a	2.93a	3.00a
Day zero	4.60	4.67	4.27	4.67

^z Data are means of 15 judges' evaluation of 3 replicates of 30 apple cubes.

^y Mean separation in columns by Duncan's multiple range test, $P \leq 0.05$

No differences were detected by the panelists between firmness of air-stored and CA-stored apple cubes. Fruit treated with AA received a score lower than all the other treatments and in relation to the control (Tables 5 and 6). When storage was preceded by the ascorbic acid dip an increase in firmness loss was reported. This may be explained by the observed water soaked appearance of chemically treated samples, as reported in previous work (Rocha *et al.* 1998).

No differences were detected by panelists in relation to the flavor of air-stored and CA-stored samples after 3 and 7 days of storage (Tables 5 and 6). Drake and Eiselle (1994) also reported no differences in apple flavor (cvs. Bisbee, Red Chief and Oregon Spur Delicious) from CA (1% O₂ + 1% CO₂) established after 10 or 15 days in refrigerated storage when compared to air-stored samples. Only those apple cubes treated with AA received a lower score for flavor than those from other conditions after 3 days of storage. It is likely that the chemical dip imparted an acidic taste to the apple cubes.

When asked to rate the overall liking of the samples, panelists were not able to detect differences between CA-stored and air-stored apples after 3 days storage, except for apple cubes from 2% O₂ + 8% CO₂ + AA dip which received lower score in the first experiment (Table 5). After 7 days no differences were detected by panelists between air- and CA-stored samples (Table 6).

From the sensory analysis of results, only CA storage in 12% CO₂ was more effective than air storage and only in preserving the color of cut apple during 3 days at 4C.

Relationship Between Sensory Analysis and Objective Quality Parameters

Overall, data from physical and sensory evaluation of color were found to be correlated in all experiments except for 2% O₂ + 12% CO₂ atmosphere storage and for L* values in 2% O₂ + 4% CO₂ in the 1st series. However, this was not confirmed in the 2nd series. Good correlation ($r \geq 0.93$) were found between data from physical and sensory evaluations for all color parameters except for L* values in 2% O₂ + 8% CO₂ in the 2nd series (Table 7).

Data from physical and sensory evaluations of firmness were highly correlated ($r \geq 0.98$) for samples from 2% O₂ + 4% CO₂ and 2% O₂ + 8% CO₂ + AA in the 1st series (Table 7). However, poor correlation were found in the experiments with 2% O₂ + 8% or 12% CO₂. In the 2nd series, high correlation were found in all experiments.

No significant correlation were found between chemical evaluations of TA or pH and sensory evaluations of flavor in any experiment in either series.

Fructose content was found to be correlated ($r \geq 0.93$) with sensory evaluation of flavor in the experiments with 2% O₂ + 12% or 4% CO₂ of the 1st series, and in all experiments except 2% O₂ + 8% + AA of the 2nd series (Table 7).

TABLE 7.
CORRELATION COEFFICIENTS (r) CALCULATED FOR PHYSICOCHEMICAL CHARACTERISTICS AND SENSORY ATTRIBUTES OF MINIMALLY PROCESSED APPLE STORED AT 4C IN AIR AND DIFFERENT CA CONDITIONS

Quality parameters	Regular air	2% CO ₂ + 4% O ₂	2% CO ₂	2% O ₂ + 8% CO ₂	2% O ₂ + 8%O ₂ + AA	2% O ₂ + 12% CO ₂			
Color L*	0.87	0.58	0.96	0.81	0.59	0.96	0.90	0.42	0.91
a*	0.96	0.99	0.89	0.85	0.98	0.99	0.97	0.66	0.86
Hue	0.96	0.99	0.95	0.97	0.99	0.99	0.98	0.64	0.95
Firmness	0.99	0.96	0.99	0.67	0.96	0.97	0.97	0.51	0.98
Flavor									
TA	0.99	0.40	0.31	0.14	0.20	-	0.34	0.10	0.44
pH	-	0.29	-	0.17	0.48	-	0.36	0.32	0.24
Fructose	0.90	0.93	0.85	0.38	0.88	-	0.41	0.99	0.95
Soluble solids	0.19	0.99	0.12	0.46	0.67	0.81	0.32	0.96	0.86

(-) correlation coefficient not significant
r significant at a level $P \leq 0.05$

The 2nd, 4th, 6th and 8th columns are the results of the 1st series. The 3rd, 5th, 7th, and 9th columns are the results of the 2nd series.

Soluble solids content was found to be correlated with sensory evaluations of flavor in the experiments with 2% O₂ + 12% CO₂ and 2% O₂ + 4% CO₂ of the 1st series and well correlated in the experiment with 2% O₂ + 12% CO₂ of the 2nd series (Table 7).

CONCLUSIONS

Controlled atmosphere (4% to 12% CO₂ + 2% O₂) storage was found to be advantageous over air storage in terms of overall preservation of fresh-cut apple (cv. Jonagored). After 3 days, CA-stored apple cubes were firmer, showed better color and higher content of fructose and soluble solids than air-stored cubes. In relation to color preservation this benefit was increased

when the CO₂ in the atmosphere was increased to 12% CO₂. Controlled atmosphere storage showed no advantage over air storage in relation to sensory evaluation of firmness and flavor.

In spite of the reported benefits of CA storage, no complete inhibition of degradative reactions was achieved particularly as regards browning of the apple surface. Considering that enzymatic browning mediated by PPO has been extensively reported as the main factor involved in apple browning, and since the storage conditions tested (low temperature, low pH, low O₂, high CO₂) were supposed to reduce the activity of the enzyme, a characterization of the specific enzyme is recommended in order to contribute for the elucidation of the mechanisms by which gas concentration may affect apple color.

ACKNOWLEDGMENT

This research was funded by a JNICT scholarship (BD 2109/92-IF).

REFERENCES

- BALDWIN, E.A., NISPEROS-CARRIEDO, M.O. and BAKER, R.A. 1995. Edible coatings for lightly processed fruits and vegetables. *HortScience* 30, 35-38.
- BARRETT, D.M., LEE, C.Y. and LIU, F.W. 1991. Changes in 'Delicious' apple browning and softening during controlled atmosphere storage. *J. Food Quality* 14, 443-453.
- BEN-ARIE, R., LEVINE, A., SONEGO, L. and ZUTKHI, Y. 1991. Differential effects of CO₂ at low and high O₂ on the storage quality of two apple cultivars. *Acta Horticulturae* 287, 75-83.
- BOLIN, H.R. and HUXSOLL, C.C. 1989. Storage stability of minimally processed fruit. *J. Food Processing Preservation* 13, 281-292.
- BRECHT, J.K. 1995. Physiology of lightly processed fruits and vegetables. *HortScience* 30, 18-21.
- BURTON, W.G. 1974. Some physiological principles underlying the controlled atmosphere storage of plant materials. *Ann. Appl. Biol.* 78, 162-167.
- CHEN, P.M., VARGA, D.M., MIELKE, E.A. and DRAKE, S.R. 1989. Poststorage behaviour of apple fruit after low oxygen storage as influenced by temperatures during storage and in transit. *J. Food Sci.* 54, 993-996.
- CHITARRA, M.I.F. and CHITARRA, A.B. 1990. *Pós colheita de frutos e hortaliças. Fisiologia e manuseio*, pp. 272-278, ESAL/FAEPE, Lavras.
- DRAKE, S.R. 1993. Short-term controlled atmosphere storage improved quality of several apple cultivars. *J. Am. Soc. Hort. Sci.* 118, 486-489.

- DRAKE, S.R. and EISELLE, T.A. 1994. Influence of harvest date and controlled atmosphere storage delay on the color and quality of 'Delicious' apples stored in a purge-type controlled atmosphere environment. *Hort-Technology* 4, 260-263.
- DRAKE, S.R., EISELLE, T.A. and WAELTI, H. 1993. Controlled atmosphere storage of 'Delicious' apples in high and variable carbon dioxide. *J. Food Processing Preservation* 17, 177-189.
- FRANCIS, F.J. 1980. Color quality evaluation of horticultural crops. *Hort-Science* 15, 58-59.
- HERREGODS, M. and GOFFINGS, G. 1993. The storage of 'Jonagold' apples in U.L.O. circumstances. *Acta Horticulturae* 343, 76-82.
- HUXSOLL, C.C., BOLIN, H.R. and KING JR., A.D. 1989. Physicochemical changes and treatments for lightly processed fruits and vegetables. In *Quality Factors of Fruits and Vegetables*, ACS Symposium Series 405, (J.J. Jen, ed.) pp. 203, American Chemical Society, Washington, DC.
- JANOVITZ-KLAPP, A.H., RICHARD, F.C., GOUPY, P.M. and NICOLAS, J.J. 1990. Inhibition studies on apple polyphenoloxidase. *J. Agric. Food Chem.* 38, 926-931.
- KADER, A.A. 1982. Proper units for firmness and abscission force data. *HortScience* 17, 707.
- KADER, A.A. 1989. Modified atmosphere packaging of fruits and vegetables. *Crit. Rev. Food Sci. Nutr.* 28, 1-30.
- KE, D., RODRIGUEZ-SINOBAS, L. and KADER, A.A. 1991. Physiology and prediction of fruit tolerance to low-oxygen atmospheres. *J. Am. Soc. Hort. Sci.* 116, 253-260.
- KNEE, M. 1980. Physiological responses of apple fruits to oxygen concentrations. *Ann. Appl. Biol.* 96, 243-253.
- LABUZA, T.P. and BREENE, W.M. 1989. Applications of 'active packaging' for improvement of shelf-life and nutritional quality of fresh and extended shelf-life foods. *J. Food Processing Preservation* 13, 1-69.
- LAU, O.L. 1988. Harvest indices, dessert quality, and storability of 'Jonagold' apples in air and controlled atmosphere storage. *J. Am. Soc. Hort. Sci.* 113, 564-569.
- LAU, O.L. and LOONEY, N.E. 1982. Improvement of fruit firmness and activity in controlled-atmosphere-stored 'Golden delicious' apples by a rapid O₂ reduction procedure. *J. Am. Soc. Hort. Sci.* 107, 531-534.
- LAU, O.L. and YASTREMSKI, R. 1991. Retention of quality of 'Golden Delicious' apples by controlled- and modified-atmosphere storage. *HortScience* 26, 564-566.
- NICOLAS, J.J., RICHARD-FORGET, F.C., GOUPY, P.M., AMIOT, M.J. and AUBERT, S.Y. 1994. Enzymatic browning reactions in apple and apple products. *Crit. Rev. Food Sci. Nutr.* 34, 109-157.

- NICOLI, M.C., ANESE, M. and SEVERINI, C. 1994. Combined effects in preventing enzymatic browning reactions in minimally processed fruit. *J. Food Quality* 17, 221-229.
- PIZZOCARO, F., TORREGGIANI, D. and GILARDI, G. 1993. Inhibition of apple polyphenoloxidase by ascorbic acid, citric acid and sodium chloride. *J. Food Processing Preservation* 17, 21-30.
- ROCHA, A.M.C.N., BROCHADO, C.M. and MORAIS, A.M.B.M. 1998. Influence of chemical treatment on quality of minimally processed apple (cv. Jonagored). *J. Food Quality* 21, 13-28.
- ROUET-MAYER, M.A., RALAMBOSOA, J. and PHILIPPON, J. 1989. Reversibility of enzymic browning of frozen fruits and vegetables. In *Freezing and Refrigeration of Fruits and Vegetables*. Proceedings of the International Conference on Technical Innovations. pp. 147-150, University of California, Davis.
- SAPERS, G.M. and HICKS, K.B. 1989. Inhibition of enzymatic browning in fruits and vegetables. In *Quality Factors of Fruits and Vegetables*, ACS Symposium Series 405, (J.J. Jen, ed.) pp. 29, American Chemical Society, Washington, DC.
- SAS Institute, Inc. 1982. *SAS User's Guide: Statistics*, SAS Institute, Cary, NC.
- SFAKIOTAKIS, E., NIKLIS, N., STAVROULAKIS, G. and VASSILIADIS, T. 1993. Efficacy of controlled atmosphere and ultra low ethylene storage on keeping quality and scald control of 'Starking Delicious' apples. *Acta Horticulturae* 347, 83-94.
- STEVENS, M.A. and ALBRIGHT, M. 1980. An approach to sensory evaluation of horticultural commodities. *HortScience* 15, 48-50.
- STOW, J. 1987. Storage of 'Jonagold' apples. *Scientia Horticulturae* 31, 245-251.
- WARDOWSKI, W.F. and BROWN, G.E. 1991. Postharvest decay control recommendations for Florida citrus fruit. Circular 952. Institute of Food and Agricultural Sciences. University of Florida, Gainesville.
- WILEY, R.C. 1994. Preservation methods for minimally processed refrigerated fruits and vegetables. In *Minimally Processed Refrigerated Fruits and Vegetables*, pp. 66, Chapman and Hall, New York.