Effect of Controlled Atmospheres on Maintaining Quality of Persimmon Fruit cv. “Rojo Brillante”

L. Arnal, C. Besada, P. Navarro, and A. Salvador

ABSTRACT: Astringent “Rojo Brillante” persimmon fruits were stored in air or in 2 different controlled atmospheres: 10% CO2 + 90% N2 (CA1) or 97% N2 + air (CA2) for up to 50 d at 15 °C. After different periods, the fruits were treated with 95% CO2 for 24 h at 20 °C in order to remove astringency, and then transferred to 20 °C in air free of CO2 for 5 d to simulate shelf life. Other fruits were directly transferred to shelf life without being submitted to deastringency treatment. Storage under CA2 allowed storability of persimmon “Rojo Brillante” during 30 d at 15 °C, maintaining commercial firmness. Moreover, CA2 had an effect on removing astringency when fruits were stored for 30 d at 15 °C, or after 20 d following to shelf life. As consequence, deastringency treatment could be avoided when the fruits were previously stored under this controlled atmosphere.

Keywords: astringency, controlled atmospheres, firmness, persimmon, storage

Introduction

“Rojo Brillante” is the most important persimmon cultivar of the Mediterranean area, whose production is increasing in the last years. Fruit from this variety has an excellent size and flavor but is astringent at harvest due to high levels of soluble tannins, and a deastringency treatment before fruit commercialization is necessary. Exposure of fruit to high concentration of CO2 (95% to 98%) during 24 h is a well-known postharvest method in this cultivar to remove astringency, and it is used as a common commercial practice to market nonastringent persimmons with high firmness (Arnal and Del Río 2003). This deastringency treatment is based on the fact that soluble tannins are polymerized by acetaldehyde, produced under anaerobic conditions, to form an insoluble compound, which is not astringent (Matsuo and Itoo 1982).

The high level of persimmon production observed in the last years makes necessary prolonged fruit storage. A point to consider is that “Rojo Brillante” persimmons develop chilling injury symptoms such as firmness loss or flesh gelling when exposed to temperatures below 11 °C (Arnal and Del Río 2004a). As a consequence, “Rojo Brillante” is usually stored at temperatures close to 15 °C. However, at this temperature, prolonged storage is not possible because quality loss can be excessive, flesh softening being the principal limiting factor during storage life of this cultivar (Arnal and Del Río 2004a; Salvador and others 2005).

Under controlled atmosphere (CA), storage of some persimmon cultivar such as “Fuyu” can be prolonged considerably. In this cultivar, CA allows preservation of fruit with lower disorder incidence and higher firmness (Tanaka and others 1971; Brackmann and others 1997; Park 1999; Brackmann and Donazzolo 2001). CA-storage was also shown to retard softening of cv. “Triumph” and to reduce flesh firmness, astringency, acetaldehyde, ethanol, total soluble solids (Guellat-Reich and others 1975; Ben-Arie and others 1991). The effects of CA, such as delay senescence, decreased respiration rate, and ethylene production, depend on fruit variety or cultivar, physiology state, atmosphere composition, storage temperature, and storage time (Kader 2002). Currently, there is no information about the effect of CA-storage on the persimmon cultivar “Rojo Brillante.”

The aim of this study was to evaluate the effect of 2 different control atmospheres on maintaining quality and on removing astringency of “Rojo Brillante” persimmon during storage at 15 °C.

Materials and Methods

Fruit source and storage procedure

Persimmon fruits cv. “Rojo Brillante” were harvested in L’Alcudia (Valencia) and transported to Technidex S.A Co. (Valencia, Spain), where fruits were carefully selected for uniformity of size and color development. Afterward, the fruits were separated in lots according to the following controlled atmosphere (CA) treatments in triplicate: air or control (CTL), 10% CO2 + 90% N2 (CA1), and 97% N2 + air (CA2). The fruits were always maintained at 15 °C with 85% to 90% R.H. After 20-, 30-, 40-, and 50-d storage, a sample of fruit from each replicate was transported to Inst. Valenciano de Investigaciones Agrarias (IVIA), and submitted to deastringency treatment. Subsequently, the fruits were transferred to 20 °C in air free of CO2 for 5 d to simulate shelf life period.

Deastringency treatment was carried out in closed containers that contained 95% CO2 for 24 h at 20 °C (90% R.H.) by passing a stream of air containing 95% CO2 through the containers.

Other samples of CA-treatments were transferred directly from 15 °C to shelf life conditions, without being submitted previously to deastringency treatment. These treatments were named “direct commercialization” (DC) treatments (DC-CA1 and DC-CA2).

Fruit quality assessment

After storage periods at 15 °C and after subsequent shelf life, flesh firmness, astringency, acetaldehyde, ethanol, total soluble
Controlled atmospheres on quality persimmon... solids, pH, external color, respiration rate, ethylene production, sensory evaluation, and external and internal disorders were evaluated.

Flesh firmness was determined over 20 fruits per replicate, with a Texturemeter Instron Universal Machine model 4301 (Instron Corp., Canton, Mass., U.S.A.), using an 8-mm plunger after epicarp removal, at 2 equidistant locations in the equatorial region of each fruit. The crosshead speed during the firmness testing was 10 mm/min. Data were expressed as the maximum force in Newtons (N) required to break the flesh.

Astringency level of fruit was estimated by soluble tannins (ST) analysis, tannin content index (TI) determination, and sensory evaluation.

Many of 15 fruits per replicate were divided in 3 samples and cut in four longitudinal parts. Two of the opposite parts were sliced and frozen (–20 °C) to determine ST, which was evaluated using the Folin–Denis method as described by Arnal and Del Río (2004b) and expressed as percentage or g/100 g EW. To determine TI (Gorini and Testoni 1988), 5 fruits were visually evaluated from each replicate. Soluble tannins, which are responsible for persimmon astringency, are compounds that react with ferric chloride, forming tannin–Fe ion complexes, which are blue–black. The degree of astringency can be estimated in persimmon fruit by evaluating color development during the reaction. Persimmon fruits were cut equatorially and after 15 min the freshly cut surface was immersed in a 5% FeCl$_3$ solution for 3 min. After another 3 min out of the solution, the color of freshly cut surface was visually evaluated from 1 (less color development or minimum tannin content, no astringency) to 5 (maximum color development or maximum tannin content, high astringency).

Sensory evaluation was performed at the sensory Laboratory of the Postharvest Dept. of the IVIA in composite samples of 5 fruits from each replicate previously peeled and sliced. Eight to 10 semi-trained judges were asked to evaluate astringency and the presence of off-flavors. The judges were persons familiar with cultivar "Rojo Brillante" who were tasting fruits with different levels of astringency for several years. A 4-point scale was used for astringency, where 1 = very high astringency and 4 = no astringency. Each sample consisted of segments taken from about 5 individual fruits. Samples were presented to panelists in trays labeled with 3-digit random codes and served at room temperature (25 ± 1 °C). The panelists had to taste several segments of each fruit in order to compensate, as far as possible, for biological variation of the material. Milk was provided for palate rinsing between samples.

The opposite parts of the fruit, which were not used to measure astringency, were placed in an electric juice extractor (model 753, Moulinex, Spain) and filtered through cheesecloth; obtained juice was used to determine acetaldehyde and ethanol production, total soluble solids (TSS), and pH.

Acetaldehyde and ethanol production were measured on 3 samples per replicate of juice samples, obtained as mentioned previously and analyzed by headspace gas chromatography (Ke and Kader 1990). Five milliliters of the filtered juice were transferred to 10 mL vials with crimp-top caps, sealed with TFE/silicone septa, and frozen (–20 °C) until analysis. For the analysis, the samples were put in a water bath at 20 °C for 1 h, followed by heating at 60 °C for 10 min. A 1-mL sample of the headspace was withdrawn from the vials and injected in the gas chromatograph (Perkin-Elmer, Model 2000, Norwalk, Conn., U.S.A.), provided with flame ionization detector (FID) and 0.32 cm × 1.2 m Porapak QS 80/100 column. The injector was set at 175 °C, the column at 150 °C, the detector at 200 °C, and the carrier gas at 12.3 psi. Ethanol and acetaldehyde were identified by comparison of retention times with those of a standard solution. The results were expressed as mg/100 mL.

TSS were measured twice from 3 juice sample per replicate with a digital refractometer (model PR1, Atago, Tokyo, Japan) and the results were expressed as °Brix. Measures of pH were done with a pH-meter (model C231, Consort, Belgium).

Skin color was evaluated by a Minolta Colorimeter (Model CR-300, Ramsey, N.Y., U.S.A.) on samples of 20 fruit. L, a, b Hunter parameters were measured, and the results were expressed as skin color index as described by Jiménez-Cuesta and others (1981); color index = ($1000a$)/($Lb$).

Ethylene and carbon dioxide production rates were measured in 3 replicates of 2 fruits per replicate. The fruits were weighed and sealed in 2-L glass jars for 2 h at 20 °C. Ethylene and carbon dioxide were analyzed by injecting 3 samples per replicate of 1 mL of headspace into a gas chromatograph (Perkin Elmer), as described by Salvador and others (2005). CO$_2$ production was expressed as mL CO$_2$/kg/h and ethylene production as μL C$_2$H$_4$/kg/h.

External or internal browning was visually assessed as slight (<25% of the affected area), medium (25% to 50% of the affected area), or severe (>50% of the affected area), in samples of 50 fruits per replicate.

### Statistical analysis

Data were subjected to analysis of variance (ANOVA) and multiple comparisons between means were determined with the LSD test (P < 0.05) using the Statgraphics Plus 5.1 (Manugistics Inc., Rockville, Md., U.S.A.).

#### Results

During storage at 15 °C, fruit firmness gradually decreased, mainly in control and CA1 fruit (Figure 1A). CA2 fruit showed the highest firmness values after 20 and 30 d of storage, but no significant differences among treatments were observed after 40 d. When shelf life was simulated (Figure 1B), an important fruit firmness loss was observed. After 20 d of storage plus shelf life, all treatments presented firmness close to 10 N, a value that is considered the commercial limit in this cultivar (Arnal and Del Río 2004a; Salvador and others 2004). When the fruits were stored 30 d, only fruits from CA1 and CA2 not submitted to deastringency treatment (DC-C1A and DC-CA2) maintained commercial firmness (10N); after 40 d all treatments overcame this limit.

Just after harvest, the fruits showed a high level of soluble tannins (ST) responsible for their astringency. In CA2 fruits, an important reduction of ST was presented during storage at 15 °C (Figure 2A): so after 30 d of storage, ST values were close to 0.02%. CT and CA1 fruits did not show a reduction in the ST values. Although the CA1 values were lower than CTL ones, significant differences were not shown. Deastringency with CO$_2$ sharply reduced the ST level in all treatments (Figure 2B). Nevertheless, it is important to note that the reduction of ST in CTL fruits was lower when storage time at 15 °C increased; this fact manifests as a loss in the effectiveness to remove astringency with time storage. This effect was not observed in CA1 and CA2 fruits. CA1 fruits not submitted to deastringency (DC-CA1) showed a little ST decrease, always presenting much higher values than fruit submitted to deastringency treatment. No differences were observed between CA2 and DC-CA2 after shelf life period.

Changes in fruit astringency showed by tannin content index (TI) determination (Table 1) are in agreement with ST results explained previously. At 15 °C storage, CA2 fruits presented a reduction in TI values compared to CT and CA1. CA1 fruits presented lower TI values than CTL fruit, but no significant differences were
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Controlled atmospheres on quality persimmon... found after 20 d. The efficacy of deastringency treatment was supported by diminishing of the TI showed after deastringency treatment plus shelf life.

In the sensorial assessments, CTL fruit was evaluated as very high astringent, with values of 1, during the whole storage at 15 °C (Table 2). However, CA2 fruit showed an important astringency loss, reaching values of 4 (no astringency) from 30 d of storage at 15 °C. CA1 fruit showed a slight astringency loss throughout storage, significantly slower than CA2 fruit. After shelf life, the lower sensorial values showed by control compared to CA1 and CA2 corroborated the descent in the efficacy of CO2 treatment to remove astringency mentioned above. No significant differences between DC-CA2 and CA2 were observed, exhibiting values of 4 throughout storage. Sensory evaluation of DC-CA1 fruit did not show values of no-astringent (4) after any studied period.

Low acetaldehyde production was shown in CTL and CA1 fruit during storage at 15 °C (Figure 3A). In CA2, fruit production of this volatile increased considerably from 20 d of storage where high values were showed. Deastringency treatment after storage resulted in an increase of acetaldehyde values in CTL and CA1 fruits (Figure 3B). Nevertheless, in CTL fruit, the acetaldehyde values decreased sharply after 40 d, which also corroborated the decrease in the efficacy of CO2 treatment to remove astringency. In CA2, acetaldehyde production slightly increased, overcoming the high values registered during storage at 15 °C. DC-CA1, which was not treated with CO2, maintained low acetaldehyde values after shelf life; meanwhile DC-CA2 presented similar values to CA2 after CO2 treatment.

Ethanol production at harvest was very low (0.67 mg/100 mL). Only in CA2 fruit an important increase during storage at 15 °C was shown, reaching values close to 120 mg/100 mL after 40 d of storage (data not shown). The values shown at 15 °C remained constant after deastringency treatment, without relevant differences between CA2 and DC-CA2. After 20 d of storage, CTL fruit showed similar TSS values with respect to values at harvest (18.3 °Brix) (data not shown), but CA1 and CA2 showed lower TSS close to 17.5 and 15 °Brix, respectively. During storage at 15 °C, no relevant changes were observed. After shelf life simulation, CTL and CA1 fruits submitted to deastringency treatment presented a decrease in TSS values (16 °Brix) and values of CA2 fruit remained constant. This reduction was not observed in fruit not submitted to deastringency treatments (DC treatments). During storage at 15 °C, pH values remained constant and without differences with respect to those shown at harvest (5.9) (data not shown). After shelf life period, minor changes were shown, and no relevant differences were observed among treatments.

During storage at 15 °C, the color index of control fruit increased with respect to values at harvest (12.13), reaching values close to 33.87 at the end of storage (data not shown). But, this parameter remained constant throughout storage time when fruits were stored under CA. After shelf life simulation, color index increased in all treatments (Figure 4). No statistical differences between fruit submitted to CA conditions (CA1, CA2, DC-CA1, and DC-CA2) were shown, but they were significantly different from CTL fruit, which showed the highest values.

CO2 production at harvest was 6.02 mL/kgh. High CO2 values, close to 30 mL/kgh, were exhibited at the beginning of storage...
in CA1 (data not shown), but slowly decreased throughout time storage. Lower increase was shown in CTL and CA2 fruits, which achieved maximum values of 13 and 10 mL/kg-h, respectively. After shelf life, a reduction of CO$_2$ was shown in CA1 fruit, values achieving 11.53 mL/kg-h; as consequence, the differences between treatments decreased notably. Ethylene production during storage and after shelf life was very low (from 0 to 0.17 μL/kg-h) in all treatments tested, without remarkable differences (data not shown).

Neither development of internal or external disorders nor other visual damages were observed in fruit after treatments periods.

### Discussion

In persimmon fruit cv. “Rojo Brillante,” maintenance of fruit quality during storage mainly depends on the rate of flesh softening. Therefore delaying softening during storage is an important aim in postharvest persimmon research. In this experiment, the effect of 2 CA on fruit quality during storage at 15 °C was evaluated. From previous experiments with “Rojo Brillante” persimmon, firmness values lower than 10 N force have been considered as not acceptable from the commercial point of view (Salvador and others 2004). In the present study, flesh softening during storage and subsequent shelf life was markedly reduced in persimmon stored under CA2 conditions; this fruit maintained firmness values superior to the commercial limit up to 30-d storage plus shelf life. Meanwhile, no differences were shown between firmness fruit in CA1 and control fruit, which did not overcome commercial firmness values from 20 d plus shelf life.

#### Table 1 — Tannin content index of persimmon fruit cv.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Days at 15 °C</th>
<th>Days at 15 °C + 5 d at 20 °C</th>
</tr>
</thead>
<tbody>
<tr>
<td>CTL</td>
<td>3.0b</td>
<td>2.0b</td>
</tr>
<tr>
<td>CA1</td>
<td>2.8b</td>
<td>1.7b</td>
</tr>
<tr>
<td>CA2</td>
<td>1.0a</td>
<td>1.1a</td>
</tr>
</tbody>
</table>

#### Table 2 — Sensory evaluation of astringency on persimmon fruit cv.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Days at 15 °C</th>
<th>Days at 15 °C + 5 d at 20 °C</th>
</tr>
</thead>
<tbody>
<tr>
<td>CTL</td>
<td>1.0a</td>
<td>1.0a</td>
</tr>
<tr>
<td>CA1</td>
<td>1.0a</td>
<td>2.0b</td>
</tr>
<tr>
<td>CA2</td>
<td>3.0c</td>
<td>4.0d</td>
</tr>
</tbody>
</table>

**“Rojo Brillante” during storage at 15 °C in air (CTL) or in controlled atmospheres, 10% CO$_2$ + 90% N$_2$ (CA1) or 97% N$_2$ + air (CA2), and after being treated or not (DC-CA1, DC-CA2) to remove astringency, following 5 d at 20 °C.** Means for storage at 15 °C or for shelf life period with the same letter are not significantly different at 5% level (LSD test). Absented measures due to the lost in fruit quality. Tannin content index at harvest: 3. Tannin content index values from 1 (minimum tannin content) to 5 (maximum tannin content).

**“Rojo Brillante” during storage at 15 °C in air (CTL) or in controlled atmospheres, 10% CO$_2$ + 90% N$_2$ (CA1) or 97% N$_2$ + air (CA2), and after being treated or not (DC-CA1, DC-CA2) to remove astringency, following 5 d at 20 °C (B). Vertical bars represent LSD intervals (P = 0.05). Acetaldehyde production at harvest: 0.19 mg/100 mL. Absented measures after 50-d storage are due to the lost in fruit quality.**

Figure 3 — Acetaldehyde production of persimmon cv. “Rojo Brillante” during storage at 15 °C in air (CTL) or in controlled atmospheres, 10% CO$_2$ + 90% N$_2$ (CA1) or 97% N$_2$ + air (CA2) (A), and after being treated or not (DC-CA1, DC-CA2) to remove astringency, following 5 d at 20 °C (B). Vertical bars represent LSD intervals (P = 0.05). Acetaldehyde production at harvest: 0.19 mg/100 mL. Absented measures after 50-d storage are due to the lost in fruit quality.

Figure 4 — Color index of persimmon cv. “Rojo Brillante” after being treated or not (DC-CA1, DC-CA2) to remove astringency, following 5 d at 20 °C. Fruit were previously stored at 15 °C in air (CTL) or in controlled atmospheres, 10% CO$_2$ + 90% N$_2$ (CA1) or 97% N$_2$ + air (CA2). Vertical bars represent LSD intervals (P = 0.05). Color index at harvest: 12.13.
The positive effect on delaying fruit softening under CA2 conditions is in agreement with the results obtained by other researchers using different CA; in this way, higher firmness was found in "Fuyu" persimmon stored at 0 °C under 4% O₂ + 10% CO₂, or 6% O₂ + 10% CO₂ (Park 1997), or stored at –0.5 °C under 15KPa O₂/15KPa CO₂ (Brackmann and others 1998). Kader (2003) showed that atmospheres where O₂ is low and/or CO₂ is high slow down the activity of cell wall degrading enzymes that cause fruit softening. The different amount of oxygen in the composition of both studied atmospheres could be the cause of the different effect on firmness preservation. Kader (2003) also affirmed that maintenance of fruit firmness under CA storage could be related to a reduction in respiration and ethylene production; nevertheless, in the present experiment, the CA did not cause these effects.

Astringency removal has been sensorially evaluated and related to a reduction of ST content and TI. Treatment with CO₂ was necessary to remove astringency in fruit stored under CTL or CA1 conditions. Nevertheless, a point to note is that after deastringency treatment, CTL fruit showed less ST reduction throughout storage. This fact indicates a loss of the efficacy in removal astringency. Sensor evaluation of astringency is in agreement with this result. More studies are being carried to elucidate the causes of this fact. This effect was not observed in fruit stored under the studied CA. Moreover, it is important to consider that when fruits were stored under CA2 conditions, after 30 d at 15 °C, or after 20 d following to shelf life (DC-CA2), resulted in nonastringent fruit. As a consequence of this result, deastringency treatment could be avoided when fruits were previously stored under these storage conditions.

Removing astringency under CA2 would be related to an increase in acetaldehyde production under these conditions, since after 30-d storage CA2 fruit showed high acetaldehyde values and they were similar to those shown in fruit that was treated to remove astringency. Mitcham and others (1996) also observed an increase in acetaldehyde production in "Fuyu" persimmons stored under CA.

In all treatments, the decrease of ST coincided with a reduction of TSS values; this reduction is due to the fact that ST are included in TSS measurements and they are removed with deastringency treatment (Arnal and Del Rio 2003).

"Rojo Brillante" persimmon is characterized by a bright red color. However, when this cultivar is going to be submitted to deastringency treatment, fruits are picked when color is orange and yellow (Arnal and Del Rio 2003; Salvador and others 2005), fruits were harvested with sufficient coloration for marketing. In this way, although CA delayed the color evolution, this fact did not affect final quality. Kader (2003) affirmed that CA retard loss of chlorophyll, and biosynthesis of carotenoids and anthocyanins. In persimmon cv. "Fuyu" stored in different CA also was observed a slowdown in external color change (Yang-Yong 1996).

Regarding volatiles, Ke and others (1991) reported that changes in volatiles concentration during storage might influence the flavor. Deterioration of taste in persimmon was related to accumulation of ethanol to levels exceeding 75 mg/100 mL (Ben-Arie and others 1991). Nevertheless, the results of this study showed that 120 mg/100 mL of ethanol (maximum ethanol production in CA2 fruit) did not have a negative effect on flavor of persimmon fruit cv. "Rojo Brillante."

Other researchers reported flesh disorders in fruit stored under CA (Burmeister and others 1997). The composition of the atmosphere developed in cv. "Triumph" packed fruit could be responsible for the internal browning when acetaldehyde rose to a level of 10 mg/100 mL (Ben-Arie and others 1991). Nevertheless, in the present experiment the levels of acetaldehyde were not higher than 4 mg/100 mL and no internal browning or other flesh disorders were found.

**Conclusions**

The CA composed of 97% N₂ + air (CA2) could be considered a useful alternative to permit storability for up 30 d at 15 °C of persimmon "Rojo Brillante," maintaining commercial firmness, and without the necessity of an additional treatment to remove astringency.

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**References**


