QUALITY OF PINK TOMATOES (cv. BUFFALO) AFTER STORAGE UNDER CONTROLLED ATMOSPHERE AT CHILLING AND NONCHILLING TEMPERATURES

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ABSTRACT

Greenhouse-grown pink tomatoes (cv. Buffalo) were stored in air or in a controlled atmosphere (CA) of 4% O₂ plus 2% CO₂ to study the effect of CA at chilling and nonchilling temperatures on fruit quality characteristics. Tomatoes could be stored in CA at 12C for three weeks with no major changes in fruit appearance. CA was effective in delaying color development and ripening at this temperature. The soluble solids content was lower after CA storage, but no significant differences in firmness, pH or titratable acidity were observed between storage in air or CA. Storage in CA at 12C was also effective in reducing decay. However, at 6C, fruit showed high incidence of decay after both air and CA storage. CA did not alleviate chilling injury symptoms relative to air-stored samples and may have caused CO₂ injury. Water loss was greater under CA at both temperatures.

INTRODUCTION

Tomato (Lycopersicon esculentum) is an important crop grown in Portugal with a very large production area spread over the country and average yields of about 160,000 tons/year. Tomato growing regions comprise about 54 % of the total national area, including greenhouse production with 20% of the average yield (INIA/DHF 1991; IROMA 1991). In Portugal, tomatoes for the fresh market are usually harvested at the pink stage, and are often stored or transported to distant markets at low temperatures until the fruits ripen or reach the consumer. This procedure often results in the occurrence of chilling injury symptoms such as pitting, decay, fruit surface discoloration, and inability of the fruit to ripen.

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normally. Because of the chilling injury problems with tomato fruit, CA may be useful for storage or transport at higher temperatures, or, alternatively, by allowing tomatoes to be handled at normal chilling temperatures.

Hardenburg et al. (1986) recommended that light red tomatoes be stored for no longer than 10 days in air at 10°C in order to ensure normal shelf life during retailing. A storage temperature of 10 to 13°C has been recommended for pink-red to firm-red greenhouse-grown tomatoes (Alban 1961). The use of CA storage for extending the shelf-life of tomatoes has been studied by several researchers. However, data is still lacking for tomatoes harvested at the pink stage since most of the research has been performed using mature-green or full ripe tomatoes (Kim and Hall 1970; Parson et al. 1970; Parson and Spalding 1972; Thomas et al. 1982; Valera 1984; Brown et al. 1989; Marangoni and Stanley 1991). Low O₂ and high CO₂ have been reported to reduce the expression of chilling injury in several fruits (Forney and Lipton 1990). However, the only investigation of this effect in tomato involved fruits held under very severe chilling conditions of 0°C for 10 days and with injurious CO₂ levels of 5 to 20% (Morris and Kader 1975; 1977). Kader (1992) has reported that recommended storage conditions for partially ripe tomatoes range from 8 to 12°C and CA from 3 to 5% CO₂ and 0 to 5% CO₂.

Since no reports were found for greenhouse-grown tomatoes harvested at the pink stage, the objective of this work was to evaluate the effects of CA storage at two different storage temperatures, chilling (6°C) and nonchilling (12°C), on the quality of ‘Buffalo’ greenhouse-grown pink tomatoes. The CA composition was based on recommended values for partially ripe tomatoes (Kader 1992). The final aim is to determine whether the use of CA may bring benefits to storage at non-chilling temperature, and whether it may alleviate chilling injury at low temperature.

MATERIALS AND METHODS

Plant Material

Tomatoes (cv. Buffalo) were obtained from an experimental station at Vairao, Vila do Conde, located in the north of Portugal. Fruits were grown in a Mediterranean-type greenhouse, wood frame covered with transparent plastic (11 m × 70 m × 4 m), with static ventilation provided by windows placed all around the plastic halls. The temperature and relative humidity were not controlled, but ranged between 25 to 30°C and 70 to 75%, respectively, during August and September. ‘Buffalo’ is a tomato cultivar available from July to September in this region of the country. Fruits were harvested at the pink stage (30 to 60 % of the fruit surface showing pink to red color). Two harvests/experiments were conducted during the 1993 summer season: tomatoes were harvested on August 9 for the 6°C experiment and on September 2 for the 12°C experiment.
Treatment and Storage Conditions

Fruits were harvested and transported from Vairao to the laboratory at Porto within approximately one hour. Six replicate samples of six tomato fruits each were selected for uniformity of color and freedom from defects. Each fruit was weighed and placed in a cold room at 6°C or 12°C and 80 to 85% relative humidity (RH) for one hour in order to reach the temperature of the room before the start of the experiments. The tomatoes were then stored in ambient air or CA at 6 or 12°C and 80 to 85% RH. Three replicate containers with six fruits each were stored in ambient air and used as a control. The CA was obtained by mixing nitrogen, air and carbon dioxide from pressurized cylinders. The gas mixtures were distributed uniformly into three replicate containers per treatment, each with six fruits. Prior to entering the containers, the gas mixtures were humidified by bubbling through a container with water. The gas composition was analyzed daily by sampling from the inlet port of the containers before passing over the tomato fruits. The O₂ and CO₂ concentrations in the gas samples were determined using a Shimadzu Gas Chromatograph Chromatopac C-R6A (Shimadzu Europa GmbH, Germany) equipped with a TCD detector and connected to a 3 m × 0.3 cm column of Carbosieve SII, 80/100 mesh (Supelco). Flow rate for helium (carrier gas) was 30 mL min⁻¹. The injector and detector temperatures were set at 120 and 210°C, respectively. The temperature of the column oven was programmed for 40°C for 6 min and subsequently for 15 min⁻¹ to 170°C and held for 5 min. Oxygen and CO₂ concentrations (4% and 2%, respectively) were maintained at constant levels throughout the storage period. After three weeks storage, the tomatoes were transferred to the laboratory at room temperature (around 25°C) for one day to simulate a retail display period. Physical and chemical characteristics were then measured using three samples of six tomatoes per treatment.

Weight Loss

Fruits were individually weighed before and after storage, and weight loss was calculated. Concentrations of chemical constituents were expressed in terms of dry weight. The dry weight was determined by drying a weighed aliquot of homogenized tomato tissue at 70°C for 6 days and reweighing. The following formula was used for water loss corrections: chemical component (fresh weight) × 100 g / [5g (tomato average dry weight) + weight loss during storage (g)].

Color Assessment

Surface color of each fruit was measured after storage using a hand-held tristimulus reflectance colorimeter (Minolta CR-300, Minolta Corp., Ramsey, New Jersey, USA). Color was recorded using the CIE-L*°a°*b°* uniform color space (CIE-Lab), where L° indicates lightness, a° indicates chromaticity on a green (−) to red (+) axis, and b° chromaticity on a blue (−) to yellow (+) axis. Numerical
values of \(a^*\) and \(b^*\) were converted into hue angle \((H = \tan^{-1} b^*/a^*)\), representing the shade of color, and chroma \((\text{Chroma} = (a^{*2} + b^{*2})^{1/2})\), representing color purity (Francis 1980; Shewfelt 1988).

**Firmness Measurements**

Flesh firmness of each fruit was measured after storage by compression (10 mm deformation) with an Instron Universal Testing Instrument (Model 4501, Instron Corp., Canton, Ohio, USA). A 100 Newton (N) load cell was used for firmness determination. Crosshead speed was 20 mm min\(^{-1}\). A 35 mm diameter flat plate was used. This test measured tomato firmness based on the resistance of the fruit flesh to deformation by the plate. Results were expressed in Newtons (N) (Kader 1982).

**Soluble Solids**

Replicate samples of six tomatoes were homogenized in a laboratory blender at high speed for 2 min. The homogenate was filtered through Whatman No. 4 filter paper, and the soluble solids content of the resulting clear juice was determined at 20°C with a hand-held refractometer model Atago-ATC1 (Brecht et al. 1976; Watada and Aulenbach 1979). Soluble solids content was expressed in terms of fresh and dry weight.

**pH**

The pH of the juice was determined with a pH meter Crison MicropH 2002 (Crison Instruments, S.A., Barcelona, Spain) using a xerolyte electrode Ingold, which had been previously standardized to pH 4 and pH 7.

**Titratable Acidity**

Aliquots (6.00g) of juice from the homogenized tomato fruit were diluted with 100 mL distilled water. The titratable acidity was determined by titration with 0.1N NaOH to an end point of pH 8.1 with a pH combined electrode Ingold U402-57/120 and a Crison MicropH 2002 potentiometer (Brecht et al. 1976; Watada and Aulenbach 1979). The results were calculated as percent of citric acid \([(\text{mL} 0.1\text{N NaOH} * 0.064/6.00\text{g of juice}) * 100]\), and expressed in terms of fresh and dry weight.

**Decay**

Decay incidence was recorded by counting the number of tomatoes showing evidence of disease. The Horsfall-Barratt (HB) grading system was used to determine disease severity rated as the amount of tomato surface area affected by decay (Horsfall and Barratt 1945). This system is based on a 50% midpoint since
the human eye perceives diseased tissue below 50% and sound tissue above 50% (Horsfall and Cowling 1978). Therefore, the grades differ by a ratio of two in either direction from 50% as follows: 1 = 0%, 2 = 0 to 3%, 3 = 3 to 6%, 4 = 6 to 12%, 5 = 12 to 25%, 6 = 25 to 50%, 7 = 50 to 75%, 8 = 75 to 87%, 9 = 87 to 94%, 10 = 94 to 97%, 11 = 97 to 100%, 12 = 100%. The higher grades indicate higher percentage of decay.

Statistical Analysis

The Statistical Analysis System computer package (SAS Institute 1986) was used for analysis of the data in these experiments. Data for the two temperatures were analyzed separately. Significant differences between fruits stored in air or controlled atmosphere were detected using the t test.

RESULTS AND DISCUSSION

Temperature Effect

In this study, greenhouse-grown pink tomatoes stored in air or CA at 6C developed characteristic symptoms of chilling injury after 3 weeks' storage. Irregular coloration and development of orange rather than red pigments, pitting, water soaking, tissue browning and necrosis, and decay were all visually apparent. However, fruits stored under air or CA at 12C showed normal color development and had no pitting. These results indicate that 12C is a nonchilling temperature for greenhouse-grown pink ‘Buffalo’ tomatoes. Jackman et al. (1988) observed that tomatoes stored below 7 to 10C often develop chilling injury symptoms such as enhanced microbial spoilage, pitting due to collapse of the cells beneath the skin, softening, and poor color development. Marangoni and Stanley (1991) found that greenhouse-grown mature-green tomatoes stored at 6C in air or modified atmosphere (3% O₂ plus 2% CO₂) showed marked deterioration after 15 days’ storage. They noticed that storage in low levels of O₂ and high levels of CO₂ did not prevent the development of chilling injury symptoms in tomatoes stored at 6C. However, fruits stored at 12C for 15 days showed good quality characteristics. Brown et al. (1989) also noticed that mature-green tomatoes stored at 5C developed severe pitting after 22 days storage, while tomatoes stored at 12.5C had no pitting.

Weight Loss and Firmness

Tomatoes stored in air had lower weight loss than those stored in CA at both storage temperatures. Weight loss in air was about 50% lower than in CA at both 6 and 12C (Table 1). We attribute these differences mainly to differences in water
TABLE 1.  
EFFECTS OF TEMPERATURE AND CA STORAGE FOR 3 WEEKS PLUS 1 DAY IN AIR AT 25C ON WEIGHT LOSS AND FIRMNESS OF ‘BUFFALO’ TOMATOES

<table>
<thead>
<tr>
<th>Storage conditions</th>
<th>Weight loss (% FW)</th>
<th>Firmness (N)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stored at 6C</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Air</td>
<td>2.13b</td>
<td>61.09a</td>
</tr>
<tr>
<td>4% O₂ + 2% CO₂</td>
<td>4.24a</td>
<td>47.97b</td>
</tr>
<tr>
<td>Stored at 12C</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Air</td>
<td>0.86b</td>
<td>49.25a</td>
</tr>
<tr>
<td>4% O₂ + 2% CO₂</td>
<td>1.97a</td>
<td>54.43a</td>
</tr>
</tbody>
</table>

Data are means of 18 individual fruit replicates. Means in columns within a given temperature followed by the same letter are not significantly different at the P=0.05 level according to the t test.

vapor pressure between the gas mixtures and the produce. In fact, the RH of ambient air was maintained in a range of 80 to 85% in the cold room. In spite of humidifying the gas mixtures, the moisture content of the CA mixtures was probably not as high as in the air treatments due to the initial dry condition of the pressurized gases used to create the CA. This effect could have been prevented by improving the humidification in order to ensure that RH was in the recommended 90 to 95% range (Hardenburg et al. 1986) inside all the containers. In spite of the differences observed, water loss was less than 2% during 3 weeks’ storage at 12C.

The firmness of tomatoes stored in CA at 6C was lower than that of fruits stored in air, while at 12C, no statistical difference (P>0.05) was found between CA and air (Table 1). Valera (1984) also noticed that tomatoes stored in air at 20C were firmer than CA stored fruits at 10C. However, some other authors found that tomatoes stored in atmospheres with low O₂ content exhibited delayed softening. Kim and Hall (1976) associated the delayed softening with delayed ripening of tomatoes stored in CA. The contradictory firmness results at 6C in the present study could be related to the higher weight loss of tomatoes during CA storage. Although fruit surface shriveling due to water loss was not noticed visually, the pericarp tissue may have been more flaccid in the CA fruit, which could have resulted in lower firmness readings. Another possibility is that CO₂ injury occurred
in the tomatoes at the lower temperature. Symptoms of CO₂ injury in tomato include softening, as well as discoloration and uneven ripening (Saltveit 1993), which could be confused with chilling injury symptoms. We have previously observed greater sensitivity to CO₂ injury in snap beans at lower temperatures (Costa et al. 1994), presumably due to greater solubility of CO₂ in the cytoplasm at lower temperatures.

Color

Color development was nonuniform in tomatoes stored at 6C due to chilling injury at this temperature. Fruits stored in CA at 6C had lower L* values and higher a* values than air-stored fruits (Table 2). However, hue and chroma values did not differ between the air and CA treatments at 6C. At 12C, CA storage delayed development of red color, as shown by lower a* and higher hue values than air-stored fruits. Chroma values were lower after CA storage when compared with air storage at 12C. The tomatoes were also lighter (higher L* value) after CA storage at 12C. As described by Shewfelt et al. (1988), color development during tomato ripening is characterized by lower L* readings, a change from negative to positive a* values, decreasing hue angle, and increasing chroma. In other studies with tomato stored at about 12C, low O₂ atmospheres inhibited degradation of

<table>
<thead>
<tr>
<th>Storage conditions</th>
<th>L*</th>
<th>a*</th>
<th>Hue</th>
<th>Chroma</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stored at 6C</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Air</td>
<td>49.62a</td>
<td>9.56b</td>
<td>69.25a</td>
<td>27.05a</td>
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<tr>
<td>4% O₂ + 2% CO₂</td>
<td>47.49b</td>
<td>12.02a</td>
<td>63.83a</td>
<td>27.35a</td>
</tr>
<tr>
<td>Stored at 12C</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Air</td>
<td>44.39b</td>
<td>22.73a</td>
<td>48.80b</td>
<td>34.50a</td>
</tr>
<tr>
<td>4% O₂ + 2% CO₂</td>
<td>48.95a</td>
<td>12.53b</td>
<td>64.38a</td>
<td>29.24b</td>
</tr>
</tbody>
</table>

Data are means of 18 individual fruit replicates. Means in columns within a given temperature followed by the same letter are not significantly different at the P=0.05 level according to the t test.
chlorophyll and synthesis of lycopene and carotene, resulting in less red fruits (Kim and Hall 1976; Salunkhe and Wu 1973). Goodenough and Thomas (1980) also reported that there was no synthesis of lycopene and other carotenoids during CA storage, resulting in less red fruits. Brown et al. (1989) noticed that tomatoes stored in air at 12.5°C for 22 days turned red, while a modified atmosphere of 3% O₂ plus 2% CO₂ effectively inhibited red color development.

Chemical Properties

The dry weight of the tomatoes after storage averaged about 6% of the initial fresh weight. This level is in agreement with other published data (McCance and Widdowson 1978; Goodenough and Thomas 1980). The higher water loss in the tomatoes stored under CA tended to mask real changes of soluble solids and titratable acidity when expressed on a fresh weight basis. Therefore, the data 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. Fruits stored in air at 6°C had higher pH than tomatoes stored in CA, while fruits stored at 12°C showed no difference in pH (Table 3). The total titratable acidity did not differ significantly between fruits stored in air or CA at either 6°C or 12°C (Table 3). Valera (1984) also noticed that tomatoes stored in air at 10 or 20°C had higher pH than those stored in CA, agreeing with Parsons et al. (1970), who observed that mature-green tomatoes stored in CA (3 or 5% CO₂) tended to be more acid that those stored in air.

<table>
<thead>
<tr>
<th>Storage conditions</th>
<th>pH</th>
<th>Titratable acidity (%) DW (%) FW</th>
<th>Soluble solids (%) DW (%) FW</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stored at 6°C</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Air</td>
<td>4.10a</td>
<td>4.80a 0.39a</td>
<td>55.35a 4.5b</td>
</tr>
<tr>
<td>4% O₂ + 2% CO₂</td>
<td>4.05b</td>
<td>4.07a 0.42a</td>
<td>48.83b 5.0a</td>
</tr>
<tr>
<td>Stored at 12°C</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Air</td>
<td>3.83a</td>
<td>6.02a 0.42a</td>
<td>65.60a 4.5a</td>
</tr>
<tr>
<td>4% O₂ + 2% CO₂</td>
<td>3.87a</td>
<td>4.98a 0.40a</td>
<td>52.70b 4.2b</td>
</tr>
</tbody>
</table>

Data are means of 18 individual fruit replicates. Means in columns within a given temperature followed by the same letter are not significantly different at the P=0.05 level according to the t test.
The soluble solids content was lower for fruits stored in CA at 6C or 12C (Table 3). These results agree with the report of Salunkhe and Wu (1973) that low O₂ atmospheres inhibit starch degradation and sugar formation in tomatoes. However, Goodenough et al. (1982) found that starch in mature-green tomatoes was degraded during 8 weeks in a CA of 5% O₂ plus 5% CO₂. Goodenough et al. (1980, 1982) also found that the concentration of sugars and organic acids decreased following CA storage. These observations are not contradictory with the results of this study, since the storage was over a 3 week period and the quality evaluation was carried out after the storage period plus one day at room temperature.

Decay

Tomatoes stored in air at 6 or 12C had higher incidence and severity of decay compared with those stored in CA at the same temperatures (Table 4). Parsons and Spalding (1972) observed that decay in mature-green tomatoes stored in a CA of 3% O₂ plus 5% CO₂ was lower than in tomatoes stored in air for 6 weeks at 12.8C. They also found that CA was more effective in reducing decay at 12.8C than at 7.2C. In the experiments reported here, the development of decay in tomatoes seemed to be affected more by the temperature than by the composition of the atmosphere. In spite of the fruits being from two different harvests, the results of this study seem to reveal a synergistic effect of CA and temperature on decay incidence: there was 75% less decay in CA at 12C as there was at 6C, but only 53% less decay in air at 12C as at 6C, suggesting that chilling injury may have been slightly reduced by CA.

<table>
<thead>
<tr>
<th>Storage conditions</th>
<th>Decay Incidence (%)</th>
<th>Decay Severity (%)</th>
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</thead>
<tbody>
<tr>
<td><strong>Storage at 6C</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Air</td>
<td>83.30</td>
<td>37.5a</td>
</tr>
<tr>
<td>4% O₂ + 2% CO₂</td>
<td>66.70</td>
<td>9.0b</td>
</tr>
<tr>
<td><strong>Storage at 12C</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Air</td>
<td>38.90</td>
<td>4.5a</td>
</tr>
<tr>
<td>4% O₂ + 2% CO₂</td>
<td>16.70</td>
<td>1.5b</td>
</tr>
</tbody>
</table>

Data are means of 18 individual fruit replicates. Means in columns within a given temperature followed by the same letter are not significantly different at the P=0.05 level according to the t test.
CONCLUSIONS

Storage of pink ‘Buffalo’ tomatoes in a CA of 4% O₂ plus 2% CO₂ at 12C was somewhat effective in delaying color development and ripening. The CA fruits were lighter and less red; soluble solids content was 24% lower, but no significant differences in firmness, pH or titratable acidity were observed. Storage in CA at 12C was also effective in reducing decay: both incidence and severity of decay were about half that of air stored fruits. Storage of these tomatoes in 4% O₂ plus 2% CO₂ at 6C did not significantly reduce the expression of chilling injury, but rather seemed to accentuate the negative effects of low temperature such as watersoaking, pitting and discoloration, and loss of fruit brightness. Also CA storage at this temperature does not seem to retard tomato ripening and the fruits showed significant loss of physical and chemical quality characteristics and high incidence of decay. The greater softening noticed under CA at 6C suggests that CO₂ injury may have occurred at the lower storage temperature. Special attention should be paid to the RH of the gas mixture in order to avoid water loss during CA storage. Storage of these fruits harvested at the pink stage for longer than 3 weeks at 12C under CA would not be possible without additional measures to control decay. Although storage in CA (4% O₂ plus 2% CO₂) at 12C may contribute to extended shelf-life of pink tomatoes, CA (4% O₂ plus 2% CO₂) did not allow storage at a chilling temperature of 6C.

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