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Modified atmosphere and modified humidity packaging alleviates chilling injury symptoms in mango fruit

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Abstract

Storage of mango (*Mangifera indica* L. cvs. Tommy Atkins and Keitt) fruits at 12°C caused slight chilling injury (CI) symptoms on the fruit peel, expressed as red spots around the lenticels (lenticel spotting). A modified atmosphere (~ 5% CO_2 and ~ 10% O_2) was created in 4-kg film-lined cartons by using microperforated polyethylene (PE) or Xtend® film (XF). For 'Keitt' fruit, a similar atmosphere was also applied using controlled atmosphere chambers. After 3 weeks of storage at 12°C plus 1 week at 20°C, both modified and controlled atmosphere treatments were effective in reducing CI. The most effective reduction was found in fruits that were packed in the XF film. A second advantage of using XF film was the reduction in the level of sap inside the package due to the lower relative humidity in the XF film (~ 90%) compared with that of PE packaging (~ 99%). © 2000 Elsevier Science B.V. All rights reserved.

Keywords: Mangifera indica; Lenticel spotting; Ethylene; CO_2; O_2; Relative humidity; Controlled atmosphere (CA); Modified atmosphere packaging (MAP)

1. Introduction

The atmosphere generated by modified atmosphere packaging (MAP) delays ripening of certain subtropical-tropical fruits, including mango (Kader, 1994). The main factors that maintain mango quality in various film packagings are increased CO_2 and decreased O_2 levels, which reduce respiration rate and prevent water loss (Chaplin et al., 1982; Miller et al., 1983; Yuen et al., 1993; Rodov et al., 1997). However, despite its success at the laboratory level (Chaplin et al., 1982; Miller et al., 1983; Yuen et al., 1993; Rodov et al., 1997), MAP is still not a commercial technique.

Mango fruits suffer from the appearance of red or green spots around the lenticels. These may result from various stresses to which the fruit has been exposed in the orchard and/or during packaging processes (O'Hare et al., 1996; Jacobi and

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Giles, 1997). In 'Kensington' mango it has been shown that drops of water can enhance the formation of lenticel spots (O'Hare et al., 1996). Fruit storage under temperatures of 10–12°C accelerates the formation of red spots, symptomatic of slight chilling injury (Pesis et al., 1997), and chilling has been found to induce the formation of anthocyanins in plant tissue (Dixon and Palva, 1995). We have shown that pretreatment with temperature conditioning, heat or anaerobiosis can reduce these red spot symptoms (Pesis et al., 1997).

Packaging of horticultural crops within plastic films creates a modified atmosphere (MA) higher in CO_2 and H_2O and lower in O_2 than ambient levels, in response to the respiration of and moisture loss from the commodity. It has been shown that elevated CO_2 and H_2O and reduced O_2 are beneficial in alleviating CI symptoms in chilling-sensitive crops (Forney and Lipton, 1990). MA packaging has been effective in alleviating CI symptoms in chilling-sensitive crops such as lime and grapefruit (Wardowski et al., 1973), avocado (Scott and Chaplin, 1978; Meir et al., 1997) and cucumber (Wang and Qi, 1997).

Chaplin et al. (1982) were among the first who tried to increase 'Kensington' mango shelf life by using MA; the gas composition in perforated polyethylene (PE) bags was 8.9% CO_2 and 15.2% O_2, and that in sealed PE bags 14.8% CO_2 and 1.8% O_2. However, Chaplin et al. (1982) concluded that storage of mangoes in PE bags at 20°C could cause development of abnormal peel and flesh colour, and off-flavours. Also, individually sealing 'Tommy Atkins' mangoes in heat-shrinkable plastic films was not promising (Miller et al., 1983); the fruits showed less weight loss, but there were no differences in firmness, colour development and decay development, compared with unwrapped fruit. Moreover, decay and off-flavour development associated with some of the films remains an obstacle to commercial acceptance (Miller et al., 1983). Shrink-wrap or sealed PE packaging delayed ripening, and after 30 days storage at 20°C had reduced peel injury of 'Kensington' mango relative to unpackaged fruit, but there was an undesirable retention of green peel colour (Yuen et al., 1993). Rodov et al. (1997) found that microperforated film was more beneficial for mango packaging, as it avoided the accumulation of dangerous levels of CO_2 that can cause off-flavours and peel injury.

Recently developed Xtend® film (XF), has a higher water vapour transmission rate than PE film and so has better potential to reduce humidity in the package. This XF film has been found to be beneficial for several fruit commodities, including cherry and nectarine (Lurie and Aharoni, 1998) and sweet corn (Aharoni and Richardson, 1997). In the present paper, the effects of sealing mango fruits in microperforated PE or microperforated XF bags in order to maintain the fruit quality, are presented.

2. Materials and methods

2.1. Fruit material

Mature mango (*Mangifera indica* L.) fruits of the cvs. Tommy Atkins or Keitt were harvested from a local orchard in the Sharon or the Jordan Valley regions of Israel. All fruits were treated the day after harvest, some in the laboratory and some in the packaging house.

2.2. Postharvest treatments, waxing and wrapping

2.2.1. Experiment 1

In the first experiment, 'Tommy Atkins' mangoes were harvested in the Sharon region in Israel and treated the next day in the laboratory. All fruits were washed in tap water and treated with the fungicide prochloraz (0.2%). The fruit were divided into lots, each of which was given one of the following five treatments: (1) control — non-waxed fruits; (2) waxed fruits; (3) non-waxed, sealed in packages made of Xtend® film XF.A12; (4) non-waxed, sealed in bags made of Xtend® film XF.A13; and (5) non-waxed, sealed in PE bags. All films were microperforated.

The XF film bags were obtained from Xtend® (Stepac, L.A., Israel). The liners measured 75 × 75 cm and their thicknesses were 20 μm for the Xtend® films and 40 μm for the low-density polyethylene (PE) film. The Xtend® films were manufactured from proprietary blends of polymeric materials having O_2 permeances between 24×10^{-14} and 48×10^{-14} mol s^{-1} m^{-2} Pa^{-1}. Gas permeance of the packages was modified by the presence of microperforations in the Xtend® and PE films ($\sim$0.0018% of the total film area). The Xtend® films used in the experiments had various water vapour transmission rates (WVTR): 6×10^{-10}, 18×10^{-10}, and 24×10^{-10} mol s^{-1} m^{-2} Pa^{-1}, respectively, for the three films, XF.A12, XF.A13, and XF.A14. The PE film had an O_2 permeance of 35×10^{-12} mol s^{-1} m^{-2} Pa^{-1} and a WVTR of $\sim11.8\times10^{-11}$ mol s^{-1} m^{-2} Pa^{-1}. The permeability data were provided by the manufacturer and converted into SI units according to Banks et al. (1995).

Each treatment consisted of three cardboard boxes, each containing 4 kg of fruit (ten fruits). All fruits were stored at 12°C in a room held at $\sim$85% RH for 21 days. After cool storage, the fruits were held for 5 additional days at 20°C and $\sim$80% RH to simulate shelf life.

2.2.2. Experiment 2

In a second experiment 'Keitt' mango fruits were treated in the packaging house near the Sea of Galilee (Zemach). All fruits were washed and brushed for 15 s in hot water (55°C) and treated with prochloraz (0.2%) (Prusky et al., 1999). The fruits were divided into five lots, each of which was given one of the following treatments: (1) non-waxed non-sealed fruits packed without special drying (wet); (2) non-waxed non-sealed fruits, dried with a hair dryer before packing (dry); (3) waxed fruit; (4) fruits sealed in microperforated XF.A12 film package; and (5) fruits sealed in microperforated XF.A14 bags. Each treatment consisted of three boxes each weighing 4 kg (ten fruits). All fruits were stored at 12°C for 21 days and at 20°C for an additional 5 days.

2.2.3. Experiment 3

In a third experiment, 'Keitt' mango fruits were harvested in the Sharon region of Israel and treated the next day in the laboratory. All fruits were brushed for 15 s in hot water and then treated with prochloraz (0.2%). The experiment had a 5×2 factorial design: five treatments and two types of fruits, non-waxed and waxed. The five treatments were: (1) control, non-sealed; (2) sealed in XF.A12 packages; (3) sealed in XF.A14 microperforated bags; (4) stored in a controlled atmosphere (CA) (5% CO_2 and 15% O_2); and (5) control, non-sealed stored at 20°C. Each treatment comprised six boxes (three non-waxed and three waxed) each containing 4 kg (eight fruit). All treatments, except the treatment that remained at 20°C, were stored for 21 days at 12°C and removed to shelf life at 20°C for an additional 5 days.

The wax used in all experiments was 12% (v/v soluble solids), polyethylene emulsion wax (Safepack, Israel).

2.3. Fruit ripening and CI indices

Ripening indices of firmness and peel colour, and injury indices of red and green lenticel spotting were determined by hand grading and visual inspection using 1–10-point scales: firm fruit 10, soft fruit 1; green fruit 1, colourful 10. In 'Tommy Atkins' fruit colourful means red and in 'Keitt' yellow. The indices were calculated by the following formulae:

$$\text{Firmness Index} = \sum_{10}^{1} \frac{(\text{index level})*(\text{no. of fruits in this level})}{\text{total no. of fruits}}$$

$$\text{Peel colour Index} = \sum_{1}^{10} \frac{(\text{index level})*(\text{no. of fruits in this level})}{\text{total no. of fruits}}$$

The chilling injury (CI) index was based on the peel area covered with red or green spots: 0, no injury; 1, low injury (i.e. 10% of the peel covered with spots); 5, medium injury (i.e. 30% or more of the peel covered with spots); 10, high injury level (i.e. 50% or more of the peel covered with spots). The indices were calculated according to the following formula:

$$\text{Red or green spots Index} = \sum_{0}^{10} \frac{(\text{index level})*(\text{no. of fruits in this level})}{\text{total no. of fruits}}$$

The gases accumulated in the packages were measured by gas chromatography. A silicone septum was glued on each bag and headspace samples were taken with a 10-ml syringe. The GC was fitted with a thermal conductivity detector with a double column CTR-1 (Altech). Ethylene was determined with a flame ionization detector using an alumina column.

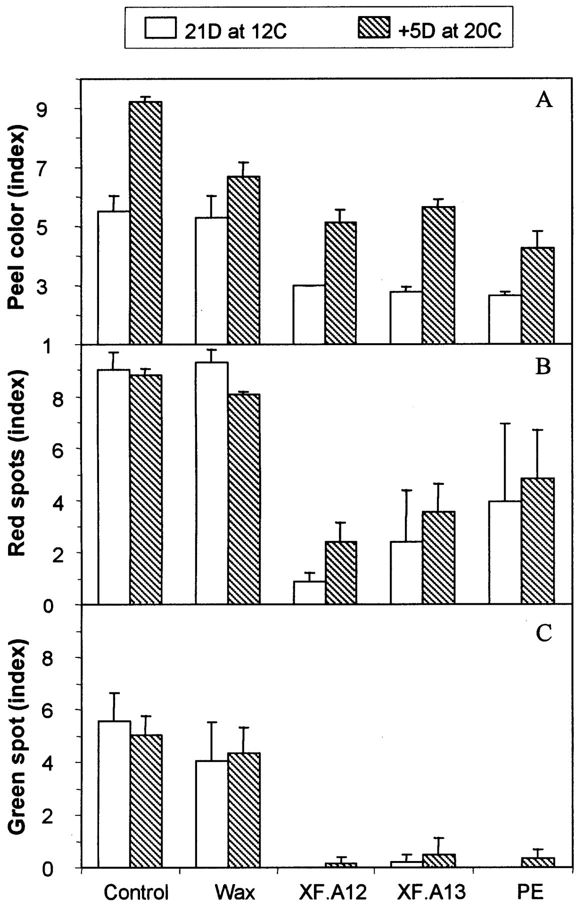

Fig. 1. Effect of waxing and MA treatment on colour development (A), and chilling injury, expressed as red spots (B) and green spots (C) in mango cv. Tommy Atkins, after 21 days storage at 12°C + 5 days shelf life at 20°C. Treatments as described in the first experiment. The results are means of three boxes ± S.E.

2.4. Measurements of relative humidity, sap and weight loss

Relative humidity (RH) was checked at the end of the cold-storage period. Shortly before the bags were opened, a probe (Delta OHM, HD 9216, Italy) was inserted into them via a small hole. The measurements were taken after 1 h of equilibration.

Immediately after the bags were opened, the amount of sap that stuck to the film was recorded visually. The sap index indicates the stickiness of the fruit to the film: 0, no sap; 4, greatest amount of sap sticking to the film. Fruits were weighed and the weight loss was calculated at the end of shelf life at 20°C.

All the results shown as columns represent means ± S.E. The results of gas analysis are means of three samples. Least significant difference (LSD) at 5% was calculated for each gas.

3. Results

The highest peel colour development in mango cv. Tommy Atkins was found in the control fruit; waxing the fruit resulted in reduced colour development after transfer to shelf-life conditions. However, packing the fruit in PE or in XF films reduced peel colour development by almost 50%, after 21 days at 12°C plus 5 days at 20°C. The lowest colour development was found in fruit packed in PE (Fig. 1A).

The chilling injury (CI) symptoms at 12°C in 'Tommy Atkins' fruit were expressed as red spots around the lenticels on the red cheek, and green spots around the lenticels on the green cheek. In the control or waxed fruit the red or green spots around the lenticels were very noticeable (Fig. 1B, C). Packing the fruits in XF or PE film dramatically reduced the development of red or green spots, while the most effective packaging for the reduction of red spots was the XF.A12 film.

Extraction of the area around the red lenticel with acidic methanol, revealed more anthocyanins absorbance at 530 nm and more chlorophyll absorbance at 420 nm compared to the unspotted area (Fig. 2). The chlorophyll absorbance at 650

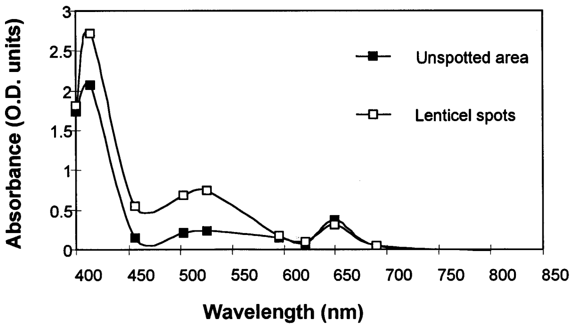

Fig. 2. Absorbance spectra at visual wavelength (400–800 nm) of red lenticel spots and unspotted peel area extracts of mango cv. Keitt. The fruit were stored for 24 days at 12°C followed by 8 days shelf life. The graph represents a typical absorbance spectrum (five extracts).

nm was the same in the red-spotted area and in the unspotted area (Fig. 2).

The highest levels of CO_2 (~ 5 kPa) and the lowest levels of O_2 (~ 15 kPa) were found in the atmosphere that surrounded the fruit packed in XF.A12 (Fig. 3). Packing in XF.A13 yielded lower levels of CO_2 and higher levels of O_2. The PE bags had the most gradual accumulation of CO_2 and showed a pronounced ethylene peak (6.4 nmol l^{-1}) after 15 days at 12°C (Fig. 3). In the XF.A12 bags the levels of ethylene were generally low (2.64 nmol l^{-1}) after 15 days at 12°C.

The level of sap adhesion differed between films at the end of the experiment (Table 1): the greatest accumulation occurred in the PE bags, where the RH was highest, while in XF.A13 bags, where the lowest RH was measured, almost no sap was found on the film (Table 1).

Non-packed, non-waxed fruits declined in firmness most rapidly, and dry fruits softened before the wet-packed fruits. Packing in XF.A12 bags kept the fruit firmer than packing in XF.A14 or only waxing (Fig. 4A). 'Keitt' is a green mango cultivar that develops only slight colour; nevertheless, packing 'Keitt' fruit in XF.A12 caused a significant reduction in peel colour development after 19 days at 12°C followed by 5 days at 20°C (Fig. 4B). The level of red lenticel spots was least in the fruits packed in XF.A12 and XF.A14, but fruits that were dried immediately after hot water brushing, and the waxed fruits, also had lower levels of red spots than the wet controls (Fig. 4C).

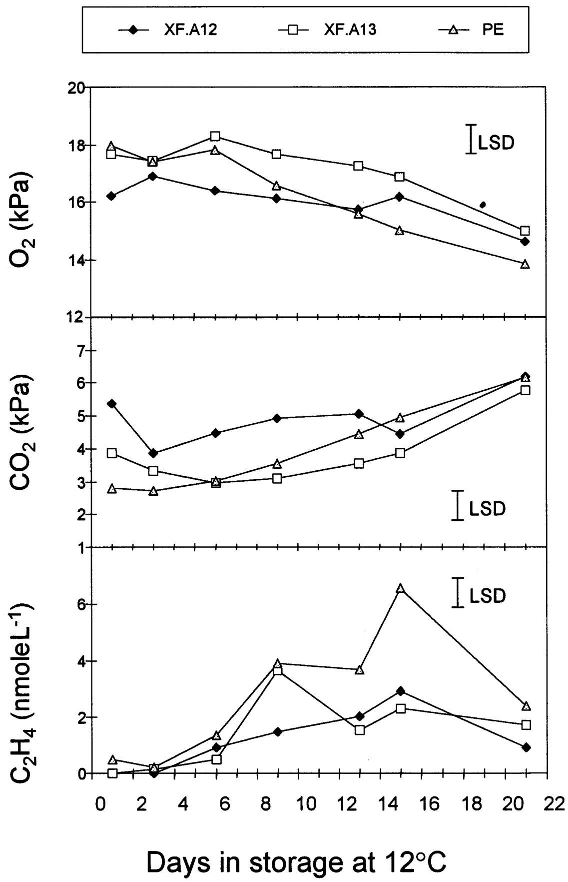

Fig. 3. Effects of different types of film in MA treatments (XF.A12; XF.A13 and PE) on O_2, CO_2 and C_2H_4 levels in non-waxed mango cv. Tommy Atkins stored at 12°C. The results are means of three boxes. LSD at 5% level for $O_2 = 1.1$; $CO_2 = 0.9$; $C_2H_4 = 0.96$.

Table 1
Effect of various film packaging on relative humidity (RH) and adherence level of the sap (0–4) in 4-kg boxes of non-waxed mango cv. Tommy Atkins stored for 3 weeks at 12°C, in XF.A12, XF.A13 and PE bags[a]

Treatment	Relative humidity (%)	Sap index (0–4)
XF.A13 bags	91.6 ± 1.2	0.3 ± 0.5
XF.A12 bags	95.3 ± 1.4	2.0 ± 1.0
PE bags	98.9 ± 0.9	3.7 ± 1.3

[a] The results are means of three boxes ± S.E.

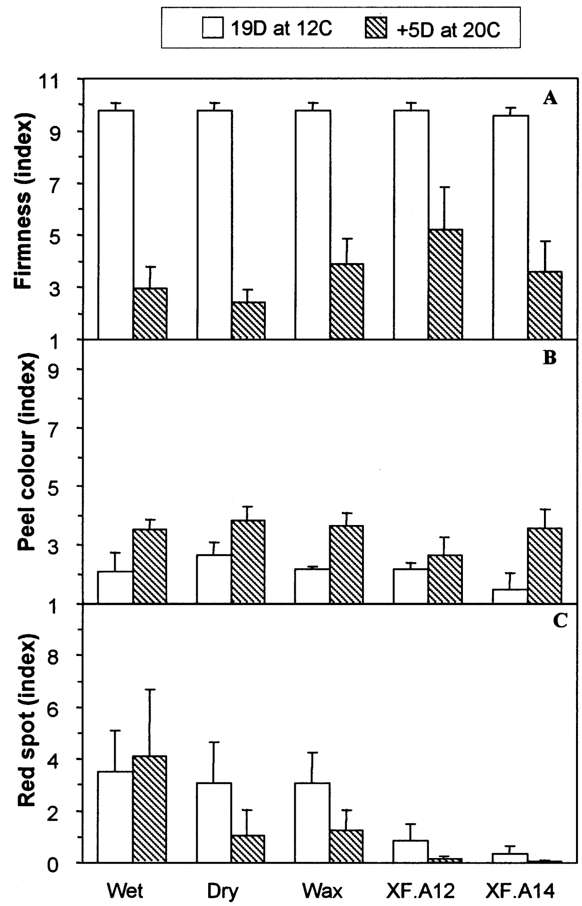

Fig. 4. Effect of drying, waxing and MA treatments on firmness of the fruit (A), colour change (B) and chilling injury expressed as red spots (C) in mango cv. Keitt after 19 days storage at 12°C + 5 days at 20°C. Treatments as described in the second experiment. The results are means of three boxes ± S.E.

The greater firmness which was maintained in fruits stored in XF.A12 bags than in those stored in XF.A14 bags (Fig. 4A) correlates with the lower weight loss from fruits packed in XF.A12 bags, for both non-waxed and waxed fruits (Table 2). The greater firmness retention and the lower weight loss are probably due to the higher RH in the XF.A12 bags (95%) than in the XF.A14 bags (88%) (Table 2).

The effects of packing non-waxed and waxed 'Keitt' mango in XF bags, on CO_2 and ethylene accumulation inside the bag, were different (Fig. 5). In bags containing waxed fruit the same accumulation of CO_2 and ethylene were found in both XF.A12 and XF.A14 bags (Fig. 5A, C), while in bags containing non-waxed fruit the accumulations of CO_2 and ethylene were higher in the XF.A14 bags than in the XF.A12 bags (Fig. 5B, D).

The level of red spots in 'Keitt' following cold conditions and shelf-life storage was lower than that after removal from cold storage (Fig. 4C, Table 3). This might be because there was an enhancement of peel colour during shelf life (Fig. 4B, Table 3) which masked the colour of the lenticel spots.

Non-waxed fruits packed in XF.A14 bags had a lower level of red spots than those packed in XF.A12 bags (Fig. 4C), probably because of higher CO_2 accumulation inside the packaging (Fig. 5B). However, when the fruits were waxed the levels of red spots were similar in both packages (Table 3), for similar accumulated CO_2 levels (Fig. 5A).

Waxing caused a reduction in peel colouring of 'Keitt' fruit in all treatments, after cold storage and 5 days of shelf life at 20°C or even in fruits stored only at 20°C (Table 3). The highest colour level developed in non-waxed fruits that were not

Table 2
Effect of various film packaging and waxing on weight loss in mango cv. Keitt in 4-kg boxes after 3 weeks at 12°C and 5 days at 20°C[a]

Treatment	Weight loss (%)	Relative humidity (%)
Control non-waxed	4.75 ± 0.32	~85 (room, 12°C)
Waxed	3.25 ± 0.30	~85 (room, 12°C)
Non-waxed in XF.A12 bags	2.05 ± 0.14	95.3 ± 0.73
Non-waxed in XF.A14 bags	3.10 ± 0.23	88.3 ± 0.71
Waxed in XF.A12 bags	1.83 ± 0.12	95.8 ± 0.52
Waxed in XF.A14 bags	2.61 ± 0.22	88.7 ± 0.81

[a] RH was checked in the packaging and in the room during 12°C storage. The results are means of three boxes ± S.E.

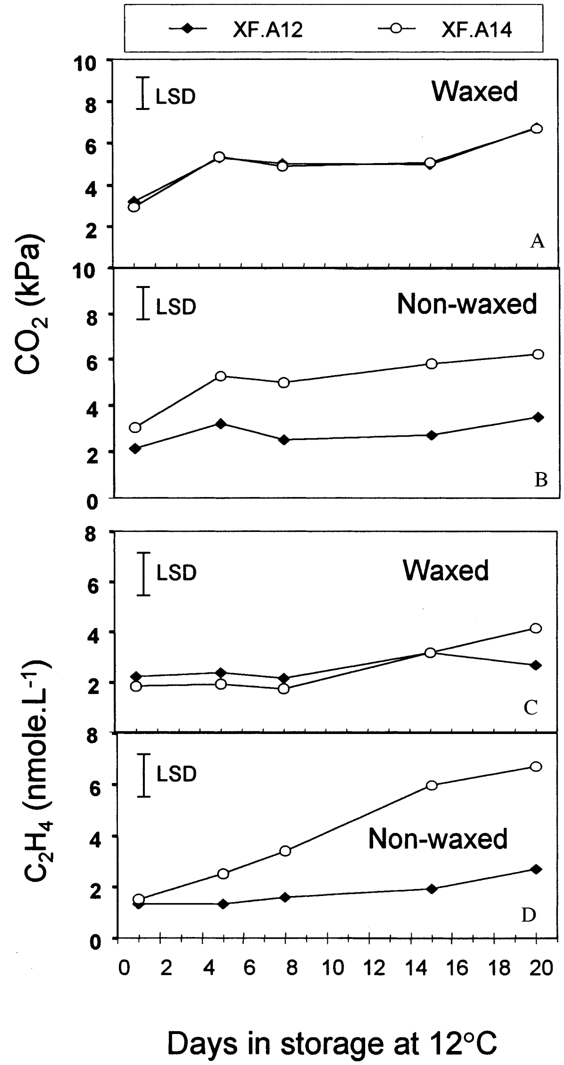

Fig. 5. Effects of fruit waxing and type of film (XF.A12 and XF.A14) on accumulation of CO_2 and ethylene in 4-kg bags of mango cv. Keitt: waxed mango (A and C) and non-waxed mango (B and D). The results are means of three boxes. LSD at 5% level for $CO_2 = 1.8$; $C_2H_4 = 1.6$.

cold-stored, but stored for 21 days at 20°C (Table 3).

Incidence of red spots was reduced by holding fruit under a controlled atmosphere (CA) for 21 days at 12°C, in conditions similar to those in the MA (5 kPa CO_2 and 15 kPa O_2), followed by 5 days at 20°C in air. The level of spotting was similar to that in the MA storage for packaging XF.A12 and XF.A14 films (Table 3). Storing the fruit only at 20°C (without cold storage at 12°C) resulted in the lowest incidence of red spots on waxed or non-waxed 'Keitt' fruit peel (Table 3), which might indicate that the red spots resulted from cold storage at 12°C.

4. Discussion

Lenticel spotting in mango peel can occur in the orchard, in response to many stresses, such as wind or cold. In addition, packing house treatments can enhance lenticel spotting as a result of water droplets, bruising, or vapour heat or hot water treatment (O'Hare et al., 1996; Jacobi and Giles, 1997). We found previously that the level of red spots increased in mangoes exposed during storage to slight chilling temperatures, 10–12°C (Pesis et al., 1997).

The accumulation of anthocyanins in plant tissue in response to chilling stress is a well known phenomenon (Dixon and Palva, 1995). Development of green spots on the green cheeks is not uncommon in mango fruits and is attributed to the response of the lenticels to various stresses. In 'Kensington' mango, this disorder is, in general, described as lenticel spots (O'Hare et al., 1996). In 'Kensington' fruit polymeric film packaging reduced the levels of peel injury in calcium-treated fruit (Yuen et al., 1993).

In 'Kensington' mango, peel injury and lenticel spotting occurred as a result of calcium treatments, although, bagging the treated fruits in PE reduced pore browning on the mango surface (Yuen et al., 1993). Yuen et al. (1993) concluded that it was not clear whether the injury was reduced by the MA surrounding the fruits, or by increased humidity in the air surrounding them (Yuen et al., 1993).

The high effectiveness of XF.A12 bags in reducing lenticel spotting (Figs. 1 and 4, Table 3) can possibly be attributed to two factors: the modified atmosphere surrounding the fruits (Figs. 3 and 5), and the maintenance of an RH in the bags of $\sim 95\%$ (Tables 1 and 2).

With 'Kensington' mango it was shown that avoiding condensation in the bags eliminated the appearance of lenticel spots (O'Hare et al., 1996). In the present study, too, a higher level of red lenticel spots was correlated with very high RH (98%) in the PE bags, while fewer lenticel spots were found under the lower RH (95%) in the XF.A12 bags (Fig. 1B).

Lower RH in the MA packaging accelerated ripening in non-waxed mangoes, as indicated by higher respiration and ethylene levels (Fig. 5). Fruit packed in XF.A14 bags (RH 88.2%) (Table 2) produced higher CO_2 and ethylene levels (Fig. 5B, D) compared with those produced by fruit packed in XF.A12 bags (RH 95.3%) (Table 2, Fig. 5B, D). These results are in agreement with findings for bananas, for which lower humidity around the fruit accelerated ripening, through increased ethylene and CO_2 production (Burdon et al., 1994).

However, in the present study, waxing the fruit reduced the effects of lower humidity on respiration and ethylene production. This was probably the reason for the fact that no differences were found in peel colour development, respiration and ethylene production, between waxed fruits packed in XF.A12 or in XF.A14 bags (Fig. 5A, C, Table 3).

The delay in peel colour development caused by waxing (Table 3) is similar to that found in many other studies, in which it was shown that coating mangoes with wax or polysaccharides, caused delay in ripening, expressed as reduced colour development (Mitra and Baldwin, 1997).

The efficacy of CA storage with mango is still a controversial issue. CA storage was found to be effective in prolonging the storage life of 'Tommy Atkins' and 'Kent' fruit from Chile (Lizana and Ochagavia, 1997), but not of 'Kent' fruit from Mexico (Trinidad et al., 1997). In our present study, a CA of 5 kPa CO_2 and 10 kPA O_2 was effective in reducing peel colour development and CI symptom development (red spots) (Table 3). These results are consistent with other findings for mango: CA storage of 'Kensington' fruit with 5–10% CO_2 alleviated CI symptoms (O'Hare and Prasad, 1993); short treatments of 'Tommy Atkins' fruit with low O_2 induced higher CO_2 levels and was effective in reduction of CI symptoms at 5°C (Pesis et al., 1997). It has been shown that atmosphere modification, either by CA or MA, can alleviate CI symptoms in many chilling-sensitive crops (Forney and Lipton, 1990).

Our results show that MA storage, including moderate relative humidity, eliminated the CI symptom of red spots around the lenticels in mangoes stored at 12°C. These result are in agreement with findings for 'Kensington' mango, that

Table 3
Effect of MA and CA treatments on peel colour development and chilling injury expressed as red spots in mango cv. Keitt[a]

Treatment	After 21 days at 12°C		After +5 days at 20°C	
	Waxed	Non-waxed	Waxed	Non-waxed
Peel colour index (1–10)				
Control	1.9 ± 0.56	1.9 ± 0.12	2.9 ± 0.66	3.6 ± 0.42
MA (XF.14A)	1.4 ± 0.31	2.0 ± 0.30	2.8 ± 0.37	3.5 ± 0.85
MA (XF.12A)	1.4 ± 0.12	1.6 ± 0.44	3.0 ± 0.00	2.9 ± 0.22
CA	1.4 ± 0.31	1.5 ± 0.20	2.9 ± 0.31	2.8 ± 0.37
Storage at 20°C	2.0 ± 0.27	3.8 ± 0.54	–	–
Red spots index (1–10)				
Control	3.1 ± 0.43	1.8 ± 0.33	3.7 ± 0.74	2.4 ± 0.71
MA (XF.14A)	1.4 ± 1.31	0.3 ± 0.12	0.6 ± 0.26	0.3 ± 0.04
MA (XF.12A)	0.8 ± 0.26	0.5 ± 0.36	0.5 ± 0.11	0.9 ± 0.65
CA	1.1 ± 0.27	0.6 ± 0.24	0.3 ± 0.24	0.4 ± 0.23
Storage at 20°C	0.4 ± 0.07	0.3 ± 0.10	–	–

[a] Fruit was stored for 21 days at 12°C, followed by 5 days at 20°C. Control fruit was also stored for 21 days at 20°C. Treatments were as described in the third experiment. The results are means of three boxes ± S.E.

MA storage reduced peel injury around the lenticels caused by calcium treatment (Yuen et al., 1993). Also MA can reduce CI in other chilling-sensitive fruits such as lime and grapefruit (Wardowski et al., 1973), avocado (Scott and Chaplin, 1978; Meir et al., 1997), and cucumber (Wang and Qi, 1997).

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