

Physiological changes in peaches related to chilling injury and ripening

J.P. Fernández-Trujillo, A. Cano, F. Artés *

Postharvest and Refrigeration Laboratory, Food Science and Technology Department, CEBAS-CSIC, PO Box 4195, E-30080, Murcia, Spain

Received 27 May 1997; accepted 16 January 1998

Abstract

Firm-breaker (FB) and firm-mature (FM) peaches cv. 'Paraguay' were either stored for 4 weeks at 2°C or subjected to three cycles of 1 day of intermittent warming (IW) at 20°C every 6 days at 2°C. Normal postharvest ripening and post-storage ripening at 20°C were also studied in order to relate postharvest physiology with the onset of chilling injuries (CI) (woolliness, gel breakdown and scald). As far as we know, both gel breakdown and scald have been described and reported on peaches for the first time. FB peaches were more sensitive to CI than FM ones. A high respiration rate and ethylene production in conventionally stored fruit after 2 weeks of storage, followed by a drop in ethylene production, was accompanied by the development of CI in fruit of both maturity stages. IW strongly reduced CI during storage. Periodic warming acclimatised chilled fruit to subsequent periods of chilling by allowing them to ripen due to the production of a suitable amount of ethylene, depending on their maturity stage at harvest. The increase in ethylene production during post-storage ripening could be related to the development of over-ripeness. © 1998 Elsevier Science B.V. All rights reserved.

Keywords: Chilling injuries; Woolliness; Gel breakdown; Scald; Intermittent warming; *Prunus persica*; Respiration; Ethylene; Pectolytic enzymes

1. Introduction

Chilling injury (CI) is a limiting factor in the shelf life of peaches stored at low temperatures (Lill et al., 1989; Luza et al., 1992; Artés et al., 1996). Low temperatures of around 2.5°C (Von Mollendorff and De Villiers, 1988a,b) normally exacerbate CI. Immature peaches are usually

* Corresponding author. Tel.: + 34 68 215717; fax: + 34 68 266613; e-mail: fr.artes@natura.cebas.csic.es

more susceptible to woolliness (internal breakdown) according to Kailasapathy and Melton (1992). Intermittent warming (IW) has been demonstrated to be a good method for alleviating CI during storage (Lill et al., 1989; Artés et al., 1996).

Slight but consistent differences have been found in the respiration rate between chilling-damaged and sound peaches (Ben-Arie and Lavee, 1972). In nectarines, ethylene production increased after transfer to ripening temperatures when storage at 0°C progressed. However, the loss of ethylene production capacity after prolonged storage could be influenced by CI (Brecht and Kader, 1984; Valero et al., 1997). The firmness and the respiration rate of peaches decreased at the onset of woolliness, while woolly peaches had a low ethylene production throughout the storage and ripening periods (Von Mollendorff and De Villiers, 1988a).

In a non-melting flesh clingstone peach cultivar, the development of CI during cold storage has been related to continuous activity of pectin-methyl-esterase (PME) during storage and inhibition of endo-polygalacturonase (endoPG) activity after the second week of storage. The exoPG activity did not seem to affect the development of CI (Artés et al., 1996). In melting flesh clingstone peaches, one of the questions is the possible linkage between the decrease in flesh firmness and the onset of a respiration increase, ethylene emission and pectolytic enzyme activities during ripening, and the usefulness of these physiological responses either to measure or to predict the effect of CI on fruit metabolism.

In the present work the extent of some physiological changes, particularly those related to the development of CI, both in conventionally or intermittently-warmed stored peaches, harvested at two stages of maturity, and in subsequently ripened peaches, has been determined.

2. Materials and methods

2.1. Plant material and experimental design

Firm-mature (FM) and firm-breaker (FB)

peaches (*Prunus persica* L. Batsch cv. 'Paraguay') were harvested on the 5th July at two different stages of maturity according to their skin ground colour (see below) from a commercial orchard in Cieza (Murcia). Fruit was transported by ventilated car 5 km to a packing house, where they were sorted and selected for uniform size, maturity and appearance and freedom from defects. Sound fruit were quickly transported by ventilated car in about 40 min to the laboratory, where they were immediately forced-air pre-cooled to reach 5°C (stone temperature) 5 h later, and 2°C 12 h later. Other fruit were subjected to normal ripening at 20°C. Temperatures were recorded at 15-min intervals with an LCD portable thermometer (Digi-Thermo, Tecnoquim, Spain) with an external sensing probe inserted beside the stone of each of five fruits. The following morning, peaches were randomly divided into batches of 32 fruits each and put in clean plastic boxes with the fruit touching. The mean measurements (\pm S.E.) of the FM fruit were: axial diameter 36.0 ± 0.4 mm; longitudinal diameter 62.4 ± 0.6 mm; weight 82.9 ± 2.6 g. For FB fruit these were: axial diameter 36.9 ± 0.9 mm; longitudinal diameter 61.5 ± 1.0 mm; weight 83.8 ± 4.5 g. Measurements of ground colour at harvest were recorded using standard C.I.E. L*a*b* colorSpace coordinates on three equidistant points of the equatorial zone of 15 fruits (Minolta CR-300 colorimeter, 8 mm ϕ viewing aperture, white plate reference plate, D65 standard C.I.E. illuminant, 2° observer). Hue angle [$H^* = \tan^{-1}(b^* \cdot a^{*-1})$] values of FB and FM fruit at harvest were 100.4 ± 1.2 and 94.9 ± 2.5 units respectively.

To induce CI, the peaches were placed in a cold room at $2.0 \pm 0.5^\circ\text{C}$ and 90–95% RH. IW was achieved by transferring the boxes to a room at 20°C and 95% RH. After 1 day of warming, the fruit were returned to 2°C (Artés and Escriche, 1994).

Treatments were: normal postharvest ripening for 11 days at 20°C and 90–95% RH; conventional storage for 1, 2, 3 or 4 weeks at 2°C as control; IW, storage at 2°C with four cycles of IW of 1 day at 20°C every 6 days of storage. The samples of IW and control fruit taken for analysis at weekly intervals were subjected to 3 days of post-storage ripening at 20°C and 70–75% RH,

simulating the normal period of retail sale, to measure the effects on the CI index (see below). Different samples of control or IW fruit stored for 2 and 3 weeks, or control fruit stored for 4 weeks, were then left for another 4 days in the same conditions to enable measurement of respiration and ethylene production.

2.2. Quality analysis

Quality measurements were made on three replicates of five fruits each, and included firmness (Fruit Pressure Tester Effegi 327 penetrometer, 7.9-mm probe tip, readings at 20°C), soluble solids content (Atago N1 hand refractometer, readings at 20°C), titratable acidity (Artés et al., 1996) ($\text{mmol H}^+ \text{ l}^{-1}$) and extractable juice (Von Mollendorff and De Villiers, 1988a). The measurements were made at harvest, after 3 days of ripening at 20°C and 90–95% RH, and after 3 weeks of storage at 2°C and 95% RH plus 3 days of post-storage ripening at 20°C and 70–75% RH.

2.3. Chilling injury indices

The degree of CI was assessed by eye and fruit were divided into five classes (0: absent (A); 1: very slight (VS), 2: slight (S); 3: moderate (M); 4: severe (E)). Symptoms are described in the Results section.

Moderate to severe injured fruit were not commercial since these disorders affected >20% of the mesocarp surface in a double cut parallel to the axial diameter and between themselves (opposite sides of the stone). Very slight or slight woolliness resulted in visible juice on the cut surface of the fruit, but with slight pulp disruption and pastiness. In the cut surface of the fruit with moderate or severe woolliness, flesh appeared dry on the cut, pulp was mealy and with evident disruption, and free juice was barely detectable or even undetectable (Rushing and Dinamarca, 1993).

The CI index recording the extent of damage on a 0–100 scale was calculated as follows: $\text{CI index} = (1*N_{\text{VS}} + 2*N_{\text{S}} + 3*N_{\text{M}} + 4*N_{\text{E}})/4$ where N_{VS} , N_{S} , N_{M} and N_{E} were the percentages of fruit showing the different degrees of CI. The CI index

was evaluated weekly in three replicates of 32 fruits during storage and after further 3 days of ripening at 20°C. The severity of the damage was expressed by CI losses recorded as the percentage of fruit on a fresh weight basis with moderate and severe symptoms.

2.4. Respiration rate and ethylene production

For CO_2 and C_2H_4 measurements, five replicates of two fruits each were held in 620 ml gas-tight jars for 1 h at 2°C or 20 min at 20°C, prior to gas sampling. The gas samples collected were 1 ml for CO_2 and 5 ml for C_2H_4 . Measurements of C_2H_4 were made with a Perkin Elmer gas chromatograph with FID detection. For CO_2 , a Hewlett Packard 5730A gas chromatograph was used.

The measurement error was about 0.1% for CO_2 and 1.5% for C_2H_4 , with a detection limit of 0.01 ppm of C_2H_4 and 0.01% of CO_2 . With this method ethylene production levels of up to $0.025 \mu\text{l C}_2\text{H}_4 \text{ kg}^{-1}$ fruit were detected.

2.5. Pectolytic enzyme activities

Pectolytic enzymes during ripening were extracted by homogenising peach flesh in an aqueous solution containing 0.5 M NaCl and 1% polyvinylpyrrolidone. The homogenates were centrifuged at 12000 rpm for 10 min, filtered and used for the assays (Artés et al., 1996). ExoPG and endoPG (PG, EC 3.2.1.15) activities were determined according to Artés et al. (1996). PME (EC 3.1.1.11) activity was evaluated according to Hagerman and Austin (1986).

2.6. Statistical analysis

Experiments were carried out in a completely randomized design. A 2-way analysis of variance (ANOVA) was carried out on quality parameters with maturity and storage treatments as factors. Firmness was transformed to their respective log for anova according to their normal probability plot. LSD or Duncan's multiple range test were used to compare treatment means (Snedecor and Cochran, 1980).

Table 1

Chilling injury index of firm-mature (FM) and firm-breaker (FB) peaches after storage at 2°C with or without 3 days of post-storage ripening at 20°C

Treatments and maturity stage	Chilling injury index							
	Continuous storage (weeks)				Intermittent warming (weeks)			
	1	2	3	4	1	2	3	4
End of cold storage								
FM	1a*	2a	9b	12b	0a	1a	0a	10b
FB	1a	0a	16c	40d	0a	0a	3b	3b
End of post-storage ripening								
FM	0a	14b	69c	71c	0a	0a	0a	10b
FB	0a	34b	82c	74c	0a	1a	2a	7b*

* Mean separation within rows by Duncan multiple range test ($P = 0.05$).

3. Results

3.1. Chilling injury and senescence symptoms

CI was mainly developed as woolliness (dry and mealy tissues) and to a lesser extent gel breakdown and scald. Gel breakdown developed primarily around the stone, reaching in some cases, a large part of the mesocarp. The disorder was characterised by gelatinous texture, a typical water-soaked and vitreous appearance and dark olive-green colour accompanied by slight reddening to browning. Scald was identified by dry and brown tissues forming a crown in the cortical part of the mesocarp (subepidermal tissues). As far as we know, both CI symptoms have not been previously described in the scientific literature on peaches, separately from woolliness (or internal breakdown). When woolliness progressed (after 2 weeks of storage), gel breakdown or scald were frequently indistinguishable from woolliness.

Early symptoms of CI during the first week of storage were negligible (< 1 unit of CI index). After 1 week of storage of control fruit, early CI symptoms such as slight brown punctures in the subepidermal tissues appeared. With increasing time, damaged tissues became slightly brown and dry, accompanied by a cork-like texture starting at the cortical tissue near the skin (woolliness), which progressed until reaching the pit cav-

ity. In FM fruit, very slight symptoms of damage in the cortical tissue (scald) also appeared after 1 week of storage, but these primary symptoms disappeared after 3 days of post-storage ripening, showing that the injury was still reversible.

Gel breakdown symptoms were not visible until after 2 weeks of storage in FM fruit. After 2 weeks of storage plus post-storage ripening, all CI losses in control FM fruit (Table 2) were caused by gel breakdown, which did not appear in fruit subjected to IW at any time. After 2 or 3 weeks plus post-storage ripening (in control FB and FM fruit respectively), losses due to woolliness in control fruit ranged from 23 to 69% for FB fruit or from 46 to 55% for FM fruit. In IW fruit, losses from woolliness appeared to a lesser extent (around 3.1% after 4 weeks plus post-storage ripening). Losses from gel breakdown ranged from 9 to 20% (FM control fruit) or 7 to 24% (FB control fruit), while in the IW treatment, gel breakdown only appeared in FB fruit at less than 3.1%. Losses from scald only appeared in FB fruit after 2 weeks plus post-storage ripening ranging from 2 to 7%, although under IW this disorder did not cause losses.

FB peaches were generally more sensitive to CI than FM peaches (Tables 1 and 2). IW fruit at both stages of maturity were free from woolliness up to 3 weeks of storage with or without post-

Table 2

Chilling injury losses of firm-mature (FM) and firm-breaker (FB) peaches after storage at 2°C with or without 3 days of post-storage ripening at 20°C

Treatments and maturity stage	Losses (% on a fresh weight basis)							
	Continuous storage (weeks)				Intermittent warming (weeks)			
	1	2	3	4	1	2	3	4
End of cold storage								
FM	0a*	2.2b	7.2c	8.3c	0a	0a	0a	0a
FB	0a	0a	8.2b	26.4c	0a	0a	4.1b	4.1b
End of post-storage ripening								
FM	0a	8.7b	64.4c	63.4c	0a	0a	0a	3.0b
FB	0a	8.6b	80.1c	71.8d	0a	1.0a	0.9a	3.2b*

* Mean separation within rows by Duncan multiple range test ($P = 0.05$).

storage ripening. However, IW induced shrivelling and over-ripeness (senescence symptoms) after 4 weeks of storage, causing 7.5% losses in FB peaches and 12% in FM peaches. Compared with IW fruit, control fruit only showed slight symptoms of senescence.

3.2. Respiration rate and ethylene production

A characteristic climacteric onset was found in fruit at 20°C as well as in those undergoing post-storage ripening (Figs. 1 and 2). Compared with fruit under normal postharvest ripening at 20°C, the climacteric rise during subsequent post-storage ripening was earlier and more pronounced. Ethylene production in fruit during normal postharvest ripening at 20°C and during post-storage ripening of control and IW fruit at the different times assayed followed the typical pattern, with a maximum peak almost coincident with the respiratory peak, although the preclimacteric phase was longer in fruit ripened after harvest.

The slightly less pronounced ethylene peak in FM fruit compared with FB fruit could be related to the development of over-ripeness in the former, as occurred in IW fruit ripened after 3 weeks of storage (Fig. 1).

After 2 weeks of storage, respiration and

ethylene production in control FM fruit were higher than in IW fruit (Fig. 1), although the differences were more pronounced for ethylene. In FB fruit ripened after 2 weeks of storage, these differences were only detected in respiration. The maximum CI levels observed in the third week of storage were followed by a loss of ethylene production in control fruit at this time and during the post-storage ripening after 4 weeks at 2°C (Figs. 1 and 2).

The higher incidence of CI (mainly woolliness) in control fruit of both maturity stages after 4 weeks of storage was accompanied by a sharp decrease in ethylene production rather than by a high respiration rate during post-storage ripening (Figs. 1 and 2). The higher the incidence of CI (in FB control fruit), the lower the ethylene production during post-storage ripening will be.

IW fruit of both maturity stages showed a high level of ethylene production for 6 days after the third warming period, whereas in control fruit, this rapidly decreased. Such behaviour might be associated both with the normal ripening and senescence of IW fruit and with CI in control fruit.

3.3. Quality measurements

As can be expected, all parameters changed

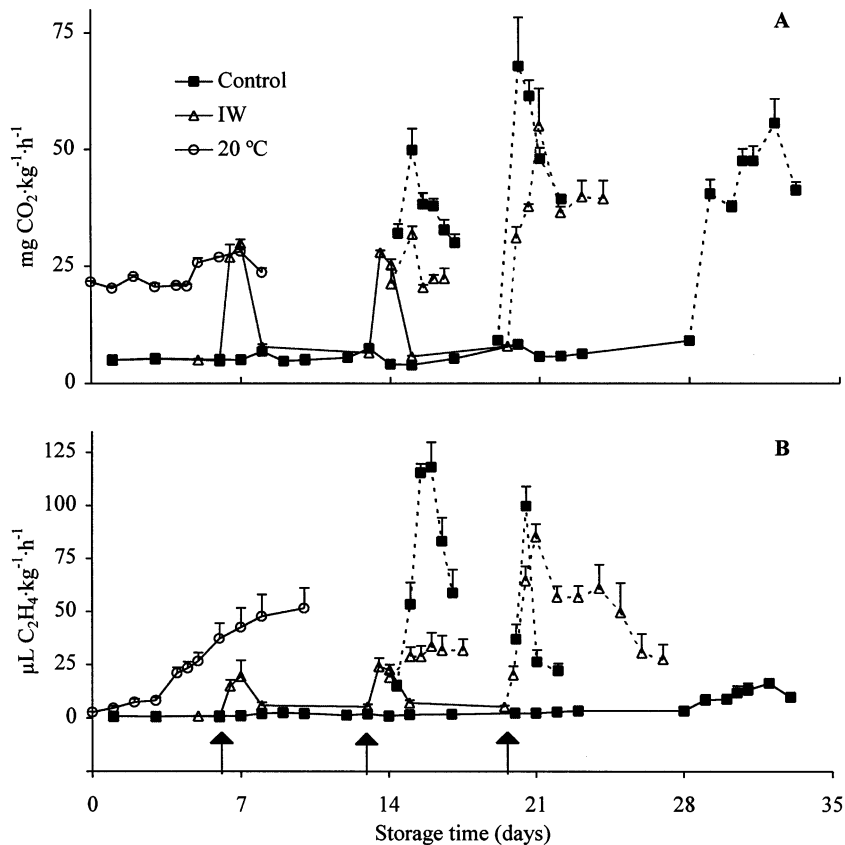


Fig. 1. Respiration rate (A) and ethylene production (B) during postharvest ripening at 20°C, conventional storage at 2°C (control) or storage at 2°C with intermittent warming cycles of 1 day at 20°C every 6 days (IW) in firm-mature peaches ($n = 5$). Arrows indicate IW. Dashed lines represent ripening at 20°C. Bars represent \pm S.E.

compared with values at harvest. Flesh firmness and titratable acidity were lower in FM than in FB fruit ($P < 0.01$), while total soluble solids (TSS) values were higher in FM fruit (Table 3).

Compared with fruit at 20°C, IW fruit showed normal quality parameters after storage mainly due to the maintenance of extractable juice (Table 4). In general, abnormal ripening of control fruit resulted in reduced flesh firmness. TSS slightly increased in both treatments during storage, although variability of the results in FM fruit subjected to ripening for 3 days at 20°C or IW was higher. Compared with control fruit, IW fruit after two warming periods plus 3 days at 20°C maintained an acceptable flesh firmness and total acidity (TA) during storage.

3.4. Pectolytic enzyme activities and flesh firmness during normal postharvest ripening

A significant difference in exoPG activity was observed in FM fruit compared with the firmest FB ones (Fig. 3C and D). In this cultivar, PME activity was considerably higher in both stages of maturity when compared with that in a clingstone cultivar (Artés et al., 1996) (Fig. 3A).

In FM 'Paraguay' peach, the climacteric phase and the upsurge in ethylene production after 4 days of storage was accompanied by an increase in PME and endoPG activities and a subsequent but not equivalent response in exoPG activity (Figs. 1 and 3). However, in FB peaches these changes were relatively lower, particularly with PG isoenzymes.

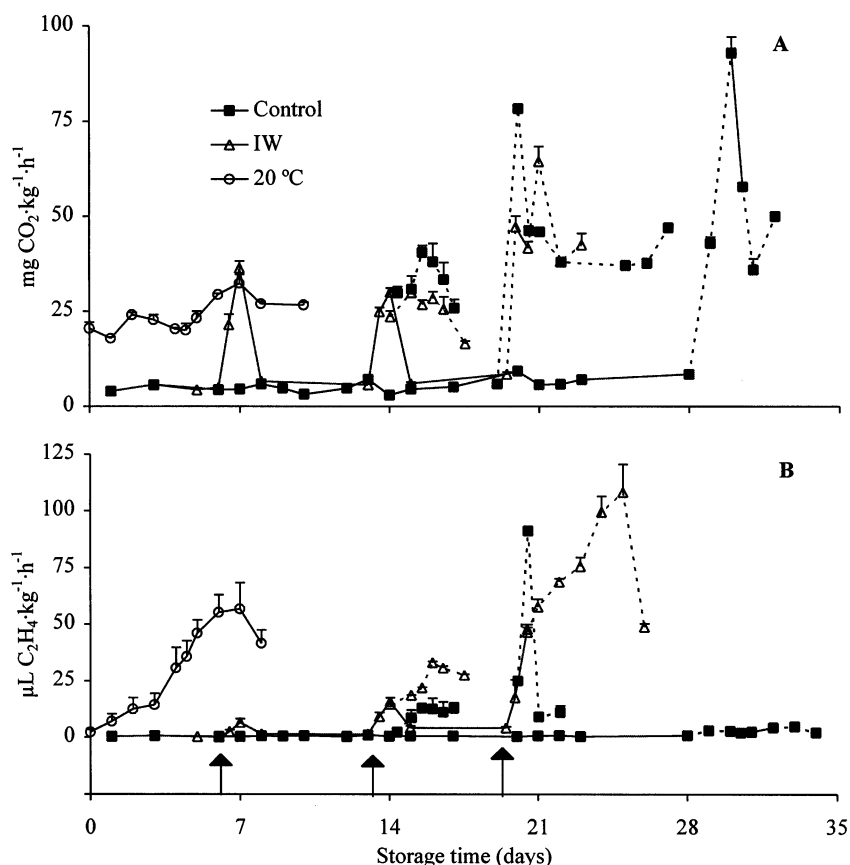


Fig. 2. Respiration rate (A) and ethylene production (B) during postharvest ripening at 20°C, conventional storage at 2°C (control) or storage at 2°C with intermittent warming cycles of 1 day at 20°C every 6 days (IW) in firm-breaker peaches ($n = 5$). Arrows indicate IW. Dashed lines represent ripening at 20°C. Bars represent \pm S.E.

4. Discussion

Compared with FM fruit, the higher susceptibility of FB fruit to CI confirmed previous reports in other cultivars (Lill et al., 1989; Kailasapathy and Melton, 1992). The main effect of IW was the reduction of losses due to moderate and severe symptoms of CI until the fourth week of storage by promoting a gradual ripening of the fruit, thus confirming previous reports (Wang, 1993; Dawson et al., 1995; Artés et al., 1996). In fruit subjected to IW, the symptoms that appeared at the end of 4 weeks of storage were probably caused by the failure of a few fruit to ripen (a typical CI in peaches) as reported by Wang (1993), and/or by variations in

the degree of maturity at harvest and/or by the variable effect IW can have on some fruit (as shown in TSS measurements).

The lower extractable juice confirmed the woolliness in FB control fruit, as was the case with juice content of mealy nectarines when allowed to ripen (Harker and Sutherland, 1993). However, in FM fruit the lack of correlation between CI (Tables 1 and 2) and extractable juice (interaction maturity \times storage conditions significant at $P < 0.001$, Table 4) could be related to the onset of gel breakdown (index of 12) and scald (index of 2), being particularly higher at this time (all CI losses due to gel breakdown), without any symptom of woolliness in the mesocarp.

Table 3

Fruit quality of firm-mature (FM) and firm-breaker (FB) peaches after 3 days at 20°C, or 3 weeks of storage at 2°C plus 3 days of post-storage ripening at 20°C

Treatments and maturity at harvest		Flesh firmness (N)	Titrateable acidity (mmol H ⁺ l ⁻¹)	Total soluble solids (°Brix)	Extractable juice (% fw)
At harvest	FM	52 ± 3	34 ± 1	14.3 ± 0.2	65.9 ± 1
	FB	95 ± 4	42 ± 1	13.3 ± 0.2	73.0 ± 3
Normal ripening	FM	38 ± 2	31 ± 1	14.0 ± 0.8	47.8 ± 1
	FB	59 ± 1	40 ± 1	13.2 ± 0.2	48.2 ± 2
Conventional storage (2°C)	FM	12 ± 1	30 ± 1	15.5 ± 0.3	63.5 ± 1
	FB	23 ± 2	35 ± 1	14.9 ± 0.2	34.6 ± 4
Intermittent warming	FM	19 ± 1	26 ± 1	15.1 ± 1	60.4 ± 4
	FB	28 ± 1	35 ± 2	14.1 ± 0.3	49.3 ± 5
LSD (<i>P</i> = 0.01)		8.0	5	0.8	10.1

Data are means ± S.E. of three (titrateable acidity and total soluble solids), 15 (flesh firmness) and four (extractable juice) replications. Mean separation within columns by LSD test.

The reduction in CI and respiration after 2 weeks of storage induced by IW agrees with data of Anderson (1982). When control and IW fruit were compared, the higher respiration rate observed in the former during ripening after 2 weeks of storage suggested the development of an irreversible metabolic pathway. This fact is often correlated with poor keeping quality (Lill et al., 1989), and is accompanied by the appearance of CI symptoms. The lower respiration of IW fruit compared with control fruit contrasted with findings in delayed storage of peaches (Ben-Arie and Lavee, 1972; Von Mollendorff and De Villiers, 1988a), who reported higher respiration rates in warmed peaches than in control ones during storage and after post-storage ripening.

Peach pericarp tissue is known to be anisotropic (Luza et al., 1992), with differences in the ultrastructural composition of the cell wall of the outer and inner tissues of the mesocarp, as has also been reported in plums (Taylor et al., 1995). In peaches, a gradient between the epicarp and mesocarp tissue in terms of firmness (highly cultivar dependent), and in ethylene biosynthesis capability between pulp and peel (ACC concentration, efficiency in converting ACC to ethylene and ethylene production), has been reported (Amorós et al., 1988; Maness et al., 1992; Tonutti et al., 1996). These facts, and the relationship between

CI and ethylene production observed in this experiment, support the hypothesis that CI in the form of scald and gel breakdown, is strongly associated with maturity, as are other disorders previously reported for stone fruit such as peeling-injury in peaches (Sharkey and Pegg, 1984) and gel breakdown in plums (Taylor et al., 1995).

The reduced ethylene production during the post-storage period of FB control fruit compared with IW fruit after 2 or 3 weeks of storage could be related to the immature stage of these fruits (as also occurred in FB fruit subjected to normal postharvest ripening), since the exposure to IW brought forward ripening. However, this was matched by a greater increase in the CI of FB fruit compared with that showed by FM fruit (Table 1).

The reduced ethylene production in control peaches was probably due to damage in the ethylene synthesising system at temperatures which cause a high incidence of CI, confirming previous reports on ethylene production changes in peaches (Wade, 1981; Sharkey and Pegg, 1984; Von Mollendorff and De Villiers, 1988a; Valero et al., 1997) and nectarines (Brecht and Kader, 1984). Also the deterioration of ethylene production after 3 or 4 weeks of storage in both maturity stages could be associated with a possible accumulation of polyamines at low tempera-

Table 4

Analysis of variance (in percentage of the total sum of squares and probability^a) of fruit quality of firm-mature and firm-breaker peaches (maturity stage) at harvest, after 3 days at 20°C or after 3 weeks of storage at 2°C plus 3 days of post-storage ripening at 20°C (control or intermittent warming)

Source of variation	df ^b	Flesh firmness	Titrateable acidity	Extractable juice	Total soluble solids
Maturity (M)	1	12.7****	29.0***	3.1 ns	57.2****
Storage condition (S)	3	84.5****	55.3****	42.6****	27.2***
M*S	3	1.0**	2.5 n.s.	30.5****	1.0 n.s.
Residual	16 ^a	1.8	13.2	23.8	14.6

^a n.s. not significant.

^b Degrees of freedom. For extractable juice ($n = 4$), $df = 31$ for residual.

** $P < 0.01$.

*** $P < 0.001$.

**** $P < 0.0001$.

tures (Artés et al., 1996). In fact, polyamines inhibited ethylene production in peaches (Biggs et al., 1982) and an increase in putrescine with accumulation of spermidine during storage at 5°C was concomitant with the onset of woolliness, particularly in unripe fruit (Valero et al., 1997).

Levels of exoPG and flesh firmness were not strongly associated in 'Paraguay' either, because FB fruit were unable to reach the same level as FM fruit at the same softening stage. In FB peaches, exoPG activity was lower than in FM peaches because of the known accumulation of exoPG in very soft (less than 30 N) peaches, as a response to an increase in endoPG activity (Downs et al., 1992; Artés et al., 1996). According to the results obtained by Orr and Brady (1993) and Tonutti et al. (1994) in other peach cultivars, endoPG activity and textural changes in 'Paraguay' are not closely linked.

In 'Paraguay' peaches subjected to normal postharvest ripening the ethylene increase to 20 $\mu\text{l C}_2\text{H}_4 \text{ kg}^{-1} \text{ h}^{-1}$ took place relatively at about the same time in both types of fruit (Figs. 1 and 2), and preceded the climacteric peak. Only a slight rise in ethylene production was concomitant with a strong decrease in flesh firmness in FB fruit (loss of 45 N), the flesh firmness being lower than 70 N when the strong upsurge in ethylene production appeared. These results agree with those Tonutti et al. (1994, 1996) and Valero et al. (1997) who suggested that the initial decrease in flesh firmness

occurred without any change in ethylene production and before an increase in PG activities. Bonghi et al. (1992) showed that an exogenous treatment with ethylene of 0.8 l air min^{-1} containing 100 $\mu\text{l C}_2\text{H}_4 \text{ l}^{-1}$ in abscising fruit explants during 5 days at room temperature, which caused an increase in mRNA encoding endoPG, preferentially activating this isoenzyme and the basic exoPG, although acid exoPG activity was depressed.

On the other hand, PG activity was lower during storage in fruit more susceptible to CI (Von Mollendorff and De Villiers, 1988b; Artés et al., 1996). However, PG activity increased sharply during post-storage ripening at 10°C in peaches which developed woolliness with similar levels to those in sound fruit after 1 week at 10°C (Von Mollendorff and De Villiers, 1988b; Lill et al., 1989). EndoPG activity was not deactivated in IW cycles which alleviated CI (Artés et al., 1996). The higher PME activity in both types of 'Paraguay' peaches compared with other clingstone cultivars (Artés et al., 1996) could be the cause of rapid woolliness development in 'Paraguay', as has also been shown in chilled tomatoes (Marangoni et al., 1995). According to Artés et al. (1996), continuous PME activity causing the accumulation of de-esterified pectate during storage and a possible deterioration of endoPG activity would result in faster CI development and lower flesh firmness in control fruit.

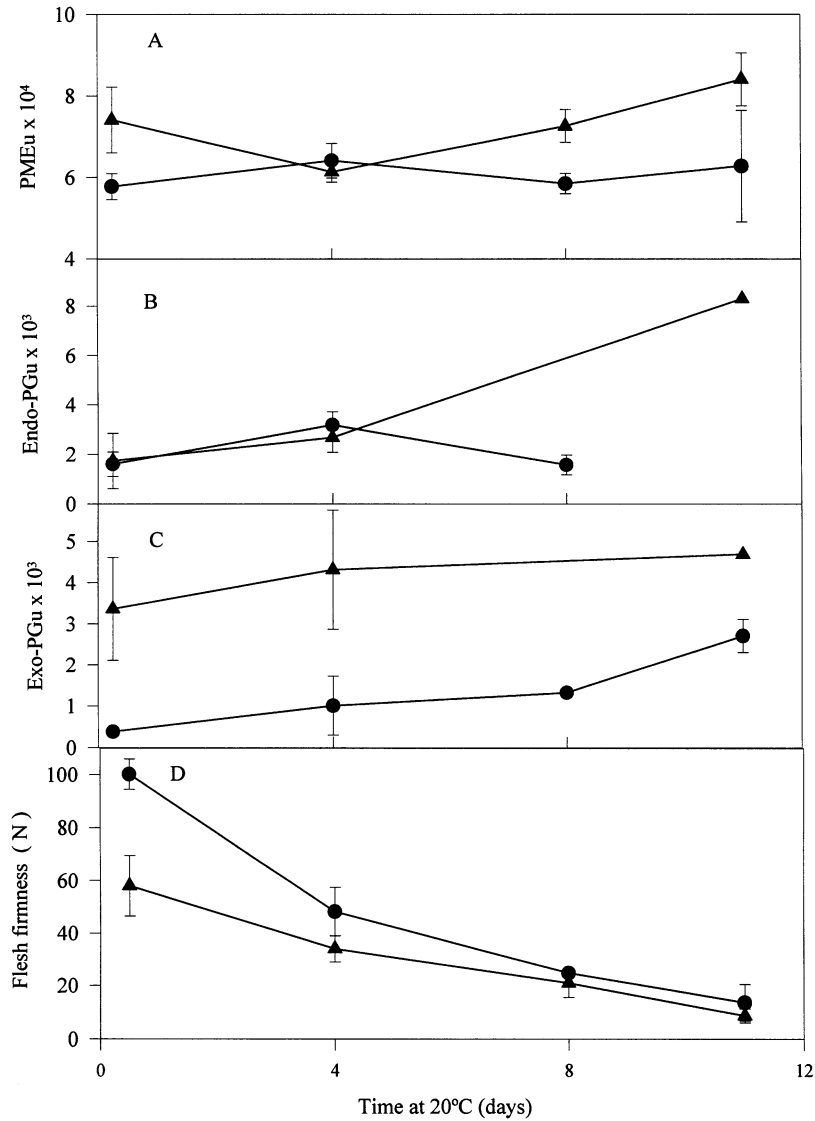


Fig. 3. Pectolytic enzyme activities (mean \pm S.E., $n = 2$) and flesh firmness ($n = 3$) in firm-breaker (●) and firm-mature (▲) peaches during normal postharvest ripening at 20°C. (A) PME activity, PEu is defined as milliequivalents of H^+ $ml^{-1} h^{-1}$. (B) endoPG activity and (C) exoPG activity, PGu is defined as $\mu M ml^{-1} h^{-1}$. (D) Flesh firmness.

Acknowledgements

Financial support was provided by CICYT (ALI-95/530 Project) and Iltmo. Ayto. de Cieza (Murcia). J.P. Fernández-Trujillo is indebted to

Spanish CSIC for a PhD grant. We thank Mr A. Hernández and Mr J.G. Marín for technical help and Cofrucieza S. Coop. and Ciezana de Frutas S. Coop. for supplying the peaches and other facilities.

References

- Amorós, A., Vivancos, G., Riquelme, F., Romojaro, F., 1988. Estudio del metabolismo del etileno en variedades de melocotón de carne blanda y dura. Symposium 'Maduración y Postrecolección'. Murcia, Spain, pp. 35–40.
- Anderson, R.E., 1982. Long-term storage of peaches and nectarines intermittently warmed during controlled-atmosphere storage. *J. Am. Soc. Hortic. Sci.* 107, 214–216.
- Artés, F., Cano, A., Fernández-Trujillo, J.P., 1996. Pectolytic enzyme activity during intermittent warming storage of peaches. *J. Food Sci.* 61, 311–313.
- Artés, F., Escriche, A.J., 1994. Intermittent warming reduces chilling injury and decay of tomato fruit. *J. Food Sci.* 59, 1053–1056.
- Ben-Arie, R., Lavee, S., 1972. On the relation between respiration and woolly breakdown of stored Alberta peaches. *Phyton* 30, 127–134.
- Biggs, R.H., El-Agamy, S.Z.A., Aly, M.M., 1982. Ethylene production by mature peach fruit. *Hortscience (Abstr.)* 17, 62.
- Bonghi, C., Rascio, N., Ramina, A., Casadoro, G., 1992. Cellulase and polygalacturonase involvement in the abscission of leaf and fruit explants of peach. *Plant Mol. Biol.* 20, 839–848.
- Brecht, J.K., Kader, A.A., 1984. Regulation of ethylene production by ripening nectarine fruit as influenced by ethylene and low temperature. *J. Am. Soc. Hortic. Sci.* 109, 869–872.
- Dawson, D.M., Watkins, C.B., Melton, L.D., 1995. Intermittent warming affects cell wall composition of 'Fantasia' nectarines during ripening and storage. *J. Am. Soc. Hortic. Sci.* 120, 1057–1062.
- Downs, C.G., Brady, C.J., Gooley, A., 1992. Exopolygalacturonase protein accumulates late in peach fruit ripening. *Physiol. Plant* 85, 133–140.
- Hagerman, A.E., Austin, P.J., 1986. Continuous spectrophotometric assay for plant pectin methyl esterase. *J. Agric. Food Chem.* 34, 440–444.
- Harker, F.R., Sutherland, P.W., 1993. Physiological changes associated with fruit ripening and the development of mealy texture during storage of nectarines. *Postharvest Biol. Technol.* 2, 269–277.
- Kailasapathy, K., Melton, L.D., 1992. Woolliness in stone fruits. *Asean Food J.* 7, 13–16.
- Lill, R.E., O'Donoghue, E.M., King, G.A., 1989. Postharvest physiology of peaches and nectarines. *Hortic. Rev.* 11, 413–452.
- Luza, J.G., Gorsel, R., Polito, V.S., Kader, A.A., 1992. Chilling injury in peaches: a cytochemical and ultrastructural cell wall study. *J. Am. Soc. Hortic. Sci.* 117, 114–118.
- Maness, N.O., Brusewitz, G.H., McCollum, T.G., 1992. Internal variation in peach fruit firmness. *HortScience* 27, 903–905.
- Marangoni, A.G., Jackman, R.L., Stanley, D.W., 1995. Chilling-associated softening of tomato fruit is related to increased pectinmethylesterase activity. *J. Food. Sci.* 60, 1277–1281.
- Orr, G., Brady, C.J., 1993. Relationship of endopolygalacturonase activity to fruit softening in a freestone peach. *Postharvest Biol. Technol.* 3, 121–130.
- Rushing, J.W., Dinamarca, A., 1993. Simulated shipment of peaches, plums and nectarines under controlled atmospheres. *Proc. VI Int. Res. CA Conf. Cornell Univ., NRAES-71*. Ithaca, NY, pp. 202–222.
- Snedecor, G.W., Cochran, W.G., 1980. *Statistical methods*, 7th ed. Iowa State University Press, Ames.
- Sharkey, P.J., Pegg, I.D., 1984. Effect of high-humidity storage on quality, decay and storage life of cherry, lemon and peach fruits. *Sci. Hortic.* 23, 181–190.
- Taylor, M.A., Rabe, E., Jacobs, G., Dodd, M.C., 1995. Effect of harvest maturity on pectic substances, internal conductivity, soluble solids and gel breakdown in cold stored 'Songold' plums. *Postharvest Biol. Technol.* 5, 285–294.
- Tonutti, P., Bonghi, C., Ramina, A., 1994. Biochemical and molecular aspects of peach fruit ripening. In: Hribar, J., Johnson, D.S., Böhling, H., De la Plaza, J., Haffner, K.E., Höhn, E., Somogyi, Z. (Eds.), *Proceedings of the COST-94 Quality criteria*. Bled, Slovenia, pp. 101–104.
- Tonutti, P., Bonghi, C., Ramina, A., 1996. Fruit firmness and ethylene biosynthesis in three cultivars of peach (*Prunus persica* L. Batsch). *J. Hortic. Sci.* 71, 141–147.
- Valero, D., Serrano, M., Martínez-Madrid, M.C., Riquelme, F., 1997. Polyamines, ethylene and physicochemical changes in low-temperature-stored peach (*Prunus persica* L. Cv. Maycrest). *J. Agric. Food Chem.* 45, 3406–3410.
- von Mollendorff, L.J., De Villiers, O.T., 1988a. Physiological changes associated with the development of woolliness in 'Peregrine' peaches during low temperature storage. *J. Hortic. Sci.* 63, 47–51.
- von Mollendorff, L.J., De Villiers, O.T., 1988b. Role of pectolytic enzymes in the development of woolliness in peaches. *J. Hortic. Sci.* 63, 53–58.
- Wade, N.L., 1981. Effects of storage atmosphere, temperature and calcium on low temperature injury of peach fruit. *Sci. Hortic.* 15, 145–154.
- Wang, C.Y., 1993. Approaches to reduce chilling injury of fruits and vegetables. *Hortic. Rev.* 15, 63–95.