

Postharvest quality and chilling injury of plums: benefits of 1-methylcyclopropene

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Abstract

The aim of this work was to evaluate the effect of 1-methylcyclopropene (1-MCP) treatment on the development of chilling injury (CI) symptoms in four plum cultivars and to determine the relationship between the climacteric behavior of the cultivar and its sensitivity to this disorder. Significant differences in ripening pattern were found between the cultivars after long-term storage. Among the climacteric cultivars, 'Royal Zee' plums showed a higher ethylene production rate than 'Linda Rosa' and 'Friar' cultivars. On the other hand, the 'Angeleno' cultivar behaved as a suppressed climacteric type. The development of translucency symptoms was higher in 'Royal Zee' than in 'Linda Rosa' and 'Friar' plums, and was almost absent in the suppressed climacteric cultivar. 1-MCP treatment significantly reduced ethylene production and the percentage of fruit affected by translucency in all climacteric cultivars. This treatment also delayed the ripening of the fruit during shelf life. In contrast, 1-MCP treatment did not affect the quality of 'Angeleno' plums. Collectively these results suggest that the development of chilling injury in plums is related to the climacteric behavior of the cultivar and demonstrated the beneficial effects of 1-MCP maintaining plum quality during storage.

Additional key words: cold storage; ethylene production; firmness; *Prunus salicina*; ripening pattern; translucency

Resumen

La calidad postcosecha y los daños por frío en ciruelas: beneficios del 1-metilciclopropeno

El objetivo de este trabajo fue evaluar el efecto del tratamiento con 1-metilciclopropeno (1-MCP) sobre el desarrollo de los síntomas de daños por frío (DF) en cuatro cultivares de ciruela y determinar la relación entre el patrón de maduración de los cultivares y su sensibilidad a este desorden. Se encontraron diferencias significativas en el patrón de maduración de los cultivares después de un almacenamiento a largo plazo. Entre los cultivares climatéricos, 'Royal Zee' mostró una tasa de producción de etileno superior a la de los cultivares 'Linda Rosa' y 'Friar'. Por otra parte, el cultivar 'Angeleno' presentó un comportamiento climatérico suprimido. El desarrollo de síntomas de transparencia fue mayor en 'Royal Zee' que en 'Linda Rosa' y 'Friar', y casi ausente en el cultivar climatérico suprimido. El tratamiento con 1-MCP redujo significativamente la producción de etileno y el porcentaje de frutos afectados por la transparencia en todos los cultivares climatéricos. Este tratamiento también retrasó la maduración de la fruta durante la vida en estante. En cambio, el tratamiento 1-MCP no afectó a la calidad de ciruelas 'Angeleno'. En conjunto, estos resultados sugieren que el desarrollo de DF en ciruelas está relacionado con el comportamiento climatérico del cultivar y demuestran los efectos beneficiosos del tratamiento 1-MCP sobre la calidad de las ciruelas durante el almacenamiento.

Palabras clave adicionales: firmeza; frigoconservación; patrón de maduración; producción de etileno; *Prunus salicina*; transparencia.

Introduction

High softening rates and susceptibility to chilling injury are the main factors limiting plums' commercial

life (Taylor *et al.*, 1993). Refrigeration is currently the most widely-used technology to reduce fruit softening, being temperatures below 0°C but above freezing point the most suitable (Crisosto *et al.*, 1999). However,

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Abbreviations used: CI (chilling injury), MACC (1-malonyl-aminocyclopropane-1-carboxylic acid), SSC (soluble solid content), TA (titratable acidity), 1-MCP (1-methylcyclopropene).

long-term storage under these conditions promotes the development of chilling injury (CI) symptoms such as flesh translucency and browning. Symptoms usually appear during the ripening period at 20°C, with consumers who detect this problem and complain. Flesh translucency is one of the most frequently observed symptoms in plums; it presents itself as a translucent and gelatinous area around the stone, also characterized by a loss of juiciness (Taylor *et al.*, 1993).

The physiological basis of CI in plums remains to be better explained. According to Lyons (1973), CI is primarily related to modifications in membrane permeability associated with a membrane-lipid transition from a flexible liquid-crystalline to a solid-gel structure. This first event would lead to a cascade of secondary events, resulting in a loss of regulatory control and metabolism imbalance, cell autolysis, and finally the development of a variety of chilling symptoms (Wang, 1989). Flesh translucency is also associated to loss of membrane permeability but also to an increase in soluble pectin that forms a gel in the intercellular spaces causing the translucency symptoms (Taylor *et al.*, 1993).

Previous study (Larrigaudière *et al.*, 2009) has demonstrated that CI symptoms in 'Larry Ann' plums are probably not caused by an oxidative stress, but related to changes in ethylene metabolism and especially in the levels of 1-malonyl-aminocyclopropane-1-carboxylic acid (MACC). In general, ethylene has been related to the development or worsening of different physiological disorders associated to chilling injury (Kader, 1985). 1-methylcyclopropene (1-MCP), an inhibitor of ethylene action, is effective to reduce the appearance of chilling injury in pineapples (Selvarajah *et al.*, 2001) and in different avocado cultivars (Pesis *et al.*, 2002). This treatment can also reduce the incidence of translucency in 'Blackamber' (Candan and Calvo, 2005), 'Fortune' (Menniti *et al.*, 2006) and 'Larry Ann' (Candan *et al.*, 2008) plum cultivars. However, this effect cannot be generalized, as both susceptibility to chilling injury (Crisosto *et al.*, 1999; Candan *et al.*, 2008) and response to 1-MCP treatment (Abdi *et al.*, 1998; Martínez-Romero *et al.*, 2003; Valero *et al.*, 2005) are cultivar-dependent.

The aim of this work was to evaluate the effect of 1-MCP treatment on the development of CI symptoms in four plum cultivars and to determine the relationship between the climacteric behavior of the cultivar and its sensitivity to this disorder.

Material and methods

Plant material

Japanese plums (*Prunus salicina* L.) of 'Royal Zee', 'Linda Rosa', 'Friar' and 'Angelino' cultivars were harvested from a commercial orchard in Río Negro (Argentina), according to fruit firmness. The fruit were immediately taken to the laboratory, where maturity at harvest was determined on 3 repetitions of 20 fruit. Fruit of two homogeneous lots from each cultivar were treated with 0 (Control) or 0.40 $\mu\text{L L}^{-1}$ of 1-MCP for 24 hours during fruit-cooling. The fruits were stored at 0°C and 90 % RH for 30 and 50 days and the quality evaluated after removal from the chamber and after 3 and 7 days of shelf life at 20°C on 3 repetitions of 20 fruit each.

Ethylene production

Ethylene production rate ($\mu\text{L kg}^{-1} \text{h}^{-1}$) was determined right after harvest and after storage on 3 repetitions of 6 fruit, during 21 days or until the climacteric maximum was reached. The fruit were weighed and enclosed in 3-L jars for 30 min at 20°C and then, 1 mL sample was extracted from the headspace. The sample was analysed with a gas chromatograph (GC-14A, Shimadzu, Japan) equipped with an alumina column (40°C) and a FID detector (210°C). Helium was used as carrier gas.

Firmness, soluble solids content (SSC), titratable acidity (TA) and epidermis color

Flesh firmness was determined with an electronic fruit texture analyzer (FTA-GS14, Güss, South Africa) with an 8 mm-diameter plunger, on both cheeks of the fruit after skin removal. Two slices of flesh were taken from each fruit and juiced to determine SSC (%) with a digital refractometer (PAL-1, Atago, Japan) and TA (%) by titration of 10 mL of juice with 0.1 N NaOH to a pH of 8.2. The epidermis color was determined i) at harvest, visually as color coverage (%) and ii) after storage, with a tristimulus colorimeter (CR-300, Minolta, Japan), on two well-colored areas on each fruit after removing the epicuticular wax. Data are expressed in coordinates L^* , a^* and b^* , used to calculate hue and chroma.

Flesh color and chilling injury symptoms

Flesh color and chilling injury (CI) development were assessed visually by cutting each fruit on half along the equatorial axis. A 4-grade visual scale according to the percentage of flesh colored or injured was used: Uninjured (0%), G₁ (up to 25%), G₂ (25-50%), G₃ (50-75%) and G₄ (75-100%). Intensity of coloration and severity of CI were calculated as the total number of fruit in each grade multiplied by the grade and divided by the total of injured fruit. Chilling injury was also expressed as percentage of affected fruits.

Statistical analysis

All data obtained from the trial were analysed using ANOVA, carried out with Infostat/Professional v.2006p.1. Treatments were compared for each storage and shelf life period and then were subjected to mean separation by Tuckey test (0.05). The least significant differences (LSD) between treatments were calculated.

Results

Fruit characteristics at harvest

The fruit were harvested at the optimum maturity, according to local recommended firmness values. 'Royal Zee' was firmer (53,4 N) than the other cultivars which are recommended to avoid over-ripeness during storage. 'Linda Rosa' and 'Friar' were harvested at middle firmness values (41-45 N) while 'Angeleno' was the less firm cultivar at harvest (34,8 N). For all the cultivars, the greater the firmness at harvest, the higher acidity and the lower soluble solids and color coverage was observed (Table 1). Ethylene production at 20°C during ripening after harvest differed significantly among plum cultivars. 'Royal Zee' presented a rapid

increase in ethylene production, reaching a maximum of 198.5 $\mu\text{L kg}^{-1} \text{h}^{-1}$ after 9 days at 20°C. 'Linda Rosa' and 'Friar' did not reach a clear climacteric peak but presented a slow increase in ethylene production with maximum values of 21.2 $\mu\text{L kg}^{-1} \text{h}^{-1}$ and 17.3 $\mu\text{L kg}^{-1} \text{h}^{-1}$ after 17 and 20 days at 20°C, respectively. Finally, ethylene production of 'Angeleno' remained steady and under 1 $\mu\text{L kg}^{-1} \text{h}^{-1}$ throughout the study period (Fig. 1).

Effect of cold storage and 1-MCP on ethylene production

The 'Royal Zee' plum fruit presented a rapid increase in ethylene production when removed from the cold storage chamber, but at lower rates and in fewer days than after harvest (Fig. 2). 1-MCP treatment significantly delayed the increase of ethylene production after 30 days of storage and reduced the maximum of the climacteric after 30 and 50 days of storage.

After storage at 0°C, 'Linda Rosa' and 'Friar' plums presented an increase in ethylene production capacity compared to the values observed at harvest, and control

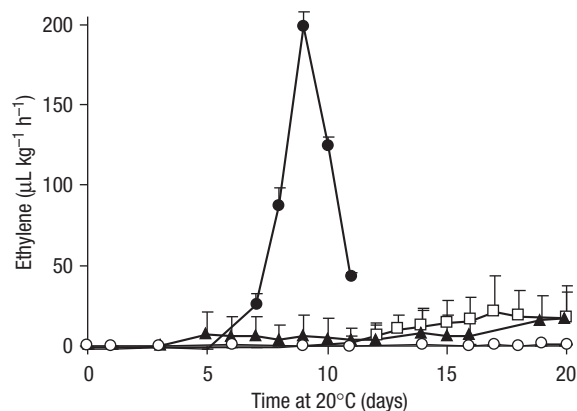


Figure 1. Evolution of ethylene production right after harvest in 'Royal Zee' (●), 'Linda Rosa' (□), 'Friar' (▲) and 'Angeleno' (○) plums. Vertical bars represents the standard deviation, for three samples of 6 fruit each.

Table 1. Maturity indexes at harvest of 'Royal Zee', 'Friar', 'Linda Rosa' and 'Angeleno' plums

Indexes	'Royal Zee'	'Linda Rosa'	'Friar'	'Angeleno'
Weight (g)	64.73 ± 2.95	98.58 ± 5.70	90.01 ± 1.26	87.34 ± 4.19
Firmness (N)	53.46 ± 4.17	41.26 ± 0.36	45.08 ± 2.30	34.84 ± 1.21
Soluble solids content (%)	11.17 ± 0.51	14.27 ± 0.71	12.03 ± 0.25	15.20 ± 0.20
Titrateable acidity (%)	2.23 ± 0.03	1.63 ± 0.10	1.61 ± 0.14	1.16 ± 0.07
Color coverage (%)	18.35 ± 2.74	82.92 ± 7.14	95.00 ± 3.25	100 ± 0.00

Each value represents the average ± standard deviation, for three samples of 20 fruit each.

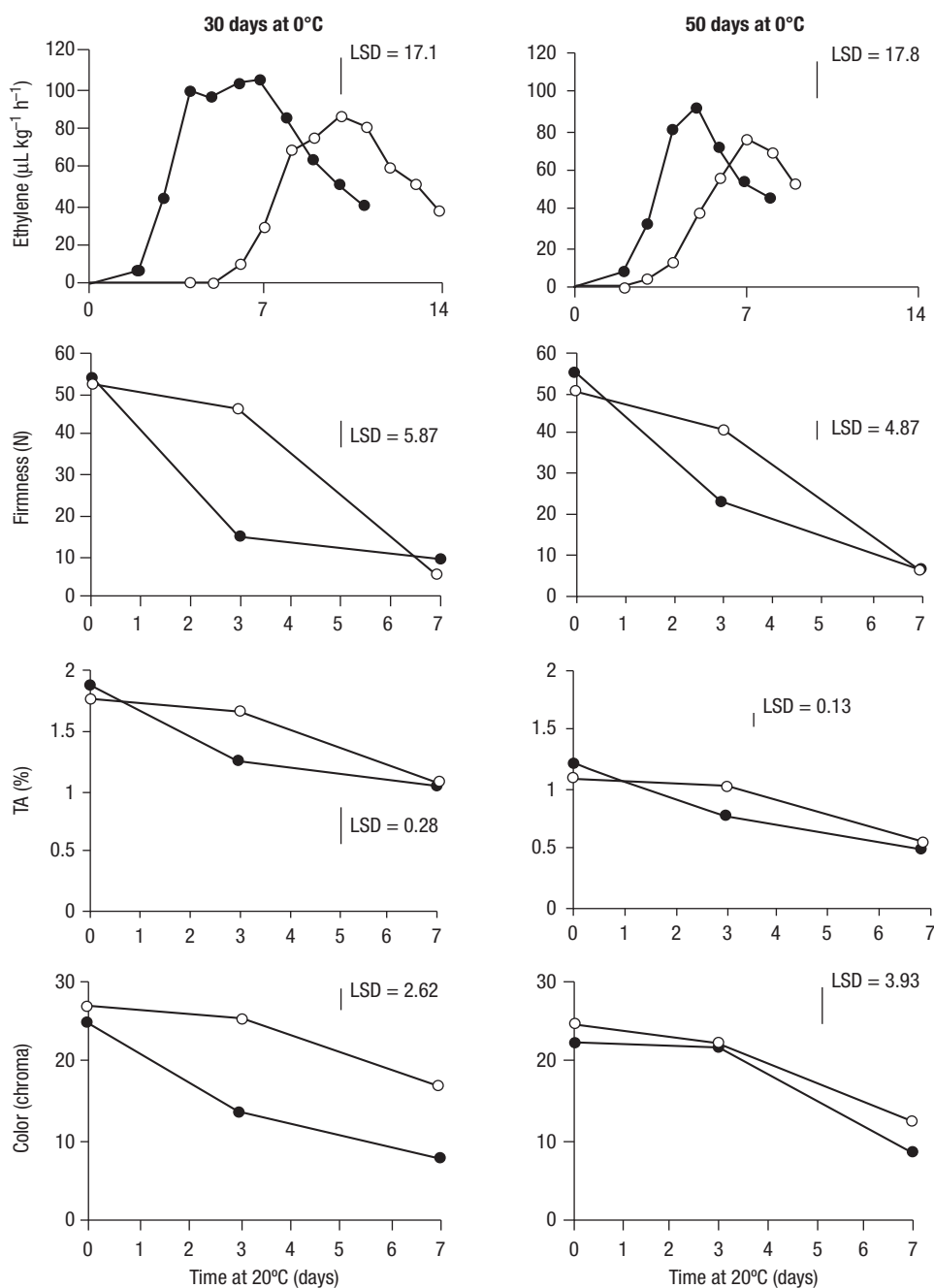


Figure 2. Evolution of ethylene production, firmness, titratable acidity and epidermal color during shelf life at 20°C, after 30 and 50 days of storage at 0°C in control (●) and 1-MCP-treated (○) 'Royal Zee' plums. Vertical bars show LSD (Tukey, 0.05) of the set of points presented in each graph.

fruit exhibited a clear climacteric peak during shelf life at 20°C. 1-MCP treatment delayed the occurrence of the climacteric peak in the 'Linda Rosa' cultivar both after 30 and 50 days of storage at 0°C (Fig. 3). A similar behavior was found for the 'Friar' cultivar with an inhibition of climacteric ethylene production after

30 days of storage (Fig. 4). Low-temperature storage did not affect the ethylene production rates in 'Angeleno' cultivar, which remained lower than $3 \mu\text{L kg}^{-1} \text{h}^{-1}$ throughout the entire study period and without any differences between control fruit and 1-MCP-treated fruit (Fig. 5).

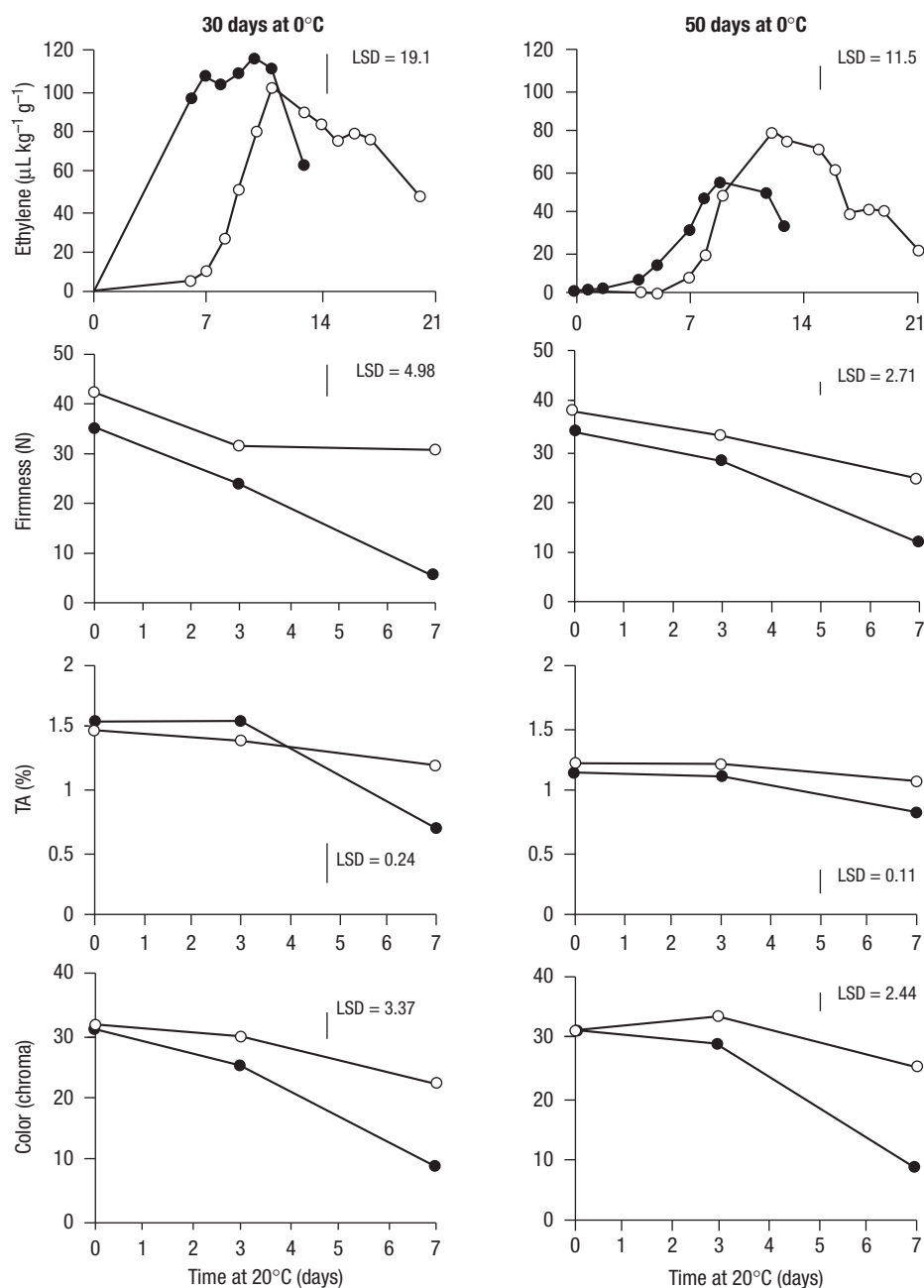


Figure 3. Evolution of ethylene production, firmness, titratable acidity and epidermis color during shelf life at 20°C, after 30 and 50 days of storage at 0°C in control (●) and 1-MCP-treated (○) 'Linda Rosa' plums. Vertical bars show LSD (Tukey, 0.05) of the set of points presented in each graph.

Effect of cold storage and 1-MCP on fruit ripening

As for ethylene production, the evolution of maturity indexes during shelf life was cultivar dependent. 'Royal Zee' control fruit softened rapidly while 1-MCP-treated fruit presented significantly higher firmness

values up to 3 days of shelf life but not after 7 days of shelf life at 20°C (Fig. 2). 'Linda Rosa' and 'Friar' control fruit rapidly lost firmness during shelf life, while 1-MCP-treated fruit softened more slowly and maintained significantly higher firmness values up to 7 days of shelf life, both after 30 and 50 days of cold storage (Figs. 3 and 4). The 'Angeleno' cultivar present-

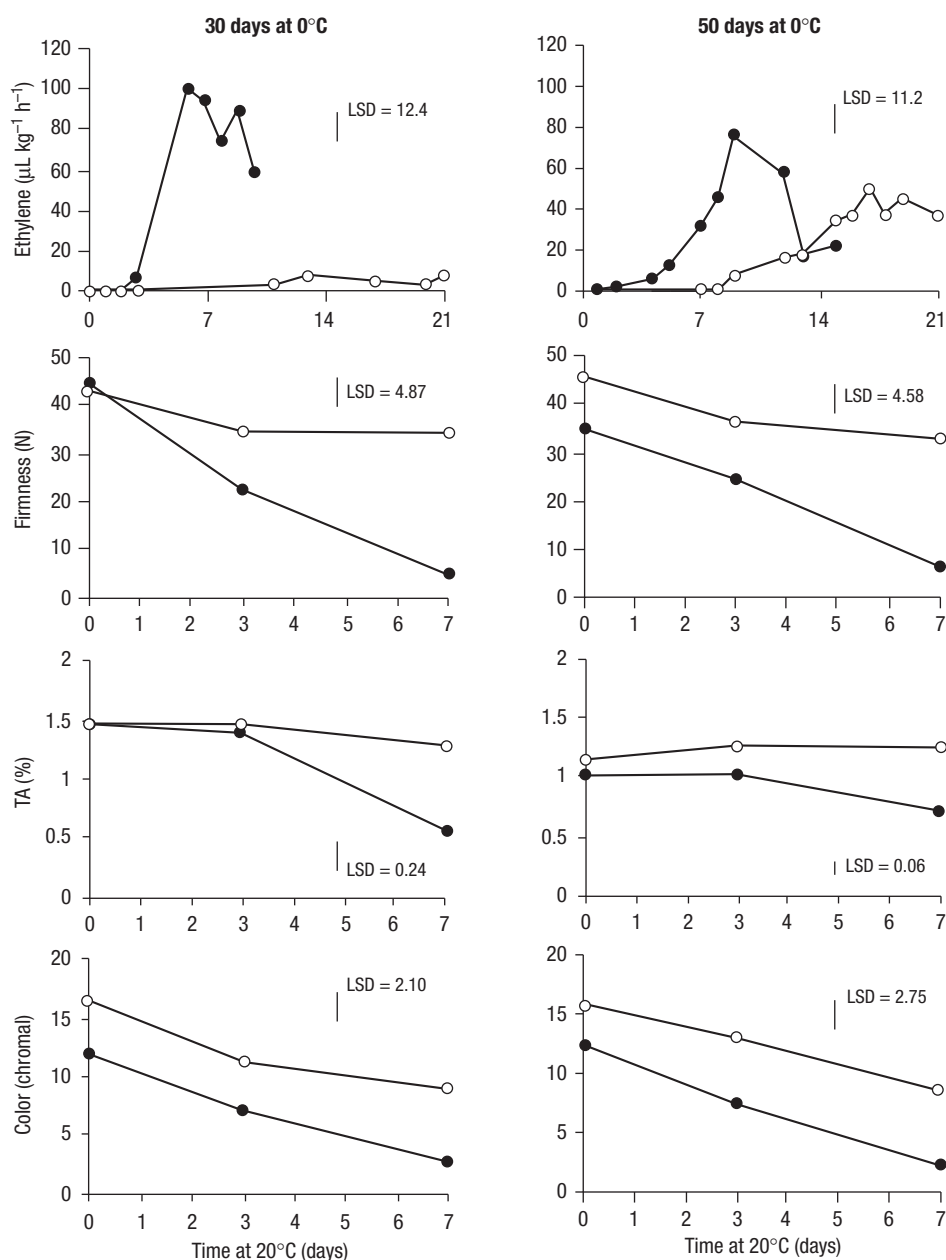


Figure 4. Evolution of ethylene production, firmness, titratable acidity and epidermis color during shelf life at 20°C, after 30 and 50 days of storage at 0°C in control (●) and 1-MCP-treated (○) 'Friar' plums. Vertical bars show LSD (Tukey, 0.05) of the set of points presented in each graph.

ted very slow softening during shelf life, without any significant differences observed between control fruit and 1-MCP-treated fruit (Fig. 5).

The fruit presented a decrease in TA during cold storage and subsequent shelf life, except for the 'Angeleno' cultivar. 1-MCP treatment delayed TA loss during shelf life and significant differences were found after 3 days of shelf life for 'Royal Zee' plums, and

after 7 days of shelf life for 'Linda Rosa' and 'Friar' plums (Figs. 2, 3 and 4). SSC (%) remained steady throughout storage and shelf life and was not affected by 1-MCP treatment in any of the cultivars (data not shown).

The 'Angeleno' cultivar did not show significant epidermis color changes and no differences were observed between control fruit and 1-MCP-treated fruit

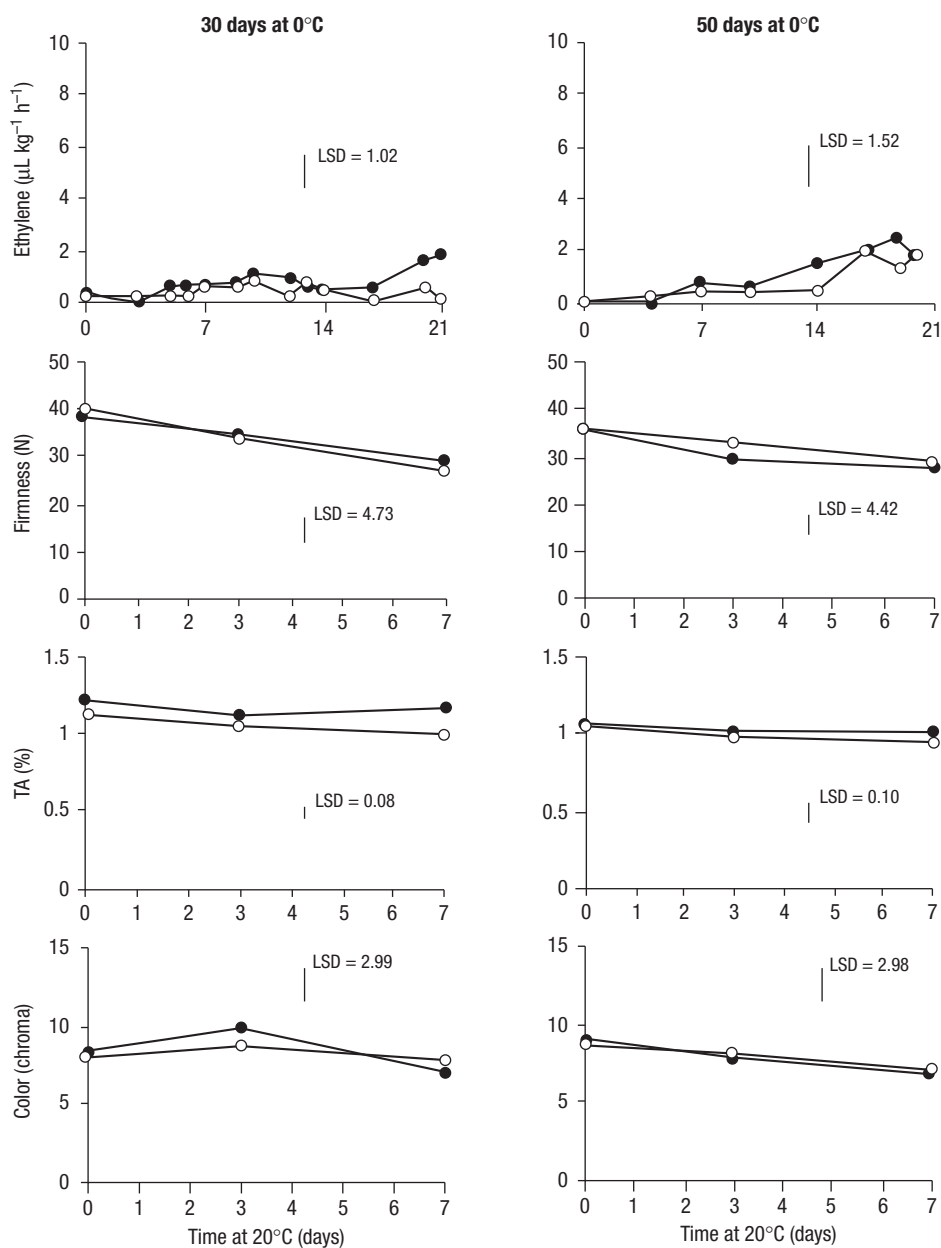


Figure 5. Evolution of ethylene production, firmness, titratable acidity and epidermis color during shelf life at 20°C, after 30 and 50 days of storage at 0°C in control (●) and 1-MCP-treated (○) 'Angeleno' plums. Vertical bars show LSD (Tukey, 0.05) of the set of points presented in each graph.

(Fig. 5). In contrast, the epidermis color changed from red to purple in 'Royal Zee' and 'Linda Rosa' plums, and from purple to black in 'Friar'. These color changes during storage of plums were reflected by the chroma better than by hue (data not shown). As visual color changed, significant decrease in chroma values was observed during shelf life. 1-MCP-treated fruit maintained higher chroma values during shelf life after 30

days of storage in 'Royal Zee' plums and after 30 and 50 days of storage in 'Linda Rosa' and 'Friar' plums (Figs. 2, 3, 4).

As observed for epidermis color, all cultivars except 'Angeleno' developed red flesh color (Table 2). The effect of 1-MCP on these characteristics was lower in 'Royal Zee' than in 'Linda Rosa' plums, and in the latter than in 'Friar' plums, in which 1-MCP totally inhibited

Table 2. Effect of 1-MCP treatment on red flesh color development (intensity: 1 to 4) in different plum cultivars stored 30 and 50 days at 0°C and exposed to 0, 3 and 7 days of shelf life at 20°C

	30 days at 0°C			50 days at 0°C		
	0	3	7	0	3	7
<i>Royal Zee</i>						
Control	1.17 ^a	1.32 ^a	1.74 ^b	1.19 ^a	1.24 ^a	1.62 ^a
1-MCP	1.00 ^a	1.38 ^a	1.47 ^a	1.22 ^a	1.10 ^a	1.45 ^a
<i>Linda Rosa</i>						
Control	1.23 ^a	1.31 ^b	3.83 ^b	1.22 ^a	1.43 ^a	2.03 ^b
1-MCP	1.00 ^a	1.00 ^a	1.33 ^a	1.17 ^a	1.19 ^a	1.35 ^a
<i>Friar</i>						
Control	0 ^a	1.50 ^b	2.21 ^b	1.07 ^b	2.20 ^b	3.03 ^b
1-MCP	0 ^a	0.00 ^a	0.00 ^a	0.00 ^a	0.33 ^a	0.67 ^a

Mean separation was carried out using Tukey's test (0.05).

the development of red flesh color up to 50 days of storage at 0°C.

Effect of cold storage and 1-MCP on chilling injury (CI)

Flesh translucency was the most important CI symptom observed in this work. In 'Royal Zee' plums, CI symptom was present after 30 days of storage at 0°C both in control fruit and in 1-MCP-treated fruit, and increased as the storage was extended and after 3 days of shelf life irrespective of the treatment (Fig. 6a). Flesh translucency affected 'Linda Rosa' and 'Friar' control fruits during shelf life especially after 50 days of storage at 0°C (Figs. 6b and 6c). 1-MCP treatment completely inhibited CI occurrence in both cultivars after 30 days of storage and significantly reduced the percentage of injured fruit even after 7 days of shelf life. No translucency symptoms were observed in the 'Angeleno' plum fruit in all the tests performed even without 1-MCP treatment.

The severity of translucency symptoms was 1.5 and 2 in 'Royal Zee', 0.5 and 2 in 'Linda Rosa' and 1 and 2 in 'Friar' plums at the end of shelf life and following 30 and 50 days of storage, respectively. As observed for percentage of injured fruit, 1-MCP treatment reduced the symptom's severity in 'Linda Rosa' and 'Friar' plums but not in 'Royal Zee' plums (data not shown).

Discussion

Ripening pattern and response to 1-MCP

Plums are traditionally considered as climacteric fruit, but in the last few years suppressed climacteric cultivars have been identified (Abdi *et al.*, 1997; Zuzunaga *et al.*, 2001). According to this work 'Royal Zee' plums are considered climacteric and show a rapid increase in ethylene production during ripening (Dong *et al.*, 2002). In contrast, 'Angeleno' plums maintained a very low ethylene production throughout the post-harvest period, which is in agreement with the observations of Holcroft *et al.* (2002), who define this cultivar as suppressed climacteric. 'Linda Rosa' and 'Friar' cultivar fruit presented low ethylene production rate after harvest, but a typically climacteric ripening pattern was observed after 30 and 50 days of storage at 0°C. These results suggest that these cultivars need low temperatures to start their climacteric ethylene synthesis, as observed in 'Songold' and 'Larry Ann' cultivars (Taylor *et al.*, 1993; Candan *et al.*, 2008). Such behavior has also been observed in other species like nectarines (Brecht and Kader, 1984), apples (Larrigaudière *et al.*, 1993, 1997) and pears (Lelièvre *et al.*, 1997).

The 1-MCP treatment reduced ethylene production and this effect was less significant in 'Royal Zee' than in 'Linda Rosa' plums and than in 'Friar' plums. This suggests that, among climacteric cultivars, the higher ethylene production rate at harvest, the less

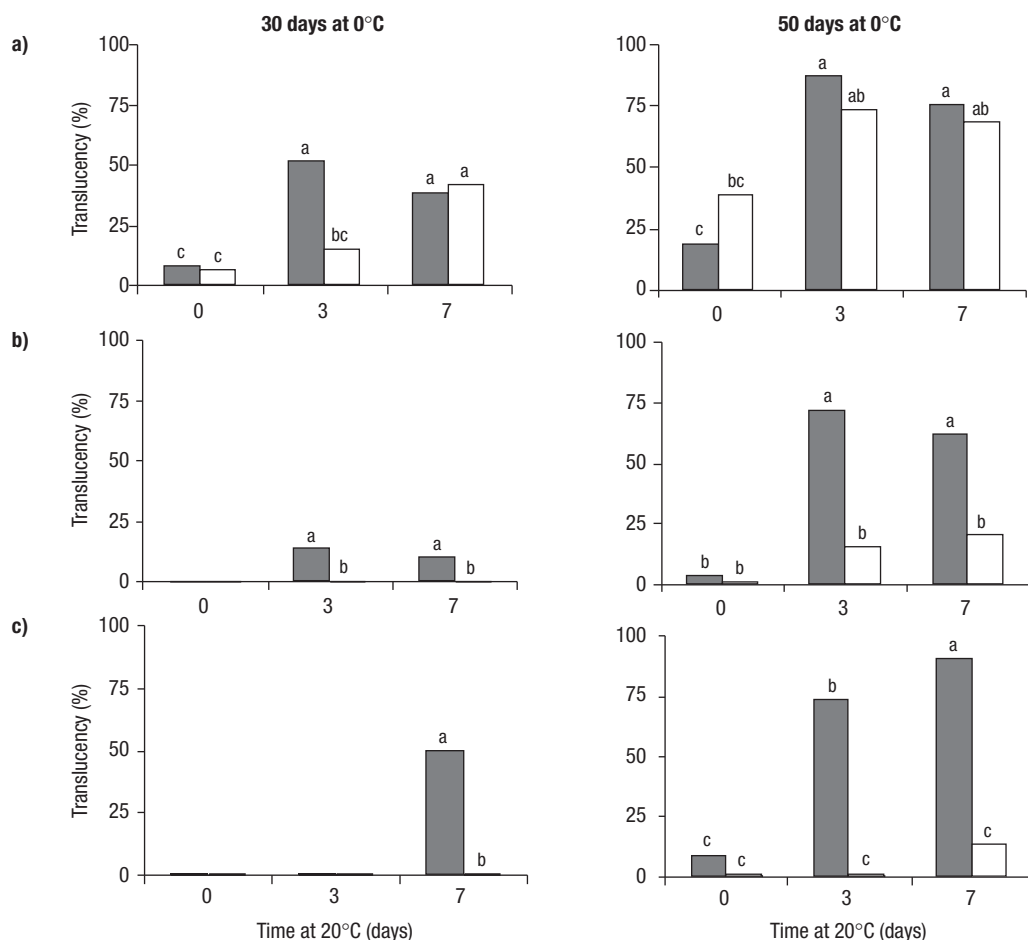


Figure 6. Percentage of fruit affected by flesh translucency during shelf life at 20°C after 30 and 50 days of storage at 0°C in control fruit (grey bars) and 1-MCP-treated fruit (white bars) of 'Royal Zee' (a), 'Linda Rosa' (b) and 'Friar' (c) plums. Within each graph, columns with different letters differ significantly according to the LSD value (Tukey, 0.05).

significant is the response to 1-MCP treatment. This result was in accordance to those observed by Valero *et al.* (2005) and showed that the 1-MCP dose required to control ripening in plum should be adjusted to the cultivar (Martínez-Romero *et al.*, 2003). In the case of the suppressed climacteric 'Angeleno' cultivar, 1-MCP treatment affected neither ethylene production nor maturity indexes (such as firmness, TA and epidermis color). Such a relationship has been observed in other cultivar and it was hypothesized that ripening changes were not related to ethylene production in suppressed climacteric cultivars (Abdi *et al.*, 1997). However, this result must be interpreted with caution, as other authors have observed that 1-MCP treatment reduces softening rates in these types of cultivars (Valero *et al.*, 2005; Menniti *et al.*, 2006).

Effects of 1-MCP treatment on fruit quality

In general it is recognized that 1-MCP treatment reduced firmness loss in plum (Dong *et al.*, 2002; Argenta *et al.*, 2003; Martínez-Romero *et al.*, 2003; Salvador *et al.*, 2003; Valero *et al.*, 2005; Candan *et al.*, 2006). In accordance with this idea, low ethylene production in 'Angeleno' fruit coincides with lower softening rate than that observed in the other cultivars studied in this work. This result was observed by other authors (Holcroft *et al.*, 2002) and showed that flesh softening is an ethylene-dependent process (Lelièvre *et al.*, 1997). Softening is known to be associated with the fruit's cell wall degradation process, and it has recently been demonstrated that 1-MCP reduces cell wall-degrading enzymes' activity, helping to maintain the plums' firmness (Khan and Singh, 2007).

'Royal Zee', 'Linda Rosa' and 'Friar' plums lost TA as the fruit's storage and shelf life extended, due to the utilization of organic acids as substrate in tissue respiration. 1-MCP-treated fruit maintained higher TA values, which could be due to a decrease in respiration rates, as observed in other cultivars (Dong *et al.*, 2002; Argenta *et al.*, 2003; Salvador *et al.*, 2003).

On the other hand, many previous studies showed that 1-MCP treatment delays epidermis color changes in plums (Dong *et al.*, 2002; Salvador *et al.*, 2003) and that such effect is cultivar-dependent (Valero *et al.*, 2005). This work showed 1-MCP treatment delayed the epidermis color change (chroma) in climacteric cultivars but not affected the color in the suppressed climacteric cultivar. Besides, 1-MCP treatment significantly reduced red flesh color development in 'Friar' and 'Linda Rosa' plums, coinciding with observations made in 'Laetitia' (Argenta *et al.*, 2003) and 'Blackamber' (Candan *et al.*, 2006) plums. Nonetheless, Dong *et al.* (2002) observed that the application of 1-MCP increased the development of red pigments in the flesh of 'Royal Zee' plums, but such behavior was not observed in this work.

According to firmness values recommended for plum consumption (Neri, 2003), the time needed to ripen varied between 3 and 5 days for 'Royal Zee', and between 5 and 6 days for 'Linda Rosa' and 'Friar'. 1-MCP treatment extended the fruit's shelf life by 4 to 7 days in 'Royal Zee' and more than 7 days in 'Linda Rosa' and 'Friar' similarly than for 'Blackamber' plums (Candan *et al.*, 2006). Besides texture, the plums' organoleptic quality is also closely related to sugar (SSC) and acid (TA) contents (Crisosto *et al.*, 2004). This and other research papers state that 1-MCP treatment does not affect SSC but maintains higher TA values, which might affect the fruit's final sensory quality (Dong *et al.*, 2002; Argenta *et al.*, 2003; Salvador *et al.*, 2003; Candan *et al.*, 2006). When 'Royal Zee' fruits reached their firmness consumption, the values of AT were similar in both controls and in treated fruits. Moreover, these values are consistent with those recommended by Crisosto *et al.* (2007) for the consumption of this variety. The same may be expected to occur with the other cultivars studied, but further studies are necessary to affirm it.

Effect of 1-MCP treatment on chilling injury

According to the findings presented in this work, the percentage of fruit affected by translucency was

higher in cultivars that exhibited higher ethylene production rate. In accordance with this idea, the 'Angeleno' cultivar fruit that produced low ethylene throughout the entire experimental period did not develop CI symptoms. These results show the relationship that exist between ethylene production and fruit susceptibility to CI and sustains the hypothesis proposed in previous research (Candan *et al.*, 2008). Moreover, 1-MCP treatment significantly reduced the appearance of translucency symptoms and even completely eliminated these symptoms in 'Linda Rosa' and 'Friar' cultivars after 30 days of storage. Similar results were also observed in 'Blackamber' (Candan and Calvo, 2005), 'Larry Ann' (Candan *et al.*, 2008) and 'Fortune' (Menniti *et al.*, 2006) plums.

Collectively, these results support the idea that ethylene participates in the development of chilling injury symptoms in climacteric plums (Candan *et al.*, 2008) and that 1-MCP is a useful tool for chilling injury control in this species. They also suggest that early ripening cultivars might be more susceptible to chilling injury than late ripening cultivars.

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