



## Could the 1-MCP treatment effectiveness in plum be affected by packaging?

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### Abstract

Plum fruit (*Prunus domestica* L., cv. 'President') were harvested at a pre-climacteric stage of ripeness. They were packaged in small cardboard boxes or in bulk, and treated with 1-methylcyclopropene (1-MCP) at 0.3 or 0.5  $\mu\text{l l}^{-1}$  for 24 h. Then fruit were stored at 1 °C during 0–5 weeks, and weekly removed for 7 days at 20 °C for a shelf-life period. All 1-MCP treatments inhibited the typical ethylene climacteric peak and delayed the change in properties related to fruit ripening, such as fruit softening, decrease in titratable acidity and colour chroma index, and increase in soluble solids content. Nevertheless, these effects were significantly higher when 1-MCP application was performed in plums packaged in small and ventilated cardboard boxes than in bulk, and the storability was extended for up to 5 weeks of cold storage plus 7 days at 20 °C. Thus, for commercial purposes, and in order to obtain the maximum benefit of the 1-MCP, treatments should be carried out in plums already packaged, since gas could diffuse homogeneously to the fruit surface, and all ethylene receptors would be blocked by 1-MCP.

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### 1. Introduction

The use of 1-methylcyclopropene (1-MCP) has been evaluated on different vegetables and fruit, both climacteric and non-climacteric. The 1-MCP treatments induce beneficial effects, such as delay of the physico-chemical changes related to the ripening process, as well as a reduction of decay, weight loss, and chilling injury (Blankenship and Dole, 2003). The main mode of action of 1-MCP on fruit has

been related to ethylene action, since 1-MCP blocks the ethylene receptors and inhibits its hormonal action (Sisler and Serek, 1997; Watkins, 2002). Thus, 1-MCP is considered an effective tool for commercial application, in order to maintain quality and extend shelf-life of fruit and vegetables.

The application of 1-MCP in species of the genus *Prunus* (apricot, nectarine, peach, plum and sweet cherry) has been performed at different temperatures, concentrations, duration of treatments and ripening stages at harvest, and there is not a general rule to achieve the maximum effectiveness of 1-MCP treatment. Thus, the range of temperature for application is wide (between 1 and 20 °C) and the relationship

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between temperature and effectiveness of 1-MCP is not clear (Blankenship and Dole, 2003), although the efficacy has been reported to be higher at low temperature in different plum cultivars (Martínez-Romero et al., 2003; Salvador et al., 2003; Valero et al., 2003). The concentration of 1-MCP and duration of treatment also varies widely, from 20 to 40  $\text{nl l}^{-1}$  and between 4 and 24 h (Abdi et al., 1998; Dong et al., 2001a,b, 2002; Fan et al., 2000, 2002; Gong et al., 2002; Blankenship and Dole, 2003; Martínez-Romero et al., 2003; Salvador et al., 2003; Valero et al., 2003). It has been suggested that a fruit saturation response to 1-MCP occurs at between 0.5 and 1  $\mu\text{l l}^{-1}$  (Argenta et al., 2003). On the other hand, differences among cultivars and fruit maturity stage at harvest have been observed for plums (Martínez-Romero et al., 2003; Valero et al., 2003).

In most of the above-mentioned papers, 1-MCP treatments have been carried out with fruit in bulk inside hermetically sealed jars or containers of different capacity, ranging from 1 to 230 l. In other reports, 1-MCP treatment has been performed in combination with modified atmosphere packaging (injecting the 1-MCP into the plastic bags), such as in banana (Jiang et al., 1999) and apples (DeEll et al., 2002). However, there are no reports on the question of whether 1-MCP should be applied with fruit in bulk or after handling and packaging in small boxes, from the commercial application point of view. Thus, the objective of this work was to compare the effect of 1-MCP treatment (at two different doses, 0.3 and 0.5  $\mu\text{l l}^{-1}$ ) applied to 'President' plum, either packaged in cardboard boxes or in bulk, on the change in fruit quality parameters during postharvest storage. To evaluate 1-MCP effectiveness, the change in fruit firmness, weight loss, colour, soluble solids concentration and titratable acidity, as well as the ethylene production and respiration rates, after different periods of cold storage plus 7 days of shelf-life at 20 °C, were determined.

## 2. Materials and methods

### 2.1. Plant material

Plum fruit (*Prunus domestica* L., cv. President) were harvested in mid September 2002 from 10-year old trees grown in an orchard in Murcia (Spain).

Plums were picked at the commercial ripening stage (weight 74.82  $\pm$  0.24 g; total soluble solids (TSS) concentration 19.19  $\pm$  0.18%; titratable acidity 0.78  $\pm$  0.01 mmol  $\text{H}^+$   $\text{ml}^{-1}$ ; chroma index 13.59  $\pm$  0.85). Over 2000 fruit were manually picked, to avoid mechanical damage, and once in the laboratory 1560 fruit were selected according to size and colour uniformity. Sixty fruit were used to analyse the fruit physico-chemical characteristics at harvest (day 0), and the remaining plums were divided at random into 15 groups of 100 fruit. Six lots of 100 fruit were used for 1-MCP treatments in bulk with 0.3 and 0.5  $\mu\text{l l}^{-1}$  doses in triplicate. Treatments were performed in 0.12  $\text{m}^3$  hermetically sealed containers. The remaining nine lots of 100 fruit were split into groups of 20 fruit, packaged in cardboard boxes and used for 1-MCP treatments in triplicate for 0 (control), 0.3 and 0.5  $\mu\text{l l}^{-1}$  doses, placed in similar containers as the treatments in bulk (shown in Fig. 1).

Following 1-MCP application, treated plums in bulk were divided into lots of 20 plums and placed in cardboard boxes. All fruit were stored at 1  $\pm$  0.5 °C, in permanent darkness and with an RH of 90% for 5 weeks. Three lots of each treatment were weekly sampled and stored at 20 °C for 7 days (shelf-life period (SL)), in which the ethylene production, respiration rate, colour, firmness, soluble solids concentration and titratable acidity were determined.

### 2.2. 1-MCP treatments

Fruit were pre-cooled at 1 °C for 6 h before treatments. SmartFresh™ for 1-MCP (0.14%) was supplied by AgroFresh Inc. (Rohm and Haas Inc., Gessate, Italy) as a powder, that after addition of warm water (40 °C) released the active ingredient as gas. Different amounts of the powder were weighed and warm water was added to obtain triplicate doses of 0.3 and 0.5  $\mu\text{l l}^{-1}$ . Treatments were performed in 0.12  $\text{m}^3$  hermetically sealed containers through injection of 1-MCP using plastic syringes (Fig. 1). Duration of treatment was 24 h at 1 °C. Control fruit were similarly treated but without 1-MCP.

### 2.3. Ethylene and respiration rate determination

Ethylene and CO<sub>2</sub> production were measured by placing each lot and replication of 20 fruit in a 3 l

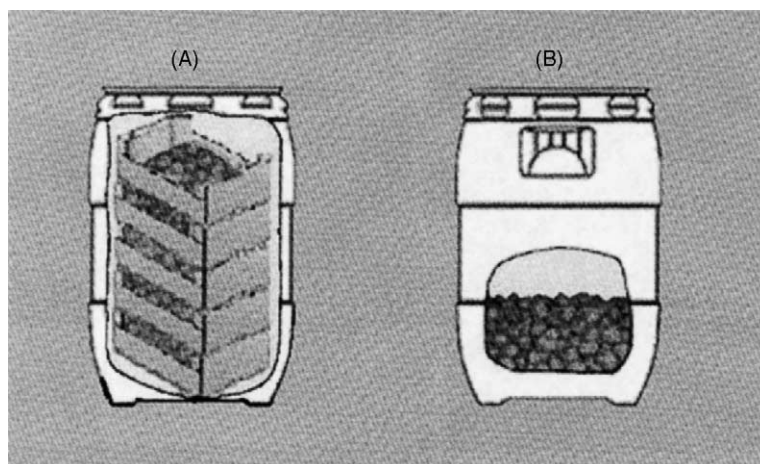


Fig. 1. Scheme for 1-MCP treatment of plums packaged in small cardboard boxes (A) or in bulk (B) in 1201 hermetically sealed containers.

glass jar hermetically sealed with a rubber stopper for 15 min. One millilitre of the holder atmosphere was withdrawn with a gas syringe, and the ethylene was quantified using a Hewlett-Packard™ model 5890A gas chromatograph (Wilmington, DE) equipped with a flame ionisation detector and a 3 m stainless steel column with an inner diameter of 3.5 mm containing activated alumina of 80/100 mesh. The column temperature was 90 °C, and injector and detector temperatures were 150 °C. Results were the mean  $\pm$  S.E. of four determinations for each one of the replicates used and expressed as  $\text{nmol g}^{-1} \text{h}^{-1}$ . For respiration rate determination, another sample of 1 ml of the same atmosphere was withdrawn and  $\text{CO}_2$  quantified using a Shimadzu™ 14A gas chromatograph (Kyoto, Japan), with a thermal conductivity detector (TCD) and a 3 m stainless steel column with an inner diameter of 3.3 mm containing Chromosorb 102. The column temperature was 55 °C, and injector and detector temperatures were 110 °C. Results were the mean  $\pm$  S.E. of four determinations for each one of the replicates used and expressed as  $\text{mmol kg}^{-1} \text{h}^{-1}$ .

#### 2.4. Fruit quality parameters

Colour was determined using the Hunter Lab System in a Minolta colorimeter CR200 model (Minolta Camera Co., Japan). Results were the mean  $\pm$  S.E. of

two determinations for each fruit of the three replicates and expressed as  $C^*$  (chroma index). For pulp firmness determination, 1  $\text{cm}^2$  of the skin was removed and penetration force measurements were individually recorded using a 5 mm diameter probe coupled on a TX-XT2i Texture Analyzer (Stable Microsystems, UK) interfaced to a personal computer. A bevelled holder prevented bruising of the opposite side. Penetration rate was 20  $\text{mm min}^{-1}$  for 10 mm after contacting the flesh, and results were the mean  $\pm$  S.E. of determination in each fruit and expressed in N. TSS concentrations were determined in each fruit with a digital refractometer Atago PR-101 (Atago Co. Ltd., Japan) at 20 °C, and results were the mean  $\pm$  S.E. expressed as a percentage. Fruit acidity was determined in each fruit by measurement of pH using 1 ml of diluted juice in 25 ml distilled  $\text{H}_2\text{O}$  and results were the mean  $\pm$  S.E. expressed as  $\text{mmol H}^+ \text{ml}^{-1}$ .

#### 2.5. Statistical analysis

Data were subjected to ANOVA analysis. Sources of variation were type of packaging system, storage duration and treatment. Mean comparisons were performed using HSD Tukey's test to examine if differences between packaging system, treatments and storage duration were significant at  $P < 0.05$  using SPSS software (SPSS, 2001).

### 3. Results

#### 3.1. Ethylene production and respiration rate

Ethylene production significantly increased in control fruit reaching the climacteric peak ( $0.25 \pm 0.01 \text{ nmol g}^{-1} \text{ h}^{-1}$ ) after 2 weeks of cold storage plus shelf-life, and then progressively decreased until the end of the experiment (Fig. 2). 1-MCP treatments were effective in inhibiting the ethylene production, although differences were observed depending on the method used for its application. Thus, ethylene production was very low (between 0.01 and  $0.02 \text{ nmol g}^{-1} \text{ h}^{-1}$ ) and remained almost unchanged until the end of the experiment in plums treated in packages, while for those fruit treated in bulk, a significant increase was detected from the third and fourth week for 0.3 and  $0.5 \mu\text{l l}^{-1}$  1-MCP dose, respectively. For this treatment, ethylene production was negatively correlated with the applied dose ( $r^2 = 0.86$ ).

Respiration rate at harvest was  $0.20 \pm 0.01 \text{ mmol CO}_2 \text{ kg}^{-1} \text{ h}^{-1}$  and progressively increased during storage, the levels being significantly higher in control than in 1-MCP treated plums, for both methods of 1-MCP treatment (Fig. 3). However, plums packaged

before performing the treatment exhibited significantly lower respiration rates than those treated in bulk for both 1-MCP doses. In addition, for packaged and bulked plums the effect of 1-MCP on reducing the respiration rate was dose-dependent ( $r^2 = 0.96$  and  $0.97$ , respectively),  $0.5 \mu\text{l l}^{-1}$  being the most effective.

The differences between packaged plums and those treated in bulk in these parameters were not due to occurrence of mechanical damage, since the load supported by fruit from the bottom was not high enough to induce fruit bruising, as was visually observed after removing the fruit.

#### 3.2. Colour changes

Colour, expressed as  $C^*$  (Fig. 4) resulted in a sharp diminution from the initial value ( $13.59 \pm 0.85$ ) within the first 2 weeks of cold storage plus shelf-life, showing a continuous decrease until the end of the storage period. On the contrary, colour changes were significantly delayed by 1-MCP treatments, especially for packaged plums, in which the chroma index remained unchanged after 3 weeks of cold storage plus shelf-life. In addition, values

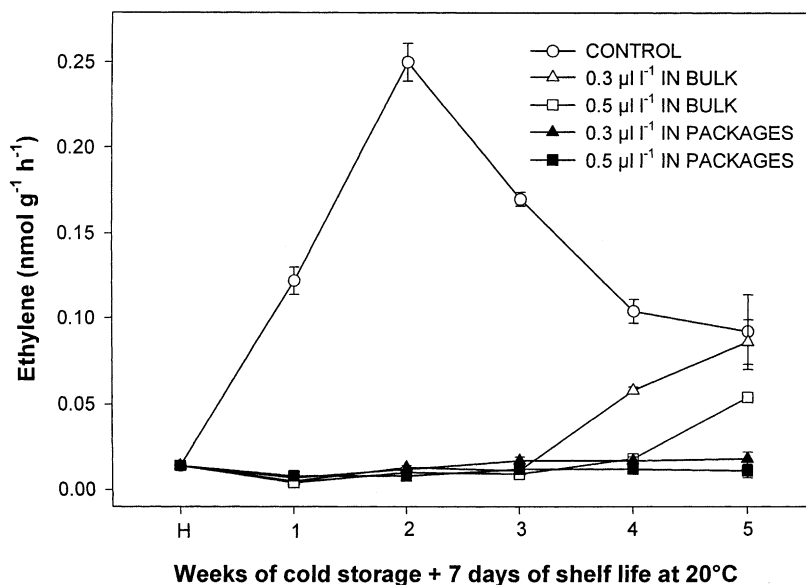


Fig. 2. Ethylene production at harvest (H) and after 1–5 weeks of cold storage at  $1^\circ\text{C}$  followed by 7 days at  $20^\circ\text{C}$  in plums treated with 0 (control, circles),  $0.3 \mu\text{l l}^{-1}$  (triangles) and  $0.5 \mu\text{l l}^{-1}$  of 1-MCP (squares) applied to packages (closed symbols) or in bulk (open symbols). Data are the mean  $\pm$  S.E. ( $n = 12$ ).

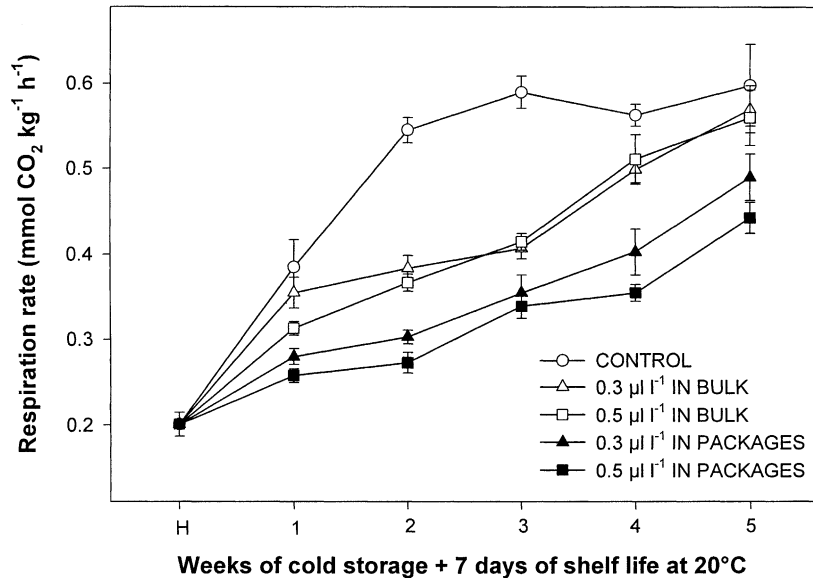


Fig. 3. Respiration rate at harvest (H) and after 1–5 weeks of cold storage followed by 7 days at 20 °C in plums treated with 0 (control, circles), 0.3 µl l<sup>-1</sup> (triangles) and 0.5 µl l<sup>-1</sup> of 1-MCP (squares) applied to packages (closed symbols) or in bulk (open symbols). Data are the mean ± S.E. (*n* = 12).

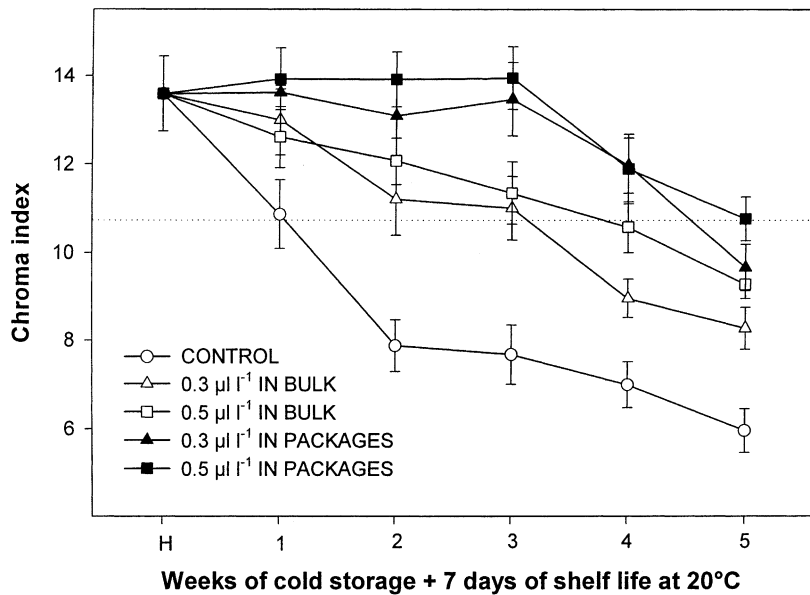


Fig. 4. Colour (chroma index) at harvest (H) and after 1–5 weeks of cold storage followed by 7 days at 20 °C in plums treated with 0 (control, circles), 0.3 µl l<sup>-1</sup> (triangles) and 0.5 µl l<sup>-1</sup> of 1-MCP (squares) applied to packages (closed symbols) or in bulk (open symbols). Data are the mean ± S.E. (*n* = 120).

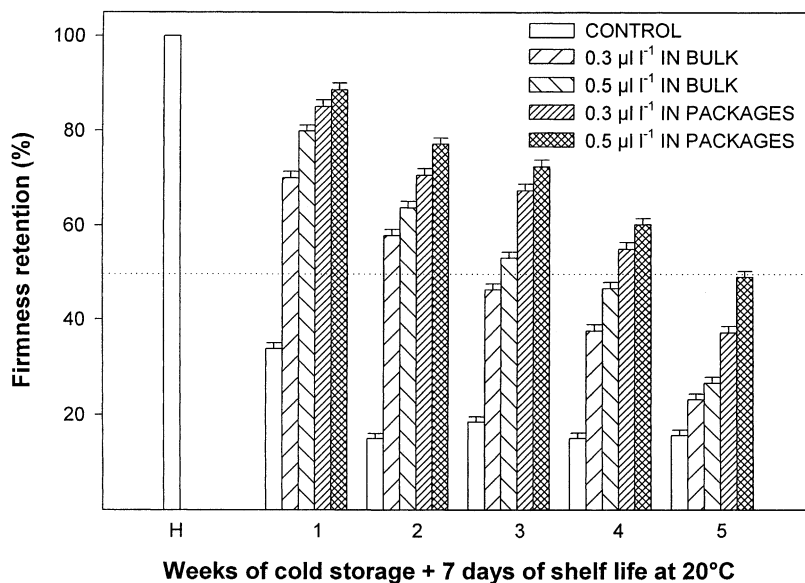


Fig. 5. Percentage of fruit firmness retention at harvest (H) and after 1–5 weeks of cold storage followed by 7 days at 20 °C in plums treated with 0 (control, open bars), 0.3 and 0.5  $\mu\text{l l}^{-1}$  of 1-MCP applied to packages (grid) or in bulk (fine pattern). Data are the mean  $\pm$  S.E. ( $n = 60$ ).

of chroma index were significantly higher in packaged fruit than those observed for plums treated in bulk along the experiment, for both 1-MCP doses. It was found that maintenance of colour was positively correlated with the 1-MCP dose with correlation coefficients of 0.90 and 0.83, for packaged and bulked plums, respectively.

### 3.3. Fruit firmness

Fruit firmness at harvest was  $8.99 \pm 0.42$  N and diminished during the postharvest storage, the decrease being significantly higher in control than in 1-MCP treated fruit. Fig. 5 shows the percentage of fruit firmness retention with respect to the levels at harvest. For control plums, after 1 week of cold storage plus shelf-life, fruit firmness was 34% of the initial level. The greater effectiveness in fruit firmness retention was achieved when 1-MCP treatment was performed on packaged plums, in which after 4 weeks of cold storage plus shelf-life, 60 and 55% of fruit firmness was retained for the 0.5 and 0.3  $\mu\text{l l}^{-1}$  1-MCP doses, respectively, while these values were 47 and 36% in plums treated in bulk. Finally, it was found that soften-

ing process was influenced by 1-MCP dose, since the higher 1-MCP dose applied led to higher fruit firmness retention, with correlation coefficients of 0.99 and 0.94 for plums treated in bulk and in packages, respectively.

### 3.4. Soluble solids concentration and acidity

The TSS concentration significantly increased during the first 2 weeks of cold storage plus shelf-life in control fruit, from levels at harvest of  $19.19 \pm 0.18$  to  $23.10 \pm 0.16\%$ , remaining unchanged until the end of the experiment (Fig. 6). However, this increase in TSS was significantly delayed in 1-MCP treated plums, with no significant differences between the two procedures of 1-MCP application or the applied dose. On the contrary, the change in fruit acidity was affected by the way of performing the treatment (Fig. 7). Thus, plums treated in packages exhibited a significantly lower reduction in acidity compared with controls or plums treated in bulk, which was found to be dose-dependent ( $r^2 = 0.98$ ). Finally, no significant differences were found between controls and fruit treated in bulk for any of the 1-MCP doses applied.

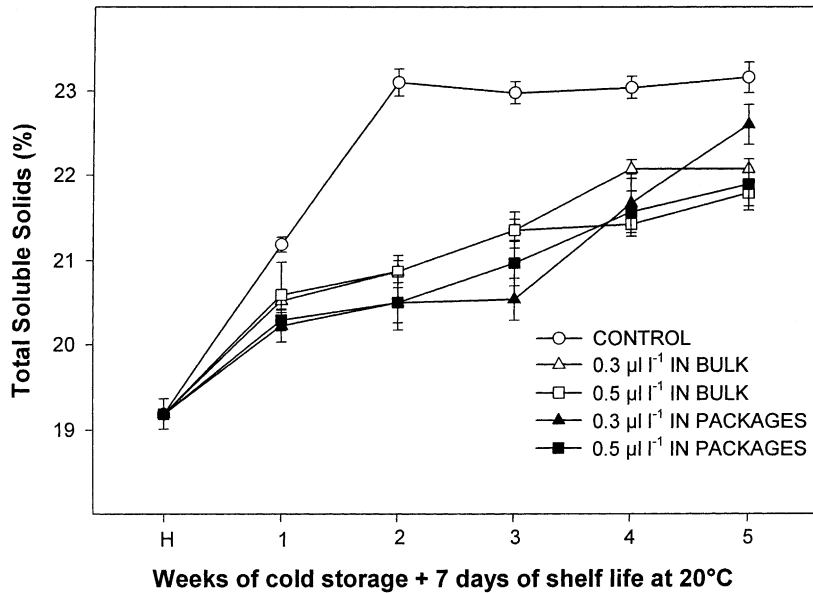


Fig. 6. Total soluble solids concentration at harvest (H) and after 1–5 weeks of cold storage followed by 7 days at 20°C in plums treated with 0 (control, circles), 0.3 μl l<sup>-1</sup> (triangles) and 0.5 μl l<sup>-1</sup> of 1-MCP (squares) applied to packages (closed symbols) or in bulk (open symbols). Data are the mean ± S.E. (*n* = 60).

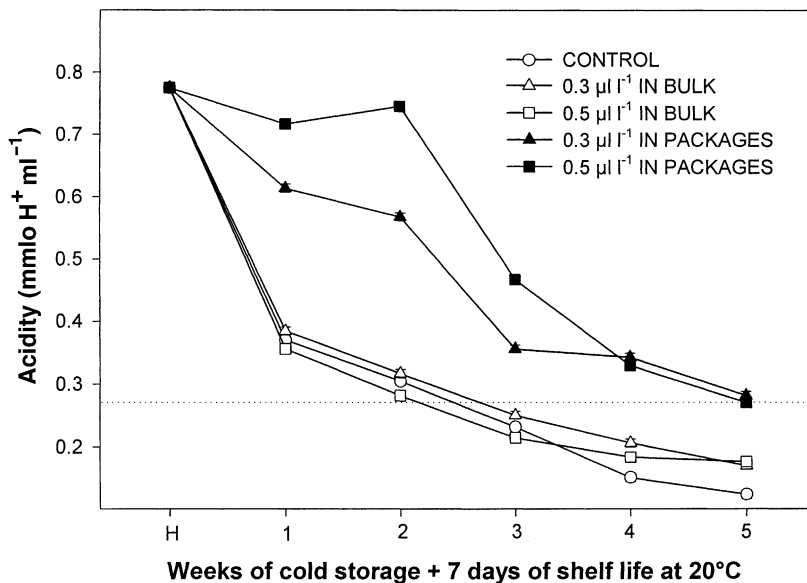


Fig. 7. Fruit acidity (mmol H<sup>+</sup> ml<sup>-1</sup>) at harvest (H) and after 1–5 weeks of cold storage followed by 7 days at 20°C in plums treated with 0 (control, circles), 0.3 μl l<sup>-1</sup> (triangles) and 0.5 μl l<sup>-1</sup> of 1-MCP (squares) applied to packages (closed symbols) or in bulk (open symbols). Data are the mean ± S.E. (*n* = 60).

#### 4. Discussion

Plums, as well as other stonefruit, are characterised by rises in ethylene production until reaching the climacteric peak, and then triggering the ripening process, with an acceleration of colour changes, softening and modification in the balance between total soluble solids and acidity (Zuzunaga et al., 2001). Thus, producers usually pick fruit at an early stage of ripeness, with enough firmness to withstand the handling and packaging process, although these fruit do not reach optimal organoleptic properties before consumption. Since cold temperature storage is not enough to inhibit ethylene production and the associated ripening process, there is a need to have new tools able to inhibit and/or delay the onset of ethylene production. Among these tools, the use of 1-MCP has been widely proved to be effective in extending shelf-life and improving quality in a large number of fruit, the main effects having been recently reviewed (Blankenship and Dole, 2003). However, there are different aspects that have not been decided yet, such as the best time for application (immediately after harvesting or after fruit handling) or the influence of the package itself on 1-MCP efficacy (type and size of packages). In cold storage studies, packaging is essential to gain rapid and effective pre-refrigeration, and in turn to maintain fruit quality until marketing, especially for small and well-aerated packages, over a 5% of their volume (Thompson et al., 1998).

In the present work, control plums exhibited a true climacteric ethylene peak after 2 weeks of cold storage plus 7 days at 20 °C, which was accompanied by increases in the respiration rate. However, 1-MCP treatment differentially affected ethylene production and respiration rate depending on the type of application. Thus, when 1-MCP was applied to fruit handled and packaged in perforated cardboard boxes, ethylene production was totally inhibited during all storage periods, while in those plums treated with 1-MCP in bulk (before handling and packaging), ethylene production increased after 3 and 4 weeks of cold storage plus 7 days at 20 °C, for 0.3 and 0.5  $\mu\text{l l}^{-1}$  1-MCP doses, respectively. The increase in respiration rate was delayed and dose-dependent for the applied 1-MCP, the effect being higher in packaged than in bulk plums. These differences could be attributed to the higher gas diffusion around the fruit when they are packaged in

small-perforated boxes. Given that the mode of action of ethylene is through a blockage of the ethylene receptors (Sisler and Serek, 1997), our results suggest that all receptors present at the time of 1-MCP application were irreversibly blocked in plums packaged in small boxes, since individualisation of fruit previously packaged could be achieved, and gas diffusion would reach the entire fruit surface. On the contrary, for those plums treated in bulk not all receptors were bound by 1-MCP, primarily due to the high number of fruit, which obstructed 1-MCP access to the whole fruit surface, since the latter was partially covered by the neighbouring fruit. However, the synthesis of new receptors in plums treated in bulk, as has been reported to occur in several fruit, cannot be discarded (Blankenship and Dole, 2003), and might be also responsible for the increase of ethylene after 3 weeks of cold storage.

The parameters related to plum ripening, such as colour chroma, TSS, fruit acidity and softening, quickly changed in control plums, while in 1-MCP treated plums significant delays were observed, which were dose-dependent with the exception of TSS. This behaviour seems to be a general effect of 1-MCP treatments in delaying the fruit ripening process during postharvest storage, related to the inhibition of ethylene production, as has been observed in a wide range of studied fruit: papaya, apple, apricot, avocado and peach (Hofman et al., 2001; Watkins et al., 2000; Fan et al., 2000, 2002; Jeong et al., 2002), and in several plum cultivars such as ‘Santa Rosa’, ‘Red Rosa’, ‘Golden Japan’, ‘President’, ‘Royal Zee’, ‘Shiro’, ‘Rubyred’, ‘Gulfruby’ and ‘Beauty’ (Abdi et al., 1998; Salvador et al., 2003; Martínez-Romero et al., 2003; Dong et al., 2001a, 2002; Valero et al., 2003).

In addition, the response to 1-MCP treatment in the parameters related to fruit ripening differed depending on the mode of its application, as did the ethylene inhibition. The greater effectiveness was achieved in packaged plum, since maintenance of colour, and fruit firmness and acidity retention were detected as compared with in those plums treated in bulk. Given the importance of these parameters for consumer’s acceptability for fruit, the maximum storage period with optimum plum quality parameters was 1 week of cold storage plus 7 days of shelf-life at 20 °C for control plums. For plums treated in bulk, the storage period was extended up to 3 weeks plus shelf-life, while up

to 5 weeks plus shelf-life for those plums treated in packages. In the latter, fruit had retained 50% of their initial firmness at this time, while fruit acidity and colour chroma had diminished 20% only, for the highest 1-MCP dose. Similar values of these parameters were reached 2 weeks earlier in plums treated in bulk.

In conclusion, it should be noticed that effectiveness of 1-MCP treatments is dependent on the fact that fruit are packed or not at time of application. Thus, from a commercial application point of view, treatment with 1-MCP in small and well-aerated packages should be carried out, in order to gain more inhibition of ethylene production, and in turn to delay the ripening process and extend fruit storability.

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