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Postharvest methyl salicylate treatments delay ripening and maintain quality attributes and antioxidant compounds of 'Early Lory' sweet cherry



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1. Introduction

Spain is the second largest producer of sweet cherries (*Prunus avium* L.) in Europe and is the sixth largest producer in the world with more than 30,000 ha and producing about 105,000 t annually. A wide range of cultivars are grown actually, from early to late season harvesting. 'Early Lory' is one of the earliest cultivars having low chilling requirements and being harvested beginning of May (Nogueroles-Pérez, 2005).

Sweet cherries are a nutritionally dense food rich in anthocyanins, quercetin, hydroxycinnamates, potassium, fiber, vitamin C, carotenoids, and melatonin. These constituent nutrients and bioactive food components support the potential preventive health benefits of cherry intake in relation to cancer, cardiovascular disease, diabetes, inflammatory diseases, among others (McCune et al., 2011; Ballistreri et al., 2013). However, several factors such as degree of ripeness at harvest, postharvest storage conditions, and processing, each can significantly alter the amounts of nutrients and bioactive components (Serrano et al., 2009, 2011; Valero et al., 2011; Díaz-Mula et al., 2012). In addition, the sweet cherry quality

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ABSTRACT

The effect of postharvest application of methyl salicylate (MeSA) at two concentrations (0.1 and 1 mM) on quality attributes, bioactive compounds and antioxidant activity was studied in 'Early Lory' sweet cherry. MeSA treatments were effective in reducing respiration rate, weight loss, softening, total acidity losses and the increase in the ripening index during storage at 2 °C for 20 days as compared with non-treated control fruit. In addition, total phenolics, total anthocyanins and total antioxidant activity (TAA) in the hydrophilic extract (H-TAA) remained at higher concentrations at the end of storage in treated fruit. High correlations were found between H-TAA and phenolics or anthocyanin concentrations and between lipophilic TAA (L-TAA) and total carotenoids. Overall, results demonstrated that MeSA applied as postharvest vapours is an effective and environmentally friendly tool to maintain sweet cherry quality during storage with enhanced bioactive compounds concentration and antioxidant activity.

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is deeply reduced during transit, storage and retailing, mainly due to visual and textural changes, increased microbial contamination and decay (Valero and Serrano, 2010).

In recent years, there has been an increasing interest in using salicylic acid (SA) and its derivatives acetyl salicylic acid (ASA) and methyl salicylate (MeSA) owing to their potential to extend the shelf-life of fresh produce and to reduce chilling injury and susceptibility to decay (Cao et al., 2010; Sayyari et al., 2011a,b). Specifically in sweet cherry, SA and ASA applied as dip postharvest treatment delayed the ripening process, maintained fruit quality attributes such colour, firmness and acidity, and enhanced bioactive compounds with antioxidant activity (Valero et al., 2011). In addition, SA and ASA when applied as preharvest treatments during sweet cherry on-tree growth led to fruit with greater size and weight, firmness, total soluble solids (TSS), total phenolics and anthocyanins and antioxidant activity at time of harvest (Giménez et al., 2014).

MeSA could be applied either as a vapour or as dips (Sayyari et al., 2011a; Glowacz and Rees, 2016) when prepared in aqueous solution. A MeSA preharvest application (at 0.5, 1 or 2 mM) in three sweet cherry tree cultivars ('Sweetheart', 'Sweet Late' and 'Lapins') induced fruit with higher fruit quality attributes at harvest and delayed the postharvest ripening process as manifested by a lower degree in colour changes, and lower acidity and firmness losses in

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fruit from treated trees with respect to controls, leading to maintenance of fruit quality (Giménez et al., 2015). In addition, preharvest MeSA treatments increased hydrophilic antioxidant activity (H-TAA) due to increased levels of phenolics and anthocyanins at harvest, which were also maintained at higher levels during storage (Valverde et al., 2015). These MeSA effects included a delay on the postharvest ripening process which was attributed to the higher content in antioxidant compounds such as phenolics and anthocyanins as well as the higher activity of the antioxidant enzymes catalase (CAT), peroxidase (POX), superoxide dismutase (SOD) and ascorbate peroxidase (APX) which would contribute to scavenging reactive oxygen species (ROS) related to fruit ripening and senescence process (Valverde et al., 2015).

On the other hand, postharvest treatment with exogenous MeSA delayed the postharvest ripening process in pomegranate, manifested by maintenance of fruit firmness, total soluble solids (TSS) and total acidity (TA) while significant losses occurred in control pomegranates (Sayyari et al., 2011a). This treatment was also effective in reducing chilling injury (CI). Accordingly, CI was retarded in tomato (Fung et al., 2006) and mango (Han et al., 2006) fruit by MeSA postharvest treatment throughout protecting cell wall and membranes structure from dysfunction caused by lipid peroxidative injury.

However, as far as we know there is no scientific literature on the role of postharvest MeSA treatments on sweet cherry quality attributes. In this sense, the aim of this work was to evaluate the effect of postharvest MeSA treatments (at 2 concentrations, 0.1 and 1 mM) on 'Early Lory' sweet cherry cultivar quality during 20 days of cold storage, with special focus on antioxidant activity and related bioactive compounds, given their implication in the beneficial effect of sweet cherry consumption for the human health.

2. Material and methods

2.1. Plant material and experimental design

Sweet cherries were manually harvested at commercial ripening stage and transferred to laboratory in 2 h. A total of 780 fruits, homogeneous in colour and size and without visual defects were selected. Three lots of 20 cherries were selected at random and used to analyse fruit properties at harvest time. The remaining 720 fruits were divided into 9 lots of 80 fruits to perform MeSA treatments at 20 °C in triplicate (0, control, 0.1 and 1.0 mM MeSA concentration, purchased from Sigma, Sigma-Aldrich, Madrid, Spain). After placing the cherries in a 10L air-tight container, the head space was calculated (9.4 L) in which the appropriate volume of MeSA (0.167 and 1.67 mL) to reach the desired concentration (0.1 and 1 mM, respectively) was deposited on filter paper placed inside a 25 mL plastic vial (to avoid direct contact with fruit) at the bottom of the container. The container was immediately hermetically-sealed and taking into account the rapid volatilization of MeSA the desired concentration was achieved very quickly, as reported by Wang et al. (2015). Control fruit were also sealed in similar containers, but without MeSA addition. Duration of the treatment was 16 h, after which the fruits from each replicate were grouped randomly in lots of 20 fruits and stored in a temperature-controlled chamber at 2 °C, in permanent darkness and with a relative humidity of 90%. After 5, 10, 15 and 20 days of storage one lot from each replicate and treatment was taken for analytical determinations.

2.2. Weight loss and respiration rate determination

Weight loss of each individual lot was calculated as % with respect to the weight at day 0. Respiration rate was measured (at 20 °C for day 0 and at 2 °C for all sampling dates) by placing each lot in a 1 L glass jar hermetically sealed with a rubber stopper for 30 min and CO₂ was quantified using a ShimadzuTMGC-14B gas chromatograph (Kyoto, Japan), equipped with thermal conductivity detector (TCD). Results were the mean \pm SE and expressed as mg CO₂ kg⁻¹ h⁻¹.

2.3. Fruit quality parameters

Colour was determined on each cheek of 20 fruit from each replicate by using a Minolta colorimeter (CRC200, Minolta Camera Co., Japan), using the CIELab coordinates and expressed as Hue angle. Fruit firmness was determined independently in 20 fruit of each replicate using a TX-XT2i Texture Analyzer (Stable Microsystems, Godalming, UK) interfaced to a personal computer, with a flat steel plate mounted on the machine. For each fruit, the cheek diameter was measured and then a force that achieved a 3% deformation of the fruit diameter was applied. Results were expressed as the force-deformation ratio (Nmm⁻¹) and were the mean \pm SE. After that the 20 fruits of each lot were cut in small pieces to obtain a homogeneous sample for each replicate. Total soluble solids (TSS) were determined in duplicated in the juice obtained from 5g of each sample with a digital refractometer Atago PR-101 (Atago Co., Ltd., Tokyo, Japan) at 20°C, and expressed as g 100 g^{-1} (mean \pm SE). Total acidity (TA) was determined in duplicate in the same juice by automatic titration (785 DMP Titrino, Metrohm) with 0.1 N NaOH up to pH 8.1, using 1 mL of diluted juice in 25 mL distilled H_2O , and results (mean \pm SE) expressed as g malic acid equivalent 100 g^{-1} fresh weight. The ratio between TSS and TA was calculated and expressed as a ripening index.

2.4. Bioactive compounds and antioxidant activity

Total phenolics were extracted according to protocol described in Serrano et al. (2009) using water:methanol (2:8) containing 2 mM NaF (to inactivate polyphenol oxidase activity and prevent phenolic degradation) and quantified in duplicate using the Folin-Ciocalteu reagent and results (mean \pm SE) were expressed as mg gallic acid equivalent 100 g⁻¹ fresh weight. Total anthocyanins were extracted and determined according to previously reported (Serrano et al., 2005) and calculated as cyanidin 3-glucoside equivalent (molar absorption coefficient of 23,900 L cm⁻¹ mol⁻¹ and molecular weight of 449.2 g mol⁻¹) and results (mean \pm SE) expressed as mg 100 g^{-1} FW. Total carotenoids were extracted according to Valero et al. (2011). Briefly, 2 g of sweet cherry fruit were extracted with acetone and shaken with diethyl ether and 10% NaCl for separation of the two phases. The lipophilic phase was washed with Na₂SO₄ (2%), saponified with 10% KOH in methanol, and the pigments were subsequently extracted with diethyl ether, evaporated and then made up to 25 mL with acetone. Total carotenoids were estimated by reading the absorbance at 450 nm and expressed as mg of β -carotene equivalent 100 g⁻¹ FW, taking into account the $\varepsilon_{\rm cm}^{1\%}$ = 2560 and results were presented as means \pm SE.

Total antioxidant activity (TAA) was quantified in duplicated for each sample as previously described (Serrano et al., 2009). Briefly, 5 g fresh flesh tissue were homogenised in 5 mL of 50 mM Na-phosphate buffer pH 7.8 and 3 mL of ethyl acetate, then centrifuged at $10,000 \times g$ for 15 min at 4 °C. The upper fraction was used for total lipophilic antioxidant activity (L-TAA) and the lower for total hydrophilic antioxidant activity (H-TAA). In both cases, TAA was determined using the enzymatic system composed of the chro-mophore 2,2'-azino-bis-(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS), the horse radish peroxidase enzyme (HRP) and its oxidant substrate (hydrogen peroxide, Download English Version:

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