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Effects of 1-methylcyclopropene (1-MCP) and modified atmosphere packaging on postharvest storage quality of nectarine fruit

Okan Özkaya[a,*], Deniz Yildirim[b], Ömür Dündar[a], S. Seyhan Tükel[c]

[a] University of Cukurova, Faculty of Agriculture, Department of Horticulture, 01330 Adana, Turkey
[b] University of Cukurova, Vocational School of Ceyhan, Adana, Turkey
[c] University of Cukurova, Faculty of Sciences and Letters, Department of Chemistry, 01330 Adana, Turkey

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ABSTRACT

The goal of this study was to compare the effects of modified atmosphere packaging (MAP), which is commonly used for the storage of nectarine fruit, and treatment with 1-methylcyclopropene (1-MCP: 0.5 or 1 μL L^{-1} for 24 h at 0 °C), an ethylene inhibitor, on the quality of **nectarines cv. 'Maria Aurelia'**. After harvest, nectarine fruits were subjected to 1-MCP or MAP treatments. Following the treatments, the fruits were then kept at 0 °C for 40 days and at 20 °C for an additional 2 days to simulate its shelf life. Fruit flesh firmness significantly decreased during storage and shelf life, for all treated fruits. Both 1-MCP doses and MAP maintained firmness better than control fruits. Chilling injury (CI) was first observed after 20 days at 0 °C + 2 days at 20 °C and then further increased during cold storage and shelf life period for all treatments. Compared with control fruits, MCP treated or MAP-stored nectarines showed significantly reduced incidence of CI and polyphenol oxidase (PPO), polygalacturonase (PG) and pectin methyl esterase (PEM) activities as well as lower hue angles (h°), total soluble solid (TSS) contents and respiration rates. These results showed that both methods maintained the post-harvest quality of nectarine fruit and provided longer storage and shelf life.

1. Introduction

Nectarines are climacteric fruit, and rapid ripening of fruit during storage results in a short storage life (Cantín et al., 2010). The conventional method for slowing the ripening and prolonging the storage life of nectarines is cold storage (Dong et al., 2013). However, chilling injury (CI), a collective term describing a physiological disorder that manifests primarily as flesh browning resulting from membrane lipid oxidation, may limit such storage (Lurie and Crisosto 2005; Fruk et al., 2014). CI symptoms are of commercial importance because shipping of peaches or nectarines to distant markets and storage before selling require low temperatures (Campos-Vargas et al., 2006). Therefore, storage and shelf life extension to avoid post-harvest losses of nectarine fruit has been a long-standing goal for producers and traders.

Polygalacturonase (PG, EC 3.2.1.15) is an enzyme that catalyzes the hydrolysis of specific linkages in galacturonides and other polysaccharides. Pectin methyl esterase (PME, EC 3.1.1.11) catalyzes the cleavage of the ester bonds of pectin molecules. During the softening process of most fruits, total pectin amounts generally decrease, and the contents of water-soluble pectic substances increase. Therefore, there is a proportional relationship between the softening of fruit and the activity of pectin degradation enzymes (Imsabai et al., 2002). Polyphenol oxidase (PPO, EC 1.14.18.1) catalyzes the oxidation of polyphenols to quinones, a process that is responsible for browning reactions in fruits and vegetables (Waliszewski et al., 2009; Özkaya et al., 2015).

1-Methylcyclopropene (1-MCP) is an ethylene action inhibitor that is used to delay the ripening process and to extend the storage as well as shelf life of climacteric fruits and vegetables (Watkins, 2006). In the literature, it has been reported that 1-MCP delays the ripening process and extends the storage life of many fruits and vegetables, including apple (Zheng et al., 2014), avocado (Meyer and Terry, 2010), pear (Liu et al., 2013), and fig fruit (Ozkaya et al., 2014).

Modified atmosphere packaging (MAP) is commercially used to increase the shelf life of packaged produce by reducing the produce respiration rate, delaying senescence, and ultimately increasing product shelf life. In MAP, the gaseous content of the package is altered by the addition and removal of gases to regulate the O_2 and CO_2 levels. This manipulation can lead to reduced respiration, delayed ripening, decreased ethylene production, and slower textural softening, thereby extending the shelf lives of fruits and/or vegetables (Jia et al., 2009; Sivakumar et al., 2012). Moreover, MAP

* Corresponding author. Fax: +90 3223386388.
E-mail address: oozkaya@cu.edu.tr (O. Özkaya).

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incurs high labor costs during the packaging and cooling processes and also reduces the processing speed. Exposing nectarines to high concentrations of 1-MCP may obviate the need for MAP storage, and the use and application of 1-MCP are easier than the MAP process. However, the application of 1-MCP does not achieve the desired level of results in stone fruits.

There is little information available regarding the effects of 1-MCP treatment and MAP on physiological and biochemical changes in the nectarine. Moreover, the modes of action of 1-MCP and MAP in alleviating CI and quality deterioration as well as the roles of PG, PME, and PPO remain unclear, and further research is needed to elucidate the physiological mechanisms involved (Fruk et al., 2014). Thus, the purpose of the present study was to elucidate the effects of 1-MCP and MAP on the development of CI in nectarine fruit. In addition, the fruit quality and physiological and biochemical changes associated with 1-MCP treatment and MAP storage were also examined.

2. Materials and methods

2.1. Materials

Nectarines (*Prunus persica* cv. 'Maria Aurelia') were obtained from a commercial orchard in Nigde province, Turkey. Pectin from citrus peel, 3,5-dinitrosalicylic acid (DNSA, 98%), galacturonic acid, pyrocatechol (≥99%) and acetone were purchased from Sigma–Aldrich (St. Louis, MO, USA). All other chemicals used in this study were of analytical grade.

2.2. 1-MCP treatment and MAP storage

Fruits were harvested from trees grafted on GF-677 rootstocks at firm-ripe stage and immediately transported via ventilated truck to cold storage facilities of Department of Horticulture, Faculty of Agriculture, Cukurova University where they were sorted and selected for similar size, uniform maturity and appearance and freedom from defects.

The nectarine fruits were divided into 4 treatment groups:

a) Control: the fruit was placed into plastic boxes without any treatment.
b) MAP: MAP was achieved using the commercially available 5-kg-capacity Xtend® bags (CO_2 transmission rate:5000–18,000 $cm^3/[m^2\ day^{-1}]$; water vapor transmission rate: 10 g (mm-$m^2\ day^{-1}$) at 25 °C and atmospheric pressure). After placing approximately 5 kg of fruit inside the MAP, the bags were sealed.
c) 0.5 $\mu L\ L^{-1}$ and d) 1 $\mu L\ L^{-1}$ 1-MCP: for the application of 1-MCP, the fruits were placed into a 65-L plastic container and exposed to 0.5 or 1 $\mu L\ L^{-1}$ 1-MCP for 24 h. This 1-MCP was released from a commercial powder formulation (SmartFresh™ AgroFresh, Inc., Dow AgroSciences, Philadelphia, PA, USA) by adding distilled water according to the manufacturer's instructions.

All treatments were placed inside 5-kg commercial plastic boxes and then transferred to a storage room at 0 °C and 90% relative humidity for 40 days. The fruit was analyzed at 10-day intervals during cold storage and after additional 2 days at 20 °C following cold storage to simulate its shelf life. Three replicates were analyzed for each trial, with 8 nectarines per replicate, for a total of approximately 300 kg in the experiments.

2.3. Evaluation of CI and fruit firmness

The CI incidence was estimated based on flesh mealiness and flesh browning and was calculated as the percentage of water-soaked or browned fruit per treatment during storage. Fruit flesh firmness was determined using a fruit firmness tester (TR-FT327, Italy) equipped with an 8-mm-diameter tip and is expressed in Newtons (N).

2.4. Fruit ethylene production and respiration rate

Ethylene production was detected using a gas chromatography equipped with an alumina column and flame ionization detector (Shimadzu 14 B, Japan). For each measurement, five nectarines for each of 3 replicates were enclosed in a 2-L glass jar for 3 h, and 1 mL of headspace gas sample was withdrawn from the jar and injected into the gas chromatography device. The ethylene production rate is expressed as nL $g^{-1}\ h^{-1}$.

CO_2 production was measured from the enclosed jars for ethylene testing using an Isolcell (Italy) CO_2 analyzer and the respiration rate is expressed as mL CO_2 $kg^{-1}\ h^{-1}$.

2.5. Fruit peel color and total soluble solid (TSS) contents

The color (hue angle) was measured using a chromameter (Minolta CR300, Japan). The color of each fruit was measured in terms of the L*, a* and b* coordinates, and from these values, the hue angle was calculated as $h^\circ = \arctan(b/a)$ (Abbott, 1999). TSS content (%) was assessed in juice samples collected from five nectarines per treatment using a hand-held refractometer (Atago, Japan).

2.6. Measurement of PPO, PME and PG activities

2.6.1. Preparation of PPO, PME and PG enzyme extracts

Approximately 100 g of fruit pulp was homogenized in 200 mL of chilled acetone using a Waring blender at high speed for 2 min, followed by filtration. The residue was returned to the Waring blender, and 200 mL of chilled acetone was added, after which the fruit pulp was homogenized. This procedure was repeated for a total of three extractions. The solvent was discarded each time. The resulting white powder was maintained at 5 °C for 12 h and then stored at −20 °C until use.

For PG analysis, 0.5 g of the powder was suspended in 50 mL of acetate buffer (50 mM, pH 4.0), whereas for PME and PPO activity analyses, 0.5 g of powder was suspended in 50 mM phosphate buffer (pH 7.5 or pH 6.5). After stirring at 4 °C for 1 h, the mixtures were centrifuged at 10,000 × *g* for 10 min, and the supernatants were used as sources of PG, PME or PPO.

2.6.2. Measurement of PPO, PME and PG activities

2.6.2.1. PPO activity. PPO activity was measured spectrophotometrically (Shimadzu 1210, Japan) at 410 nm. Briefly, 0.5 mL of the PPO extract and 0.5 mL of a catechol solution were added to 2 mL of phosphate buffer (50 mM, pH 6.5). After incubation of the reaction mixture at room temperature for 5 min, the absorbance of the formed product was monitored at 410 nm at 15 s intervals. One unit of PPO activity was defined as the amount of enzyme that caused a change in the absorbance of 0.001 min^{-1} (Huang et al., 2009).

2.6.2.2. PG activity. The measurement of pectinase activity was performed using the method described by Bernfeld (1955). Briefly, 0.5 mL of the PG extract was added to 0.5 mL of a pectin solution prepared in acetate buffer (50 mM, pH 4.0). The reaction was carried out at 25 °C for 15 min. Then, 0.5 mL of DNSA solution was added, and the reaction mixture was boiled for 15 min in a water bath. After cooling to 25 °C, 4.5 mL of distilled water was added to the mixture, and its absorbance was measured at 540 nm. One unit of pectinase activity was defined as the production of 1 μmol of galacturonic acid in 1 min/mg of protein.