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Physiological response of persimmon to 1-MCP post-harvest treatments in long-term cold storage

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Abstract: This research studies the effects of 1-methylcyclopropene (1-MCP) post-harvest treatment on chilling injuries and quality of persimmon fruit cv. 'Karaj' during a four-month cold storage. Every 15 d in the cold phase, several physiological parameters were measured. The results illustrated that 1-MCP postharvest treatment with different concentrations (equalling to 0.5, 1 and 2 μL L^{-1}) meaningfully reduces chilling injuries of the persimmon fruit. The loss of weight, flesh firmness, titratable acidity, total phenolics, soluble tannins and antioxidant capacity can be alleviated using higher 1-MCP concentrations. These results can be related with alterations in antioxidant enzymes' activities together with protection of fruit′s membrane integrity. Based on the results, the storage duration recommended for this cultivar is 45 d by application of 1–2 μL L^{-1} of 1-MCP.

Keywords: antioxidant capacity; membrane integrity; chilling injury; persimmon.

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Biographical notes: Maryam Bagheri investigated on post-harvest quality improvement of several products aiming to reduce losses and waste along the fruit supply chain and increase fruit shelf life, at the University of Bu-Ali Sina.

Her research covered effects of variable post-harvest treatments and storage conditions on persimmon quality. Currently, she is a member of Plant Technology Team at the Catholic University of Portugal.

Mahmood Esna-Ashari is a Full Professor of Plant Physiology and Biotechnology at the Department of Horticultural Sciences, University of Bu-Ali Sina (BASU). During his more than 20-year tenure at BASU, he held several positions at the university, including director of the Faculty of Agriculture. He has several research works with different topics in physiology, post-harvest technology, biotechnology and so on.

Ahmad Ershadi holds a PhD from the University of Tehran on molecular identification of S-alleles associated with self-incompatibility in apple (Malus spp.) genotypes (2003). Then, he started working at the University of Bu-Ali Sina where he holds now an associate professor position. He has co-authored +70 papers in international journals and has a long and successful track record in coordination and participation in projects related to genetic diversity, fruit biotechnology, and physiology.

1 Introduction

Persimmon (*Diospyros kaki*), as a favourite fruit for customers, has high amounts of natural antioxidant compounds such as carotenoids, tannins, flavonoids, anthocyanidin, etc. Persimmon, considered as a bioactive fruit in daily diet, alleviates threat of some diseases including diabetes and cardiovascular disorders as well as some cancers (Yaqub et al., 2016; Butt et al., 2015). So far, no significant study has been performed on post-harvest physiological problems and prolonged storage life of this fruit in Iran. This fact subsequently shortens the supply period of persimmon in market.

Concerning tropical and sub-tropical fruits, chilling injury is a critical disorder which appears in low temperature (Wang, 2004). Although normal temperature for fruit storage is below 8°C, this condition increases chilling injuries in persimmon. Chilling symptoms appear when storage phase is passed and during exposure of the fruit to room temperature. Browning, flesh gelling, darkness of skin, loss of firmness, internal tissue breakdown, accelerated senescence, and increase in acetaldehyde production are among the common chilling symptoms in persimmon fruit (Paull, 1990; Salvador et al., 2004, 2005).

Since ethylene hormone affects fruit ripening process, main goals of preserving quality of the fruit during post-harvest is established on diminution of metabolic processes. This includes respiration besides ethylene production. So, in order to moderate post-harvest lesions, use of an ethylene-receptor blocker, i.e., 1-methylcyclopropene (1-MCP), is suggested by some investigators (e.g., Ansari and Tuteja, 2015). Chilling injury symptoms may develop by disbalance in cellular oxidative metabolism and some oxidative enzymes like peroxidase (POD) and catalase (CAT) (Sala and Lafuente, 2000; Hershkovitz et al., 2005). Also, chilling disorders are directly related to oxidative stress resulted from production of reactive oxygen species (ROS) and subsequently, by peroxidation and membrane degradation (Sevillano et al., 2009; Valenzuela et al., 2017). Since diminution of cell membrane integrity is a principal index of chilling injury,

normally the membrane damage is explored by electrolyte leakage and malondialdehyde (MDA) content (Parkin and Kuo, 1989; Kratsch and Wise, 2000).

1-MCP treatment, as a complementary strategy to cold storage, has extensive usage in both research and commercial techniques (Lurie and Paliyath, 2009). Several studies have demonstrated that it is non-toxic to human, is effective at very low concentrations and the amounts of its residue on products is negligible (Watkins, 2006). Advent of this new compound has revolutionised the knowledge of maintaining quality and reducing chilling injuries of many horticultural products including cut flowers, vegetables and fruits (Cheng et al., 2015; Aghdam et al., 2016; Özkaya et al., 2016). As an inhibitor of ethylene, 1-MCP interrelates with its receptors in the fruit to prevent the hormonal action (Watkins, 2015). It causes a delay at the initial phase of ethylene production which consequently postpones increase of respiration rate, volatile production, softening and finally senescence process (Larrigaudière et al., 2009). It is also reported that oxidative metabolism changes after application of 1-MCP treatment (Zhang et al., 2010). Also, 1-MCP prevents degradation of cell walls and maintains mechanical structure of the cell (Jiang et al., 2014; Zhang et al., 2012). In total, researchers have mentioned that 1-MCP treatment on persimmon is associated with:

1 reduction of softening

2 cell membrane integrity

3 delay in internal and external browning

4 delay in the increasing phase of respiration and ethylene production

5 improvement of antioxidant system (Salvador et al., 2004; Zhang et al., 2010; Öz, 2011; Novillo et al., 2015).

To the best of the authors' knowledge, there is a lack of investigation on long period storage of persimmon using 1-MCP. Most of the works (e.g., Novillo et al., 2015; Besada et al., 2008; Rasouli and Khademi, 2018; Khademi et al., 2014; Pérez-Munuera et al., 2009) have studied persimmon, affected by 1-MCP with a single concentration 0.5 μL L^{-1}, during a period of less than 45 d; Furthermore, the mentioned works never examined higher concentrations of 1-MCP. Some investigators (e.g., Li et al., 2018; Min et al., 2018; Özkaya et al., 2016; Salvador et al., 2004) evaluated persimmon treated by various 1-MCP concentrations less than 1 μL L^{-1}, for a period of less than 70 d, based on parameters like firmness, total soluble solids (TSS), soluble tannin, POD and CAT enzyme activities; These works either were limited to a moderate cold storage duration or suffered from incomplete study parameters. It seems that there is only one investigation (Öz, 2011) where it studied persimmon under a single 1-MCP concentration of 2 μL L^{-1} based on the fruit's firmness, titratable acidity (TA) and TSS during 90 d cold storage. As it is obvious, there is a necessity for exploration of different physicochemical and chilling injury parameters, using different concentrations of 1-MCP, and during a longer period. The goal of this study is to investigate cold storage effects on persimmon cv. 'Karaj' fruit treated by 0, 0.5, 1 and 2 μL L^{-1} 1-MCP during 120 d.

2 Materials and methods

In this study, the fruit quality is measured in terms of firmness, weight loss, TSS, TA, total phenolic content (TPC), and soluble tannin. In addition, the chilling injuries are evaluated using electrolyte leakages, MAD contents, antioxidant activities, and peroxidase/catalase enzyme activities. Preparation of the samples and extraction of the enzyme from the persimmon in this investigation are as presented in Bagheri and Esna-Ashari (2021).

2.1 Orchard and 1-MCP treatment

Orange and hard persimmon (mature fruit) were harvested from an orchard in Karaj City.[1] After packing the fruit, they were sent (in ambient temperature 21°C–25°C) to Hamedan City in three hours. Fruits were checked for pests, diseases and physical damages. Four collections (each group consists of 360 fruits) were selected on the basis of firmness and size uniformity. Further, each collection (group) was divided into three sets, (each set consists of 120 fruits) per replication per group. Each group of the fruit was kept in a separate compartment. The first compartment was defined as control. The other three groups were subjected to concentrations of 0.5, 1 and 2 μL L^{-1} of gaseous 1-MCP during 24 h at 20°C. Then, the persimmon were reserved in a storage (0°C–1°C) for four months. During the storage time, quality of the persimmon and their chilling damage were examined each 15 d followed by two more days of resting in the room temperature (equivalent to the shelf life to allow chilling disorders appear). Although fruit's colour change is considered as an important parameter, this factor was not monitored in this study.

2.2 Fruit firmness and weight loss

The rate of weight loss (in percentage) was determined using:

$$\%\, Weight\ loss = \frac{W1 - W2}{W1} \times 100$$

where $W1$ and $W2$ are the fruit weights (g) before and after the storage, respectively. Five unpeeled fruit samples were arbitrarily selected and tested in each replication. To examine the fruit firmness, an analysis according to the one explained in Bagheri and Esna-Ashari (2021) was performed at three distinct sites from the equatorial ring of the fruits.

2.3 Chilling injuries

Evaluation of chilling injuries was carried out 2 days after fruit removal from the storage. Ten persimmons were considered per each replication. Chilling symptoms were scaled from 0 to 4, where 0 represented no chilling symptom. The degrees 1, 2, 3, and 4 signified respectively less than 1⁄4, 1⁄4 to 1⁄2, 1⁄2 to 3⁄4, and more than 3⁄4 browning and gelling area. Chilling injury (%) was calculated as $100 \times [(N_1 + 2N_2 + 3\ N_3 + 4\ N_4) / (4\ N)]$, with N_1 to N_4 equal to the number of the fruits with chilling scale 1 to 4, respectively, and N equal to the total number of the fruits.

2.4 TA and TSS

TA and TSS were determined based on the method explained in Bagheri and Esna-Ashari (2021). The TA and TSS values are reported as % malic acid and % brix, respectively.

2.5 Total phenolic content

Colorimetric Folin-Ciocalteu assay according to Singleton et al. (1999) and Lee et al. (2004) was applied to measure the TPC. The method used to prepare the reaction mixture can be found in Bagheri and Esna-Ashari (2021). The TPC, measured by gallic acid calibration curve, is expressed as gallic acid equivalent in g kg^{-1} of the fresh weight (FW).

2.6 Soluble tannin

Adjusted Folin-Denis method, as a rapid colorimetric technique (del Río and Navarro, 2004), was used to assess the soluble tannin. This procedure is established on reduction of Folin-Denis reagent through tannin exposed to an alkaline medium. For the calculation of calibration curve, gallic acid was employed. To find more detail on this method, one can refer to Bagheri and Esna-Ashari (2021). In this study, the amount of tannin is stated as mg kg^{-1} of FW.

2.7 Total antioxidant activity

Extracts (in methanol) at different concentrations were treated with DPPH solution based on the procedure explained in Bagheri and Esna-Ashari (2021). The DPPH scavenging capacity (inhibition rate, %) was measured at the wavelength 517 nm, using $100 \times [(A_C - A_S) / A_C]$, in which, A_S and A_C indicated, respectively, the absorbance of the samples and the blank absorbance (methanol + DPPH reagent).

2.8 Analysis of MDA

MDA was examined according to the method of Chen and Wang (2006). The supernatant absorbance, at 600, 532, and 450 nm, was defined after centrifugation. The following formulation was used to calculate the accumulated MDA: $(A532 - A600) \times 6.45 - A450 \times 0.56$. The MDA is stated as μmol kg^{-1} of FW.

2.9 Electrolyte leakage

To define the total electrolyte leakage, the procedure given in McCollum and McDonald (1991) was slightly modified. In this analysis, three fruit (in each replication) was utilised to obtain six pericarp discs (each one with 10 mm diameter). The leakage data are measured based on the expression $(E_i / EC) \times 100$ and expressed as % of the total EC.

2.10 Enzyme extraction

The activities of catalase (CAT) and peroxidase (POD) enzymes were determined once the cold storage terminated. According to Plewa et al. (1991), peroxidase activity was assessed by means of guaiacol in the role of substrate. Increase in the absorption was due to oxidation of guaiacol and tetraguaiacol formation. The absorbance was measured at 470 nm. Peroxidase enzyme activity is given as unit g^{-1} FW per minute.

CAT activity was measured according to the procedure described in Dhindsa and Matowe (1981). The amount of catalase activity was assessed by means of decrease in absorbance of H_2O_2 at 240 nm. The results are stated as unit (U) g^{-1} FW. The unit U is defined as the amount of catalase enzyme that decomposes 1 μmol of H_2O_2 per minute at 25°C.

2.11 Statistical analysis

The tests were performed in split plot experiment, by three replications. The whole-plot factor was the 1-MCP treatment (at four levels) and the split-plot factor was the storage time. Analysis of variance by statistical analysis systems (SAS) programmers was carried out to examine the experimental data. SAS was also employed in regression and correlation analysis.

3 Results and discussion

3.1 Weight loss

As Table 1 confirms, both the treated and the control fruits lost weight through the storage time. 1-MCP treatment reduced the weight loss of the fruit. After four months, the minimum weight loss (6.08 %) was seen in the fruit dealt with 2 μL L^{-1} 1-MCP in contrary to the control group which had the maximum weight loss (10.61%). No significant difference was observed between 0.5 μL L^{-1} 1-MCP treatment and the control, during the evaluation ($P \leq 0.01$).

Weight loss reduction in the treated persimmon can be due to reduction in respiration rate. It was reported in Becker and Fricke (1996) that the main factors for weight loss during storage time are loss of water, due to transpiration, and carbon loss, related with fruit respiration. The prevention of weight loss via 1-MCP was also stated in avocado (Jeong et al., 2002) and in persimmon cv. 'Hachiya' (Qasid et al., 2017).

3.2 Fruit firmness

A reduction of the fruit firmness occurred through time (Table 1). The initial firmness was 8.13 N, and after 120 d, the maximum firmness (5.73 N) was obtained in the persimmon treated by 2 μL L^{-1} 1-MCP. Differently, the minimum firmness was measured in the control group (2.76 N). No significant difference between the control group and the treated fruit by 0.5 μL L^{-1} 1-MCP was observed. Softening rate was also delayed in the persimmon dealt with 1 μL L^{-1} 1-MCP when compared against the control group.

Table 1 Mean comparison of weight loss, tissue firmness, titratable acidity, total soluble solid content and chilling injury of persimmon fruit cv. 'Karaj' treated with various concentrations of 1-MCP, through 120 d of storage at 0°C

Storage period (*d*)	*1-MCP treatment* ($\mu L\ L^{-1}$)	*Weight loss* (*%*)	*Firmness* (*N*)	*TA* (*% malic acid*)	*TSS* (*%*)	*Chilling injury* (*%*)
0	–	0.0	8.13	0.68	11.20	0.00
15+2	0	3.13^{a}	8.10^{a}	0.64^{a}	12.80^{a}	6.25^{a}
	0.5	2.76^{a}	8.06^{a}	0.66^{a}	12.33^{ab}	4.16^{a}
	1	2.44^{a}	8.10^{a}	0.67^{a}	11.66^{b}	0.00^{b}
	2	2.19^{a}	8.13^{a}	0.68^{a}	11.60^{b}	0.00^{b}
30+2	0	4.21^{a}	7.40^{b}	0.56^{c}	14.40^{a}	12.50^{a}
	0.5	3.67^{ab}	7.43^{b}	0.60^{b}	13.26^{b}	10.41^{a}
	1	2.89^{b}	7.76^{a}	0.62^{ab}	13.26^{b}	4.16^{b}
	2	2.51^{b}	7.93^{a}	0.64^{a}	12.73^{b}	0.00^{b}
45+2	0	5.18^{a}	6.86^{c}	0.43^{c}	16.06^{a}	16.66^{a}
	0.5	4.81^{a}	7.16^{b}	0.47^{b}	15.33^{ab}	12.50^{ab}
	1	3.73^{b}	7.36^{ab}	0.52^{ab}	15.00^{ab}	8.33^{abc}
	2	3.16^{b}	7.56^{a}	0.55^{a}	14.33^{b}	4.16^{c}
60+2	0	6.26^{a}	6.10^{b}	0.37^{c}	17.00^{a}	22.91^{a}
	0.5	5.90^{a}	6.26^{b}	0.40^{bc}	16.80^{a}	18.75^{a}
	1	4.99^{ab}	6.93^{a}	0.44^{ab}	15.48^{b}	12.5^{b}
	2	4.12^{b}	6.93^{a}	0.46^{a}	14.94^{c}	8.33^{b}
75+2	0	7.10^{a}	5.43^{b}	0.28^{b}	18.40^{a}	29.16^{a}
	0.5	6.90^{ab}	5.56^{b}	0.30^{ab}	17.86^{b}	22.91^{ab}
	1	5.37^{b}	6.03^{a}	0.36^{a}	17.40^{b}	16.66^{bc}
	2	4.57^{c}	6.30^{a}	0.36^{a}	16.60^{c}	10.41^{c}
90+2	0	7.96^{a}	4.06^{b}	0.22^{b}	18.56^{a}	45.83^{a}
	0.5	7.94^{a}	4.53^{b}	0.27^{ab}	18.33^{a}	27.08^{b}
	1	6.74^{ab}	5.60^{a}	0.30^{a}	17.53^{b}	18.75^{bc}
	2	5.13^{b}	6.06^{a}	0.31^{a}	16.93^{b}	12.50^{c}
105+2	0	9.36^{a}	3.56^{c}	0.20^{b}	20.46^{a}	60.41^{a}
	0.5	9.03^{a}	3.76^{c}	0.22^{ab}	20.33^{a}	35.41^{b}
	1	7.19^{b}	5.36^{b}	0.27^{a}	17.60^{b}	22.91^{c}
	2	5.76^{b}	5.80^{a}	0.27^{a}	17.33^{b}	16.66^{c}
120+2	0	10.61^{a}	2.76^{c}	0.17^{b}	21.40^{a}	72.91^{a}
	0.5	10.40^{a}	3.00^{c}	0.19^{b}	21.20^{a}	58.33^{b}
	1	7.93^{b}	4.90^{b}	0.23^{a}	18.06^{b}	31.25^{c}
	2	6.08^{c}	5.73^{a}	0.24^{a}	17.53^{b}	20.83^{d}

Note: Similar subscript letters in each column indicate non-significant differences among treatments $P \leq 0.01$.

Firmness maintenance of persimmon fruit using 1-MCP treatment is most likely due to the reduced ethylene production. Decrease of the ethylene action retains enzymes, which are responsible for cell wall degradation, of being stimulated and therefore the cell wall stiffness will be maintained (Khan and Singh, 2007). Moreover, reduction of the respiration rate has an important role in retaining fruit firmness during storage (Chen and Zhu, 2011). 1-MCP treatment was reported to delay fruit softening in persimmon cv. 'Fuyu' (Zhang et al., 2010) and cv. 'Harbiye' (Öz, 2011) and mango (Razzaq et al., 2016).

3.3 TA and TSS

TA amount decreased in both treated and untreated fruits during the storage phase (Table 1). Generally, TA of the persimmon, during storage, was not significantly affected by various 1-MCP concentrations, however TA of the treated fruit was always more than that of the untreated ones. In the final 15 d, the fruit dealt with 1 and 2 μL L^{-1} 1-MCP showed significantly more TA in comparison with those treated by 0.5 μL L^{-1} 1-MCP and the control group.

The TSS level increased in both treated and control persimmons with storage time (Table 1). This increase is probably due to hydrolysis of polysaccharides, such as starch into sugars. It also might be related to breakdown of complex organic metabolites into simple molecules. Meanwhile, dehydration can be considered as a factor of the concentration of juice content (Wills et al., 1980). During the storage, TSS level in the persimmons subjected to 1 and 2 μL L^{-1} 1-MCP was significantly lower than those dealt with 0.5 μL L^{-1} 1-MCP and the control samples.

Possibly, respiratory metabolism causes conversion of organic acid into sugars, and therefore the amount of TA decreases during storage phase (Kaur et al., 2013). Application of post-harvest 1-MCP treatment on other fruits resulted in better maintenance of TA amount, e.g., in mango (Razzaq et al., 2016). Minas et al. (2013) stated that higher TA in 1-MCP treated fruit can be related to the reduction of fruit respiration.

Blankenship and Dole (2003) mentioned that the effect of 1-MCP on the content of soluble solids depends on the crop. Utilisation of 1-MCP to maintain TSS level is stated in persimmon cv. 'Rendaiji' (Ortiz et al., 2005). In contrary to these reports, treatment by 1-MCP had no effect on TSS in persimmon cv. 'Harbiye' (Öz, 2011).

3.4 Chilling injury

As given in Table 1, the maximum and minimum chilling injury was, respectively, observed in the control (72.9%) and in the persimmon subjected to 2 μL L^{-1} 1-MCP (20.8%). Chilling symptoms were not visible in the treated fruit by 2 μL L^{-1} 1-MCP in the first month of storage, and only in the next months they appeared. There was an indirect relation between the concentration of 1-MCP treatment and the chilling disorder level.

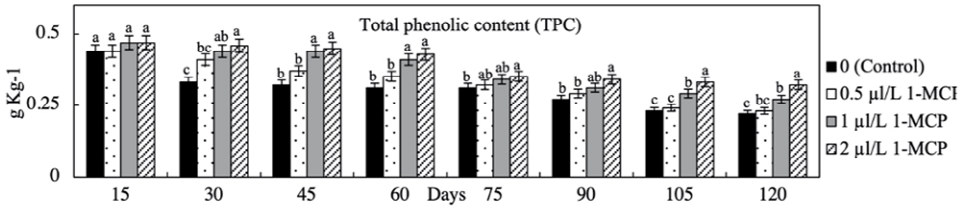
The influence of 1-MCP treatment on reduction of post-harvest chilling disorders was also reported, e.g., in pineapple (Selvarajah et al., 2001), in pear (Cheng et al., 2015) and in persimmon cv. 'Rojo Brillante' (Salvador et al., 2004) and cv. 'Fuyu' (Zhang et al., 2010), in agreement with the present results. Regarding the fact that ethylene production

is necessary for softening process (as an important chilling symptom), the application of 1-MCP treatment, as an inhibitor of the ethylene action, can decrease the chilling injuries (Salvador et al., 2004; Candan et al., 2008).

3.5 Total phenolic content

As Figure 1 displays, during four months of storage, the phenolic compound content of the fruits decreased continuously. The maximum value of the TPC was always found in the persimmon dealt with 2 µL L^{-1} 1-MCP. This content for the fruit dealt with both 2 and 1 µL L^{-1} 1-MCP was not significantly altered until the third month, but in the fourth month, the phenolic compound content was significantly different between both treated samples. The control group maintained always the minimum TPC without any significant difference with the fruit subjected to 0.5 µL L^{-1} 1-MCP. The protection effect of this treatment on TPC was also demonstrated in loquat (Cai et al., 2006), in tomato (Wang et al., 2010) and in peel tissues of apple (Hoang et al., 2011). Kalita and Jayanty (2014) mentioned the critical role of phenolic compounds in the antioxidant capacity. As storage time increases, phenolic compounds decrease by polyphenoloxidase activity and therefore leads to browning of fruit tissue. It seems that post-harvest 1-MCP treatment reduces the synthesis and the activity of polyphenoloxidase enzyme and therefore it can inhibit the reduction of phenolic compounds content (Moreno-Hernández et al., 2014).

Figure 1 Effects of 1-MCP concentrations on total phenollic content of persimmon fruit through 120 d of storage at 0°C



Note: Vertical bars show mean ± SE.

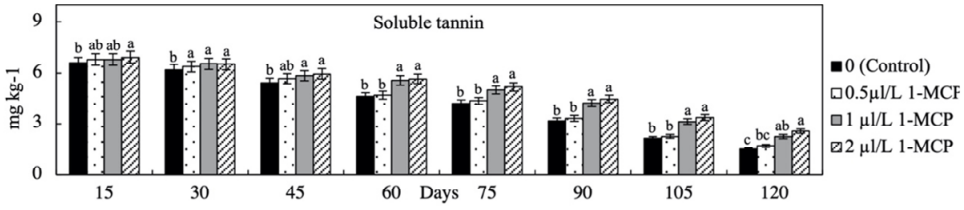
3.6 Soluble tannin

Quantity of soluble tannin in persimmon fruit is linked with the astringent taste. Decrease in astringency over time occurs as acetaldehyde reacts with soluble tannin accompanied by its change into insoluble polymers (Salvador et al., 2008). In the present study, it was seen that the amount of the soluble tannin of the fruit decreased always during the storage phase. Persimmons dealt with 2 and 1 µL L^{-1} 1-MCP had the highest content of the soluble tannin but the lowest amount was seen in the control samples and those affected by 0.5 µL L^{-1} 1-MCP (Figure 2).

The present findings are in good agreement with the results of other references, where 1-MCP treatment postponed polymerisation of the soluble tannins, e.g., in persimmon cv. 'Rojo Brillante' (Besada et al., 2008; Pérez-Munuera et al., 2009).

Besada et al. (2008) believed that the low amount of tannin insolubilisation, in persimmon treated by 1-MCP, can be related with the low rate of acetaldehyde production.

Figure 2 Effects of 1-MCP concentrations on soluble tannin of persimmon fruit through 120 d of storage at 0°C

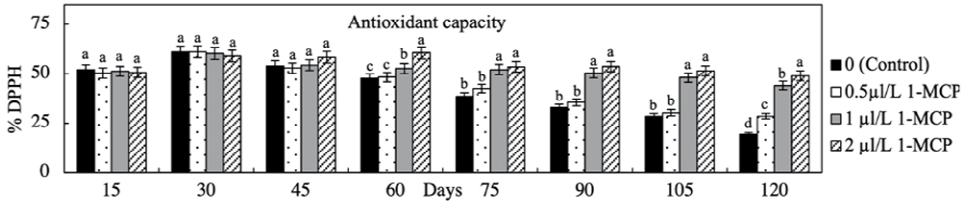


Note: Vertical bars show mean = SE.

3.7 Antioxidant capacity

As Figure 3 exhibits, the level of antioxidant activity decreased in all groups at the ending of the storage phase. In the first 30 d, scavenging activity in the control and treated fruits did not differ significantly. At the end of the first month, a peak in the scavenging activity percentage was seen in all groups. This percentage was retained until the second month, for fruit treated by 2 µL L^{-1} 1-MCP and then decreased until the end of the cold storage phase. In the case of the control group and the other fruit treated by low concentration of 1-MCP, this percentage decreased after one month until the cold storage terminated. The lowest and highest antioxidant activity was seen respectively in the control group and in the fruit subjected to 2 µL L^{-1} 1-MCP.

Figure 3 Effects of 1-MCP concentrations on antioxidant capacity of persimmon fruit through 120 d of storage at 0°C



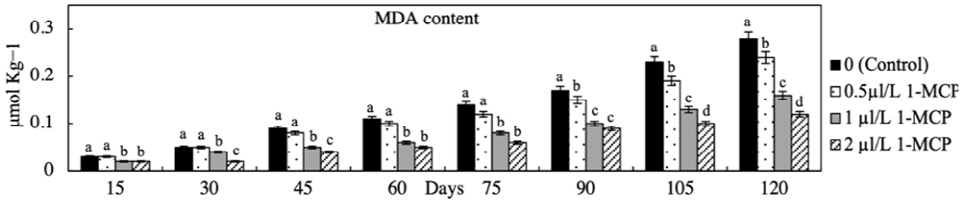
Note: Vertical bars show mean ± SE.

Based on a research by Galani et al. (2017), a decline in antioxidant capacity during cold storage may be attributed to reduced levels of TPC, phenolic acids, anthocyanins, carotenoids and flavonoids. Therefore, general decrease of antioxidant capacity in all the persimmon during the storage can be linked to the gradual decrease of the amount of phenolic content. According to the current result of the TPC measurement, high level of the antioxidant capacity in the persimmons affected by 2 and 1 µL L^{-1} 1-MCP can be correlated to their high level of TPC. Current results about the influence of 1-MCP action on the rate of the antioxidant activity agree with other studies on tomato (Wang et al., 2010) and on peach (Liu et al., 2015, 2018).

3.8 MDA content and electrolyte leakage

As an indicator of lipid peroxidation, the MDA content was evaluated throughout the storage phase. The MDA content in all the treated and the control persimmons gradually increased. The lowest amount for the MDA was seen in the fruit subjected to 2 µL L^{-1} 1-MCP (Figure 4). No significant difference in the MDA content of the fruits affected by both 1 and 2 µL L^{-1} 1-MCP was detected, during the first three months of the cold storage. However, effect of 2 µL L^{-1} 1-MCP treatment, in the last month, was significantly better than the effect of 1 µL L^{-1} 1-MCP treatment. Fruits subjected to 0.5 µL L^{-1} 1-MCP and the control persimmons had a great level of the MDA, without any significant difference between them, until 75 d of the cold storage. Afterwards, the MDA level in the persimmon affected by 0.5 µL L^{-1} 1-MCP became significantly lower than that in the control fruit.

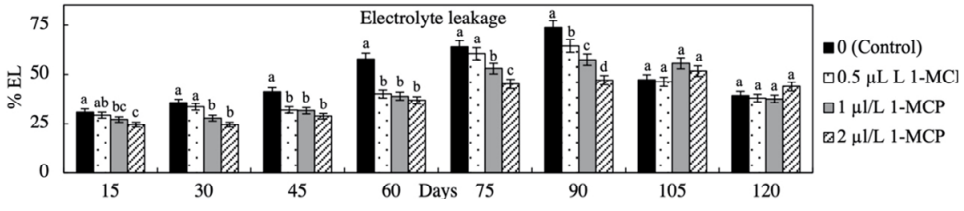
Figure 4 Effects of 1-MCP concentrations on MDA content of persimmon fruit through 120 d of storage at 0°C



Note: Vertical bars show mean ± SE.

In the present study, EL level did not change consistently. According to Figure 5, its maximum level occurred often between 90 d to 105 d for various treatments. The level of EL in the persimmons treated by 2 and 1 µL L^{-1} 1-MCP was often significantly lower than the other treated fruit as well as the control group, in the first 90 d of storage. When the storage finished, EL in the treated fruit with 2 µL L^{-1} 1-MCP was higher than that in the other samples. There was no meaningful difference among the treated and control fruits in the last month.

Figure 5 Effects of 1-MCP concentrations percentage of EL of persimmon fruit through 120 d of storage at 0°C



Note: Vertical bars show mean ± SE.

Exposure to cold temperature during a long time can damage plasma membrane integrity (Zhou et al., 2007). Changes in EL and MDA levels normally occur before a significant chilling symptom appears (Taghipour et al., 2015). ROS accumulation is due to cold stress and, subsequently, its oxidative damage to lipid membrane, which in turn can generate toxic products like MDA. Hence, there is a direct relation between chilling

damage and intensity of lipid peroxidation by MDA (Imahori et al., 2008). Possibly, 1-MCP action protects cell membrane by the extension of antioxidant capacity and the decrease in ROS damage.

The level of EL can also increase due to ripening and senescence. Low level of lipid peroxidation and EL in fruit treated by 1-MCP has also been mentioned in peach (Liu et al., 2018) and in persimmon (Zhang et al., 2010; Li et al., 2018; Min et al., 2018). The current results confirm that 1-MCP treatment is associated with higher maintenance of cellular integrity. 1-MCP treatment could suppress disruption of cell membranes by inhibitory effects on ethylene action (Hershkovitz et al., 2005).

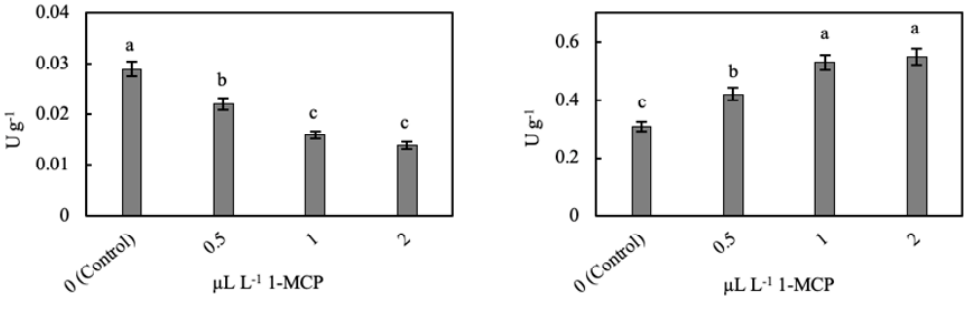
3.9 Peroxidase and catalase activities

Cold stress produces ROS and this consequently leads to oxidative stress. The normal activity of several ROS scavenging enzymes (such as CAT and POD) is required for reducing destructive effects of ROS. Some investigators believed that POD and CAT enzymes have a crucial role on chilling tolerance in persimmon, (e.g., Borrás, 2015; Zhang et al., 2010).

In the current research, CAT and POD activities were evaluated after the storage phase. The activity of POD enzyme in the persimmons subjected to 2 and 1 μL L^{-1} 1-MCP was significantly lower than that level in the control and in those affected by 0.5 μL L^{-1} 1-MCP [Figure 6(a)]. Reduction of POD activity by 1-MCP was also expressed for avocado (Hershkovitz et al., 2005) and for persimmon cv. 'Youhou' (Li et al., 2018).

Maximum CAT activities were seen in the persimmons dealt with 2 and 1 μL L^{-1} 1-MCP, but the control group experienced the minimum CAT activity [(Figure 6(b)]. In agreement with the current results, 1-MCP treatment caused up-regulated CAT and down-regulated POD activities in persimmon fruit cv. 'Fuyu' (Zhang et al., 2010) and cv. 'Rojo Brillante' (Khademi et al., 2014; Borrás, 2015). The reduction in the chilling symptoms in the persimmon treated by 1-MCP is possibly related to ability of these fruit for changing CAT and POD activities (Khademi et al., 2014).

Figure 6 Effects of various 1-MCP concentrations on (a) peroxidase (POD) and (b) catalase (CAT) enzyme activities of persimmon fruit, after 120 d of storage at 0°C



(a) (b)

4 Conclusions

This investigation showed that postharvest 1-MCP treatment, with 1 and 2 μL L^{-1} concentration, can maintain the persimmon fruit quality in the first 30–45 d. Although the fruits were evaluated for 120 d, extension of the storage period beyond 90 days is not considered to be necessary. This is because the control fruit at this stage showed the maximum electrolyte leakage due to severe chilling injury after a shelf-life period. In the persimmon fruit, diminution of some physicochemical properties, such as phenolic compounds, firmness and soluble tannin, were limited using 1-MCP treatment. Further, chilling injuries and chilling tolerance of the fruit in the cold storage phase was reduced by using this treatment. Reduction in chilling symptoms was connected to the reduction of the membrane peroxidation, the enhancement of membrane integrity, and the improvements in the antioxidant capacity. Also, an increase in the chilling tolerance and quality of the treated fruit could be correlated to an increase in the CAT enzyme activities and a reduction in POD enzyme.

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Notes

1 At 35°49′ N 50°59′ E, 1341 m above sea, Alborz Province, Iran.