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Extension of the Postharvest Life of Nectarine Using Modified Atmosphere Packaging and Potassium Permanganate Treatment

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ABSTRACT

The effects of combinations of modified atmosphere packaging (MAP) with potassium permanganate ($KMnO_4$) based ethylene scrubbers on the storage life and fruit quality of nectarine (*Prunus persica* cv. Bayramiç Beyazı) were investigated. Three different types of ethylene sachets (contained 3, 7 and 10 g $KMnO_4$) were used and placed beside fruits in polypropylene baskets then lined with MAP. Fruits were stored at 0-1°C and 90% relative humidity throughout 40 day. During the cooling storage period, O_2 and CO_2 percentage in MAP, fruit firmness, total soluble solids, titratable acidity, ascorbic acid, total flavonoid content, total phenolic content, total antioxidant content and chilling injury (CI) were determined at 10 day interval. $KMnO_4$ treated fruits had shown delayed ripening, reduced respiration and retained of higher firmness. As the dose of $KMnO_4$ treatment increased, it was determined more positive effect on fruit quality. 10 g $KMnO_4$ treatment was most effective in the retention of higher biochemical compounds and inhibition of CI symptoms. The results indicate that $KMnO_4$ treatment, as well as MAP application, should be highly recommended for retaining the fruit quality of cold-stored 'Bayramiç Beyazı' nectarines and fruits treated with 10 g doses of $KMnO_4$ could be stored for 40 days with good quality.

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Introduction

Nectarine is one of the most important and popular fruit consumed worldwide due to its delicious flavour and attractive appearance. Nectarine fruits are usually rich in vitamins and the other antioxidant compounds, including various phenolics (Garcia-Parra et al., 2011). However, they deteriorate rapidly after harvest and usually results in a short limited postharvest life (Özkaya et al., 2016). Cold storage is the most commercially adopted technique, and a cold chain has been integrated into the post-harvest management systems (Lurie and Crisosto, 2005). The storage life of nectarines under ideal conditions of 0°C and high relative humidity (90-95%) is limited to 2-4 weeks (Karen, 1991). When cold temperature combined with modified atmosphere, it causes reducing the produce respiration rate, ripening delay, inhibiting the growth of many spoilage organisms and keeps fruit according to market needs (Kader and Saltveit, 2003). Modified atmosphere is a quite versatile, relatively simple, and low cost technology that can be applied to various types of fruit and vegetables (Silva et al., 2009). Its efficacy can be increased by combining it with ethylene absorbers.

The presence of ethylene in the storage and packing atmosphere has been a major concern for unripe climacteric fruits during postharvest handling, because it accelerates ripening, senescence, abscission and physiological disorders and subsequent postharvest pathogenic infection (Sujayasree and Fasludeen, 2017). The action of ethylene must be avoided during storage and transportation. In peaches and nectarines the greatest increase in ethylene is often after the ripening related changes have already occurred (Lill et al., 1989). The removal of ethylene and/or inhibition of the effect of ethylene in stored environments are fundamental to maintaining postharvest quality of climacteric produce (Saltveit, 1999). There are several compounds that can be used as inhibitors of ethylene, for example aminoethoxyvinylglycine (AVG), an inhibitor of ethylene synthesis; 1-Methylcyclopropene (1-MCP), an inhibitor of ethylene action and potassium permanganate ($KMnO_4$), an oxidizing agent (Sen et al., 2012).

One of the main mechanisms of action of ethylene scavengers is based on the use of $KMnO_4$, which oxidizes ethylene to carbon dioxide and water (Abe and Watada, 1991; Wills and Warton, 2004). Due to the presence of double bond, ethylene ($H_2C=CH_2$) is very reactive and is rapidly oxidized to acetate and ethanol by $KMnO_4$ (Lopez-Rubio et al. 2004). $KMnO_4$ is usually imbedded in different inert substrates such as silica gel incorporated inside high permeable sachets, films or filters (Mehyar and Han, 2011). These products remove unwanted ethylene gas through the oxidation process, thereby ensuring the quality of freshness of the product in a packaged or bulk environment while in transportation or storage. $KMnO_4$ oxidizes ethylene and changes colour from purple to brown, and thus, a colour change indicates its residual ethylene absorbing capacity (Smith et al. 2009). Dual application of $KMnO_4$ and film liner would be highly desirable for maintaining fruit quality during cold storage. The use of $KMnO_4$ in conjunction with modified atmosphere in polyethylene films delayed fruit ripening, maintained quality and extended shelf life in many climacteric fruits such as avocado (Illeperuma and Nikapitiya, 2002), apple (Chaves et al., 2007; Sardabi et al., 2014), mango (Azad et al., 2008; Elzubeir et al., 2017), papaya (Silva et al., 2009; Reboucas et al., 2013), kiwifruit (Bal and Celik, 2010), apricot (Ishaq et al., 2009; Ali et al., 2015), peach (Bal, 2016), tomato (Kostekli et al., 2016) and sapodilla (Freitas et al., 2017). However, there is no information available regarding the effects of $KMnO_4$ treatment and MAP on physiological and biochemical changes in nectarine fruits.

The objective of this work was to evaluate the effect of different doses of $KMnO_4$ on the extension of postharvest life of 'Bayramiç Beyazı' nectarine, stored under modified atmosphere.

Materials and Methods

Plant Materials

Nectarines (*Prunus persica* cv. Bayramiç Beyazı) used in this study were hand-harvested from a local orchard (Çanakkale, Turkey) at firm-ripe stage and immediately transported via ventilated truck to cold storage facilities of Department of Horticulture, Faculty of Agriculture, Namik Kemal University where they were sorted and selected for similar size, uniform maturity and appearance and freedom from defects.

$KMnO_4$ and MAP Treatments

The fruits were distributed among the four treatments in a completely randomized design with three replicates. Three different types of ethylene sachets (Dongguan Dingxing Industry Co., Ltd), contained 3 g ethylene absorber (45×50 mm), 7 g ethylene absorber (45×60 mm) and 10 g ethylene absorber (65×70 mm) were used in the study. The ethylene absorbent was ethylene sachets contained a mixture of natural clays and $KMnO_4$. Sachets were made from Tyvek, a high quality material suitable for contact with food products, which allows gaseous interchanges. $KMnO_4$ sachets were placed beside fruits in polypropylene baskets (1 kg) then lined with MAP. Control fruits packed only with MAP. After all treatments, packages were transferred to a storage room at 0-1°C and 90% relative humidity for 40 days. Nectarine fruits were evaluated by analysing the physicochemical and biochemical attributes using the following parameters at 0 days and at a regular interval of 10 days until the end of storage period.

Assessment of Fruit Quality Attributes

In MAP packages, O_2 and CO_2 gas composition percentages were measured with a checkpoint O_2/CO_2 gas analyzer (Systech Gaspace advance GS3L, England) during storage.

Fruit firmness was determined using a hand-held penetrometer with an 8 mm long measuring plunger on the pared equatorial surface on 2 sides of the fruit and was expressed as Newton (N).

For the analysis of soluble solids content (SSC) and titratable acidity (TA) of each sample, tissue sap was squeezed out from fresh fruit materials with a press. In this juice, SSC were determined with a hand refractometer (%). TA content was determined by titrating method and calculating the result as grams of malic acid per 100 g fresh weight (%).

Ascorbic acid content of the samples was determined according to the recommended method of AOAC (2000) using 2, 6-dichlorophenol indophenol and expressed as mg kg^{-1}.

Total phenolics of the nectarine extract were quantified spectrophotometrically using Folin-Ciocalteu reagent based on the method (Slinkard and Singleton, 1977). Results were expressed as mg (gallic acid equivalent) 100 g^{-1}.

The total flavonoid contents were measured by a colorimetric assay (Zhishen et al., 1999) and the results were expressed as mg (routine equivalent) 100 g^{-1}.

Total antioxidants was determined by 2, 2-diphenyl-1-picrylhydrazyl (DPPH) free radical-scavenging method as described by Brand-Williams et al. (1995) and was expressed as μmol (Trolox equivalent) g^{-1}.

For evaluation of chilling injury, nectarine fruits were longitudinally cut into halves for the evaluation of the occurrence of chilling injury (CI) according to the severity of exocarp browning and flesh translucency (Khan et al., 2011). CI was estimated visually as the percentage of the affected area compared with the total surface area of each section on a scale where: 0 = no change; 1 = less than 10%; 2 = 10-25%; 3 = 25-50%; 4 = 50-75%; and 5 = more than 75%.

Statistical Analyses

The experiment was of a completely randomized factorial design of three replications with two kilogram of fruit per plot. Analysis of Variance (ANOVA) was the means for analysing the difference between means and while LSD test being applied for mean separation at $P<0.05$. All the analyses were carried out through SPSS as statistical software. Results are reflected as the mean ± SE.

Result and Discussion

O_2 and CO_2 Levels in MAP

MAP technology has been successfully used to maintain the postharvest quality and to prolong the storage period of many fruit and vegetables by creating higher CO_2 and lower O_2 concentrations in the surrounding atmosphere of the commodities (Kader and Watkins, 2000). In the study, O_2 and CO_2 levels detected in the sample packages during storage are reported in Figure 1. As expected, a decrease in the headspace O_2

concentration, as well as an increase in the headspace CO_2 concentration, was observed during storage for all of the treatments. Up to 30th day of storage, there was no significant difference in respiration rate in the fruits treated with $KMnO_4$. However, marked difference was recorded in O_2 and CO_2 level at 40th days of storage under all the treatments. At 40th day of storage, in control and 3 g $KMnO_4$ treated fruits, CO_2 level increased more and O_2 level decreased more than other applications. These results indicate that 7 and 10 g $KMnO_4$ doses are more effective than the other two treatments in inhibition of respiration rates during 40 days of storage at 0°C which are in agreement with the previous reports suggesting the beneficial effects of $KMnO_4$ in reducing respiration rates by oxidizing ethylene in avocado (Illeperuma and Nikapitiya, 2002), papaya (Silva et al., 2009) and peach (Bal, 2016). Comparatively higher respiration levels under control and 3 g $KMnO_4$ treated fruits were mainly contributed by two factors namely, acceleration of ripening and occurrence of chilling injury. Chilling injury is known for abrasion of cell membrane and other cell organelles which leads higher cell respiration rate (Valenzuela et al., 2017).

Firmness

Firmness is an important quality attribute in nectarine, and excessive softening is one of the main factors reducing quality and limiting commercialization for fresh consumption (Kader and Mitchell, 1989). In the present study, fruit firmness at harvest was 66.6 N and progressively decreased during storage (Figure 2). Softening of nectarine fruits was remarkably delayed with $KMnO_4$ treatments compared to control. At the end of the storage, the highest firmness was recorded in 10 g $KMnO_4$ treated fruits (55.8 N) followed by 7 g $KMnO_4$ treated fruits (52.9 N) while lowest firmness was recorded in control fruits (39.2 N). According to the results, within doses of $KMnO_4$, lower firmness loss was seen in fruits treated with 3 g $KMnO_4$. It may be due to the increasing in concentration of $KMnO_4$ absorber and decrease in the ripening rate, hence, softening of fruit were minimum. Similar results were obtained by Bal and Celik (2010), Sharma et al. (2012) and Kostekli et al. (2016) who reported that $KMnO_4$ induced ethylene retardation and caused a delay in fruit softening in kiwifruit, plum and tomato respectively.

SSC

Soluble solids content is one of the main internal qualities of nectarine fruits. In the present study, there was significantly an increase in SSC as the fruit ripens in all treatments. Similarly, this increase was reported in nectarine fruits by Akbudak and Eris (2004), Kaynas et al. (2005) and Özdemir et al. (2006). The increase in SSC was mainly due to conversion of starch to soluble forms of sugars. Figure 3 shows the increasing pattern of SSC in nectarine fruit during storage. Higher rises in SSC were observed in control where initial contents (11.7%) increased to 14.4 % followed by 3 g $KMnO_4$ (14%), while 7 g $KMnO_4$ and 10 g $KMnO_4$ maintained a minimum SSC (12.6% and 12.4%) at the 40th day respectively. The reason for delaying the metabolic activity of fruits treated with $KMnO_4$ (especially 7 and 10 g) during storage due to delay in increasing SSC. Maintaining SSC by $KMnO_4$ treatments has been observed in apple (Chaves et al., 2007), papaya (Silva et al., 2009) and mango (Elzubeir et al., 2017).

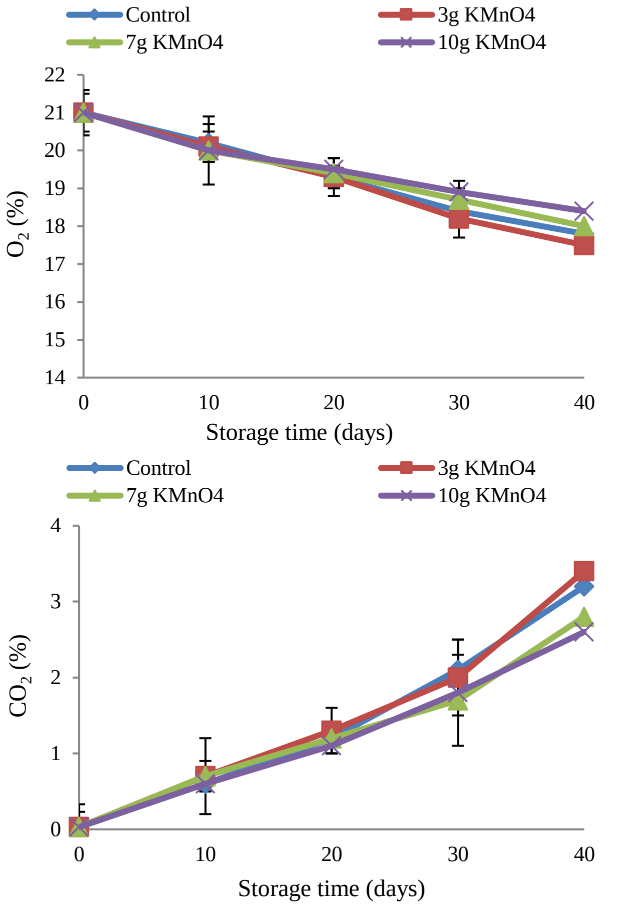

Figure 1 Effects of ethylene absorbent treatments on O_2 and CO_2 concentrations in MAP

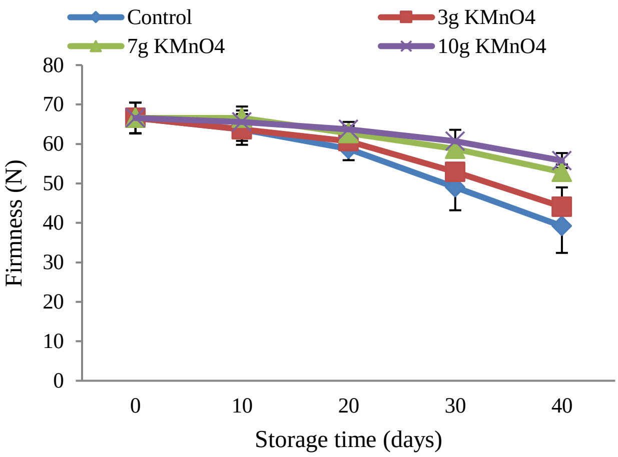

Figure 2 Effects of ethylene absorbent treatments on firmness of nectarine fruits

TA

TA is directly associated with organic acids in fruits, and a reduction in acidity may be expected as a result of metabolic changes in fruit due to the use of organic acids in the respiratory process (Maftoonazad et al., 2008). The content of TA in the nectarine fruit decreased gradually during the storage and this decline was retarded by especially 7 and 10 g $KMnO_4$ treatments (Figure 4).

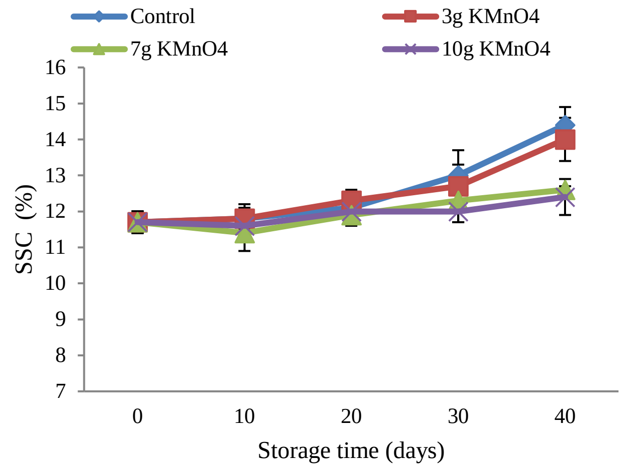

Figure 3 Effects of ethylene absorbent treatments on SSC of nectarine fruits

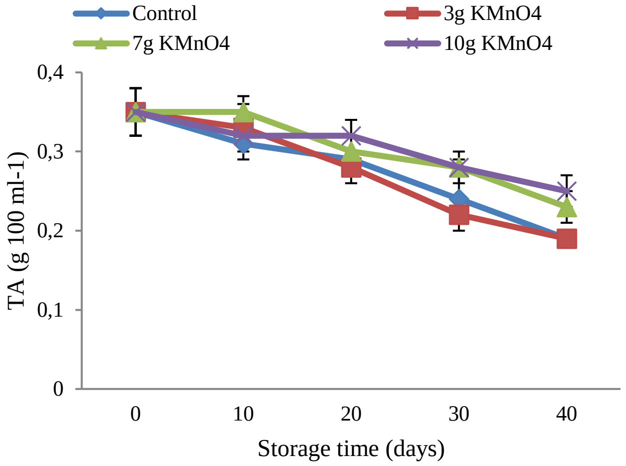

Figure 4 Effects of ethylene absorbent treatments on TA of nectarine fruits

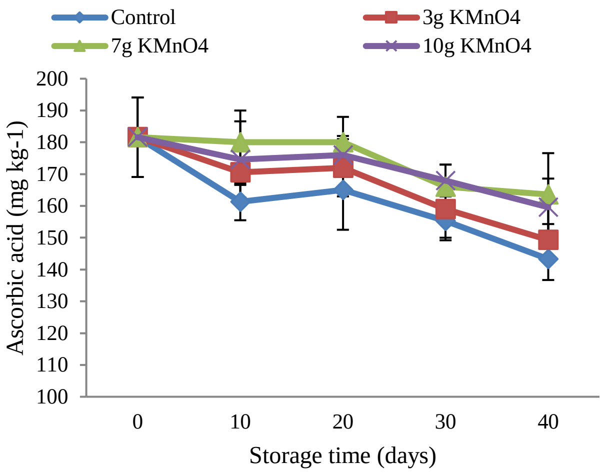

Figure 5. Effects of ethylene absorbent treatments on ascorbic acid of nectarine fruits

The treatment with low levels (3g) of $KMnO_4$ was no effective in delaying the loss of TA. At the beginning of storage, TA of nectarines was 0.35%. At 40th day of storage, while the lowest TA content of nectarines was detected in control (0.19%) and 3 g $KMnO_4$ (0.19%) treatments, the highest TSS content was determined in 7 (0.23%) and 10 g $KMnO_4$ (0.25%) treatments. It was observed that an increase in the permanganate level inside the packing significantly reduced the titratable acidity. The retention of acidity in higher concentrations of $KMnO_4$ might be due to reduction in metabolic changes of organic acid into carbon dioxide and water. Similarly, fruits, such as banana, kiwifruit, tomato and mango, have also been reported to be more acidity when treated with $KMnO_4$ (Elamin and Abu-Goukh, 2009; Bal and Celik, 2010; Kostekli et al. 2016; Elzubeir et al., 2017).

Ascorbic Acid Content

Ascorbic acid is an important nutrient and is highly sensitive to degradation due to its oxidation compared to other nutrients during processing and storage in fruits and vegetables (Rickman et al., 2007). Lee and Kader (2000) reported that the ascorbic acid levels generally tend to decrease during the storage of the most fruits. The data belonging to ascorbic acid content is shown in Figure 5. The ascorbic acid content of the fruit was the highest (181.6 mg kg^{-1}) at the beginning of storage and it decreased with the advancement of storage period. However, this trend was slower in 7 and 10 g $KMnO_4$ treated fruits. At the end of the storage, the highest ascorbic acid content was determined in 7 and 10 g $KMnO_4$ treated fruits (163.6 mg kg^{-1} and 159.6 mg kg^{-1}), while the lowest ascorbic acid content was determined in control fruits (143.3 mg kg^{-1}). Vitamin C is one of the respiratory metabolites, which means it is possible that the faster diminution could cause by the higher respiration rate observed in these treatments. This effect can be associated with the property of $KMnO_4$ on reducing or delaying the activity of ascorbate oxidase and consequently maintaining ascorbic acid content. These results are in line with Reboucas et al. (2013) and Freitas et al. (2017) who had used the ethylene absorbers. Ishaq et al. (2009) also reported that $KMnO_4$ treatment maintained higher ascorbic acid concentrations compared to non-treated apricot fruits due to slower respiration or less oxidation of ascorbic acid in fruit treated with $KMnO_4$.

Total Flavonoid Content

Flavonoids, a special group of polyphenols, are pigments responsible for the color and flavor of many fruits, vegetables, flowers, nuts, and seeds (Harborne and Williams, 2000). In the study, total flavonoid contents of nectarines generally tended to increase during storage even though there were fluctuations in flavonoid content (Figure 6). However, this rise was more pronounced in control fruits depending on maturation. The total flavonoid contents of samples at the beginning of storage were about 83.2 mg 100 g^{-1}. The highest total flavonoid content was obtained from control treatment (102.3 mg 100 g^{-1}) on 30th day, and then sharply decreased to 85.3 mg 100 g^{-1} on 40th day. The results indicated that the total flavonoids of control fruits gradually decreased as maturity proceeded. Whereas, in fruit treated with $KMnO_4$, total flavonoid retained better. 7 and 10 g $KMnO_4$ treated fruits showed to be the most effective treatment for maintaining flavonoid content in fruits. Similarly, Bal (2016) also reported that variations in phenolic compound content and flavonoid content were slower in $KMnO_4$ treated fruits than control group throughout storage period. Slower reduction of biochemical compound in $KMnO_4$ treated fruits than control might be attributed to antisenescence properties of $KMnO_4$ (Abe and Watada, 1991; Kadu and Gajipara, 2009).

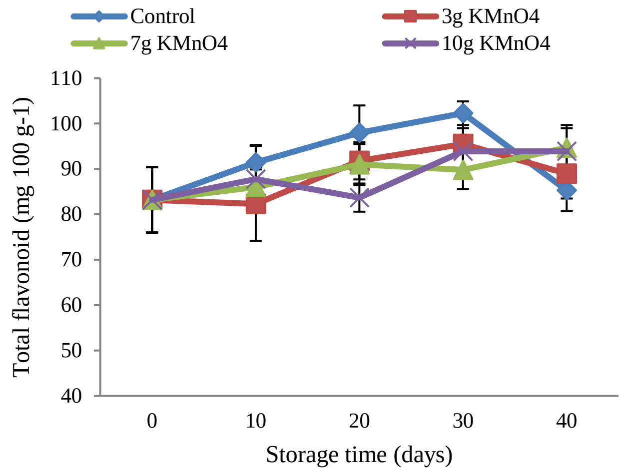

Figure 6 Effects of ethylene absorbent treatments on total flavonoid of nectarine fruits

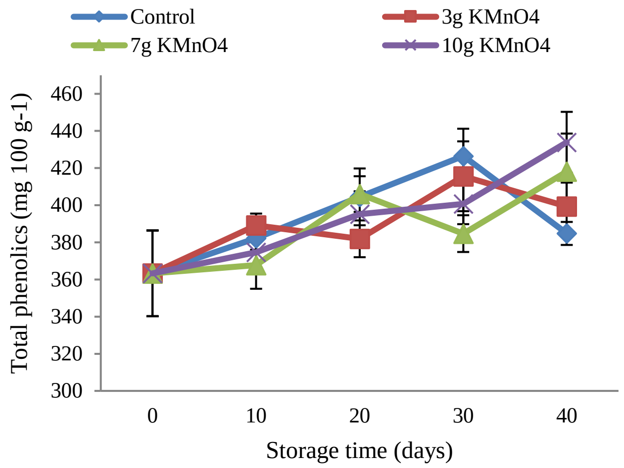

Figure 7 Effects of ethylene absorbent treatments on total phenolic content of nectarine fruits

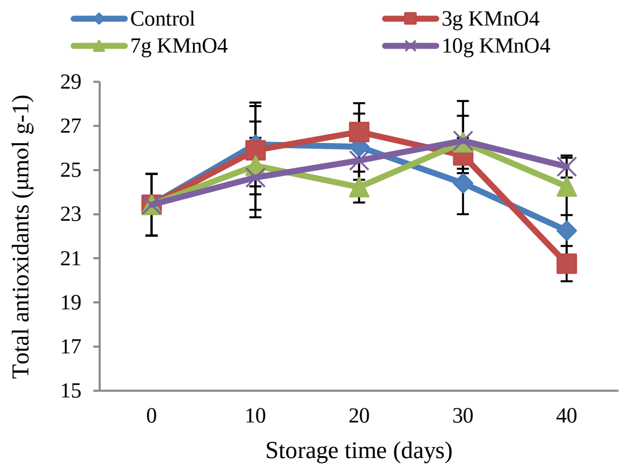

Figure 8 Effects of ethylene absorbent treatments on total antioxidant content of nectarine fruits

Total Phenolic Content

Phenolic compounds are widely distributed in fruit and vegetables and represent, together with ascorbic acid, one of the major contributors to antioxidant capacity (Cefola et al., 2014). Nectarine fruit is also usually rich in vitamins and the other antioxidant compounds, including various phenolics (Garcia-Parra et al., 2011). The initial total phenolic content in the nectarines was 363.3 mg 100 g^{-1} (Figure 7). Changes in total phenolic content over the postharvest period were also similar in trend to those for flavonoid content. Generally, the content of the detected phenols showed an increase in the first 10 days of storage, followed by fluctuations in the next days. However, $KMnO_4$ treatments maintained total phenolic content compared to control at the end of the storage. At 40th day of storage, the highest total phenolic content was recorded in 10 g $KMnO_4$ treated fruits (433.9 mg 100 g^{-1}) followed by 7 g $KMnO_4$ treated fruits (418.1 mg 100 g^{-1}), while lowest total phenolic content was recorded in control fruits (384.8 mg 100 g^{-1}). $KMnO_4$ treatments delayed the onset of the maximum value of total phenol content, and maintained a higher level of total phenol in the fruit at the end of storage. This phenomenon of total phenolics accumulation can be attributed to the role of ethylene in phenolic metabolism (Blankenship and Unrath, 1988). The results were in agreement with previous studies (Ali et al., 2015; Bal, 2016), which found that when ethylene production of fruit was suppressed by ethylene absorbers, the accumulation of total phenolics could be delayed or reduced. Moreover, it is thought that from the thirtieth day, increased progressively chilling injury in control and 3 g $KMnO_4$ treated fruits leaded to a fast decrease in phenolic compounds. Tomas-Barberan et al. (2001) also reported that the chilling injury and browning promoted polyphenol oxidase activities and caused a decreasing of total phenols of fruits.

Total Antioxidants

Antioxidant activity of fruit is contributed by several bioactive compounds like phenolics, flavonoids, ascorbic acid (Wang et al., 1996). The increase in antioxidant content during storage is an indicator of ripening process where phytochemicals attained maximum accumulation. Similarly, a decrease in antioxidant content during subsequent storage is attributed to the oxidation of phenolic contents and ascorbic acid (Ali et al., 2015). In the present study, antioxidant activity was 16.9 µmol g^{-1} at the beginning of the experiment (Figure 8). The results indicated that antioxidant content exhibited an initially increasing trend until to maximum and then began to decrease during subsequent storage. The increase in the antioxidant content could be attributed to the synthesis of some compounds, such as phenols, flavonoids depending on maturation (Zhao et al., 2015). At 40th day, the highest antioxidant content was recorded in 10 g $KMnO_4$ treatment (25.16 µmol g^{-1}) followed by 7 g $KMnO_4$ treatment (24.26 µmol g^{-1}), while lowest antioxidant content was recorded respectively in 3 g $KMnO_4$ (20.76 µmol g^{-1}) and control (22.26 µmol g^{-1}) treatments. 7 g $KMnO_4$ or 10 g $KMnO_4$ treatments effectively postponed the onset of the maximum of antioxidant activity due to retaining maturation period. This indicated that ethylene absorbers were able to maintain higher antioxidant activity in nectarines by delaying the senescence and decay in fruits, as also reported by Ali et al. (2015) and Mir et al. (2018). In addition, the results of this study confirmed that the highest accumulation of antioxidant activities in chilling-tolerant nectarine fruits (7 g and 10 g $KMnO_4$) was associated with resistance to CI. Kostekli et al. (2016) also reported that the percent loss of ascorbic acid and antioxidant capacity was significantly lower in tomatoes stored with ethylene absorber sachets as compared to tomatoes.

Evaluation of CI

CI reduces postharvest quality of fruits and is genetically influenced by a combination of storage temperature and storage period (Crisosto et al., 1999). In this study, compared with the control nectarines, ethylene absorber treatment delayed and reduced the CI in nectarine fruit during the cold storage (Figure 9). All of treated nectarines did not show any CI symptoms till 20 days of storage. However, chilling injury index of the control fruits progressed rapidly after 30 days and got the highest score during other analysis periods. CI symptoms were first visible as exocarp browning and flesh translucency on the 30^{th} day in control and 3 g $KMnO_4$ treatments. At the end of storage, the highest CI value was determined in control fruits (2 point), while no CI symptom was recorded in 10 g $KMnO_4$ treated fruits followed by 7 g (0.3 point) and 3 g $KMnO_4$ (1.6 point) treatments. The lower CI symptoms in nectarines treated with $KMnO_4$ may be due to slower metabolic rates and retention of various bioactive compounds in fruits. This result is in agreement with previous reports showing that use of absorbent sachets can be used to reduce internal breakdown development during storage (Pesis et al., 2002; Bal, 2016). Moreover, CI may be related to tissue deterioration or senescence, which leads to changes in membrane permeability and in biochemical compounds (Lurie and Crisosto, 2005). Therefore, it was seen that 10 g $KMnO_4$ treated fruits which had no CI symptom maintained better firmness and biochemical compounds of fruits.

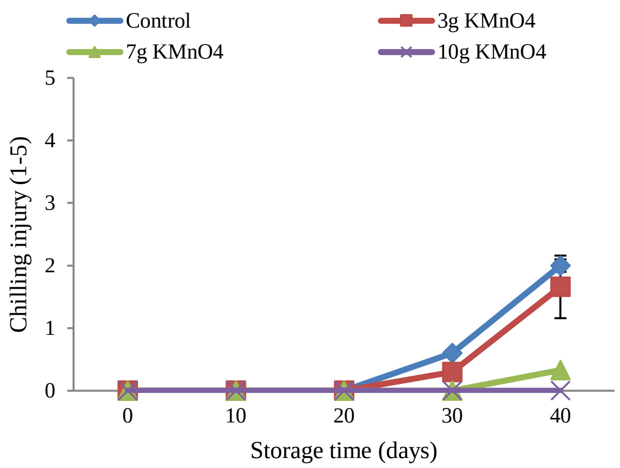

Figure 9 Effects of ethylene absorbent treatments on chilling injury of nectarine fruits

Conclusion

The results of this study indicated that $KMnO_4$ treatment ethylene absorbent along with MAP appeared to maintain fruit quality of nectarine during cold storage. $KMnO_4$ dose influenced the fruit quality, where in the concentration of 10 g of $KMnO_4$ per kg of fruit was the most efficient in inhibition of respiration rates and CI symptoms, retarding the loss of firmness and maintaining biochemical compounds of nectarine fruits. Results showed that 10 g $KMnO_4$ treatment combined with MAP showed promising results for maintaining nectarine quality and extending storage life at 0-1°C for 40 days.

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