

Postharvest AminoethoxyVinylGlycine (AVG) Treatment Affects Maturity and Storage Life of Plum

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ABSTRACT

The aim of this study was to determine the effects of different concentrations (0, 100, 200 and 300 mg L⁻¹) of postharvest AminoethoxyVinylGlycine (AVG) on fruit quality, chilling injury, and bioactive compounds in cold-stored plum fruit (*Prunus salicina* L. cv Friar). Fruit were stored at 0-1°C with 90±5% Relative Humidity (RH) for 60 days. Weight loss, flesh firmness, Soluble Solids Content (SSC), titratable acidity, total anthocyanin content, total phenolic content, antioxidant capacity, respiration rate, and chilling injury were determined at the harvest and during the storage period at 15-day intervals. As compared to the control, AVG treatment delayed ripening and prolonged storage life, as indicated by prevented fruit softening, and retarded the increase in SSC. The 200 and 300 mg L⁻¹ AVG treatments considerably reduced respiration rate and maintained higher bioactive compounds contents than other treatments. The severity of the chilling injury was reduced by AVG treatments compared to the control during storage. The results indicated that postharvest 200 and 300 mg L⁻¹ AVG treatments could be an effective tool for prolonging storage of 'Friar' plums.

Keywords: Anthocyanin, Antioxidant, Chilling injury, *Prunus salicina*, Respiration rate.

INTRODUCTION

Plums have short postharvest life and low temperature storage is recommended for delaying effectively fruit ripening and extending the postharvest life of plums during prolonged market period or long distance transport (Mitchell *et al.*, 1974). Plums can be safely stored at 0°C for 3-5 weeks or more (Crisosto *et al.*, 1999). However, storage potential of plums is influenced by several factors such as cultivar, environmental factors, harvest maturity, storage conditions, and susceptibility to postharvest physiological disorders and diseases (Abdi *et al.*, 1997; Taylor *et al.*, 1995). Therefore, delaying or reducing flesh softening and low temperature deterioration should be important strategies to extend storage life and maintain quality of plum fruit (Wu *et al.*, 2011).

Fruit ripening is a sequence of biochemical processes, which transform a physiologically

mature but inedible fruit into an edible one. Generally, it is known that ethylene plays a key role in inducing ripening processes, especially in climacteric fruits (Streif *et al.*, 2010). Ethylene biosynthesis and production normally lead to reduced storage potential of fruits. For long term cold storage, it is important to reduce ethylene to a minimum level. Recently, a number of approaches for delaying ripening in many fruits have involved inhibiting ethylene production and its action. Notable among the ethylene inhibitors, because of their commercial importance, are AVG and 1-methylcyclopropene (Valdes *et al.*, 2009).

AVG acts as one of the most effective competitive inhibitors in conversion of S-AdenosylMethionine (SAM) to ethylene precursor, that is, AminoCyclopropane-1-Carboxylic acid (ACC). AVG, an organic product and naturally occurring amino acid, is commercially sold under the trade name of

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ReTain® (Greene and Schupp, 2004; Ozturk *et al.*, 2012). Due to its capacity to block the reversibility of ethylene biosynthesis pathway, both pre- and postharvest AVG treatments have been reported as a possible way to delay ripening and to improve the storage potential of various climacteric fruits (Lara, 2013; Siddiqui, 2017). Pre-harvest application of AVG in stone fruits has had a significant effect in reducing ethylene production and softening rate of the fruit (Bregoli *et al.*, 2002; Jobling *et al.*, 2003). Ozturk *et al.* (2012) and Kucuker *et al.* (2015) reported that pre-harvest AVG treatments in plums retarded ripening and slowed down peel color development and fruit flesh softening; decreased SSC values, total phenolics, individual phenolics and total antioxidant activity. However, most of the documented effects of AVG in fruits related to pre-harvest treatment, which may be less target-specific and may require more product than postharvest treatment (Morales-Payan *et al.*, 2009). Postharvest AVG dipping treatment on tomato (Candir *et al.*, 2017), apple (Fadhil and Al-Bamarny, 2010), pear (Wang and Mellenthin, 1977; Andreotti *et al.*, 2004; Tarabih, 2014), apricot (Palou and Crisosto, 2003; Valdes *et al.*, 2009), and peach (Garner *et al.*, 2001) reduced the rate of ethylene production and fruit softening during storage or shelf life period. Presently, there is no data about the effects of postharvest application of AVG on the activity of plum fruit softening and changes in bioactive compounds during storage period.

The objective of this study was to investigate the effect of postharvest dipping of 'Friar' plums with AVG on ripening, bioactive compounds, and maintaining quality during storage.

MATERIALS AND METHODS

AVG Treatment and Storage Condition

The experiment was carried out in the postharvest laboratory of the Department of Horticulture, Agriculture Faculty, Namik Kemal University, Tekirdag, Turkey. Plums

(*Prunus salicina* L.) cv. Friar were harvested at a pre-climacteric stage [Soluble Solids Concentration (SSC)= 12%, Firmness= 45 N] from a commercial orchard and transported immediately to the laboratory (September 1, 2016). Fruit were selected for uniformity of shape, color, and size, and any blemished or diseased fruits were discarded.

The fruits (96 kg) were taken to postharvest laboratory and dipped in an aqueous solutions of AVG (ReTain® Valent Biosciences Corp., USA) at different concentrations: 0 (control), 100, 200 and 300 mg L⁻¹ for 2 minutes. The surfactant Tween 20® (polyoxy ethylene sorbitan monolaurate polyethylene glycol, Sigma-Aldrich Co., USA) at 0.5% was also added to enhance infiltration of the solutions. After treatments, all fruit were dried at 20°C for 3 hours. AVG treated fruits together with controls were placed in polypropylene baskets (2 kg) and then stored at 0-1°C and 90±5% RH for 60 days. Plum fruits were evaluated by analyzing the physico-chemical and biochemical attributes using the following parameters at 0 days (at the harvest) and at a regular interval of 15 days until the end of storage period of 60 days.

Fruit Quality Analysis

Fruit weights were determined using a 0.01 g sensitive digital scale (Radvag WLC/6/A2, Poland). Weight loss during storage was determined by measuring the fruit weight before and after the storage period and was expressed as the percentage of weight loss with respect to the initial weight, and expressed as percent (%).

Flesh firmness was determined using a hand-held penetrometer (Fruit Pressure Tester, FT-327, Facchini SRL, Alfonsine, Italy) with a 7.9 mm long measuring plunger on the pared equatorial surface on 3 sides of the fruit and was expressed as Newton (N).

For the analysis of SSC and Titratable Acidity (TA) of each sample, tissue sap was squeezed out from fresh fruit materials with a press. Nine fruit of each replicate were

used for determination of SSC and TA. In the juice, SSC was determined with a hand refractometer (%) (Atago Pocket Pal-1, Atago Co. Ltd., Tokyo, Japan). TA content was determined by titration method and calculating the result as grams of malic acid per 100 g fresh weight.

Total anthocyanin content was determined by using pH differential method and expressed as milligrams of cyanidin-3-glucoside equivalent per kilogram of fresh weight (mg cyn3-glu 100 g⁻¹) (Wrolstad *et al.*, 2005).

Folin-Ciocalteu reagent method was used to determine the total phenol content of plums as mg gallic acid equivalent 100 g⁻¹ (Slinkard and Singleton, 1977).

The antioxidant activity was evaluated by 2, 2-diphenyl-1-picrylhydrazyl (DPPH) free radical-scavenging method as described by Brand-Williams *et al.* (1995) and was expressed as $\mu\text{mol Trolox Equivalent (TE) g}^{-1}$ fw.

Respiration rate in plums was recorded in the headspace of the container using an auto gas analyzer (Systech Gaspac advance GS3L). The individual fruit was enclosed in a hermetic container of known volume (5000 mL) for 30 minutes and from the headspace gas, concentration of CO₂ was measured by piercing the probe of auto gas analyzer in the container through the septa fixed on the lid of container and direct reading was noted from instrument screen. The CO₂ evolution was calculated in mL of CO₂ kg⁻¹ h⁻¹ by using formula (Demirdoven and Batu, 2004).

For evaluation of Chilling Injury (CI), plum fruit were longitudinally cut into halves 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 damage; 1= Less than 10%; 2= 10-25%; 3= 25-50%; 4= 50-75%; and 5= More than 75%.

Statistical Analysis

The experiment had a completely randomized factorial design and three replications with six kg of fruit per treatment. Analysis Of Variance (ANOVA) was used for analyzing data using the SPSS statistical software. Results are represented as the mean $\pm$ SE.

RESULTS AND DISCUSSION

Weight Loss

Weight loss is an important factor responsible for quantitative as well as qualitative loss of produce leading to shriveling and reduced consumer acceptance (Kader, 2002). The weight loss of plum fruits is shown in (Figure 1-a) and increased with the advancement of storage period irrespective of treatments. However, fruits treated with 200 and 300 mg L⁻¹ AVG treatments showed a lower average (4.8% and 4.4%) in weight loss than the control (5.2%) and 100 mg L⁻¹ AVG (5.4%) treatments at the end of the storage period. Lower weight losses in these treatments may be because these fruits had less active metabolism in respect to respiration and transpiration thus losing lower amount of water during storage in comparison to the control and 100 mg L⁻¹ AVG treated fruits. Karaman *et al.* (2013) and Kucuker *et al.* (2015) also observed lower weight loss in plum fruits during cold storage when they were treated with pre-harvest AVG in different doses. Similarly, Ozturk *et al.* (2017) also indicated that weight loss retarding effect of AVG probably resulted from retarded ethylene synthesis, which consequently retarded ripening.

Respiration Rate

Fruits and vegetables are living commodities and their rate of respiration is of key importance to maintenance of quality (Bal, 2013). Therefore, it is crucial to maintain the respiration rate at a minimum level as much

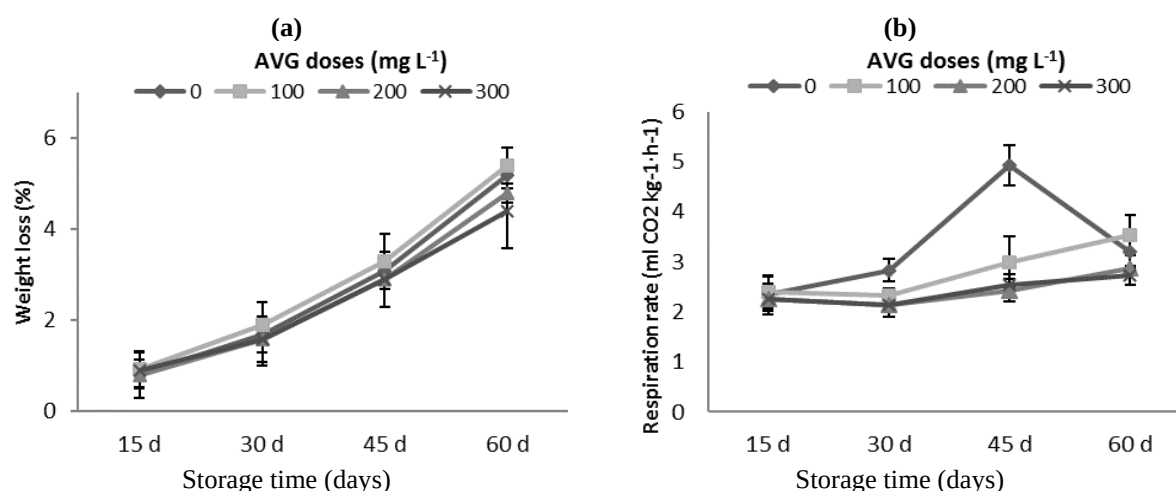


Figure 1. Changes in (a) weight loss and (b) respiration rate, of plum fruits treated with AVG during cold storage. Data are mean $\pm$ SE.

as possible to prolong the storage life of fruit. In general, respiration rates were reduced with AVG treatments in plums. The respiration rate of plum fruit exhibited a typical climacteric pattern during storage (Figure 1-b). The climacteric respiratory peak of the control fruit ($4.9 \text{ mL CO}_2 \text{ kg}^{-1} \text{ h}^{-1}$) was observed on the 45th day, and respiration rate decreased rapidly afterward. At the end of 60 days cold storage, the highest respiration rate was recorded in 100 mg L^{-1} AVG ($3.5 \text{ mL CO}_2 \text{ kg}^{-1} \text{ h}^{-1}$) followed by the control ($3.2 \text{ mL CO}_2 \text{ kg}^{-1} \text{ h}^{-1}$); while the lowest respiration rate was recorded in, respectively, 300 mg L^{-1} AVG ($2.8 \text{ mL CO}_2 \text{ kg}^{-1} \text{ h}^{-1}$) and 200 mg L^{-1} AVG ($2.7 \text{ mL CO}_2 \text{ kg}^{-1} \text{ h}^{-1}$) treatments. Treatment with AVG significantly inhibited the peak value of respiration rate. This suppression in respiration rate by AVG has been reported in other fruits including plum (Wang *et al.*, 2016), peach (Cetinbas and Koyuncu, 2011), apple (Fadhil and Al-Bamarny, 2010), and pear (Tarabih, 2014).

Flesh Firmness

Fruit firmness is one of the most important quality parameters affected by maturity stage, storage time and conditions, and pre- and postharvest applications. In this study, flesh firmness at harvest was 45.9 N, and

significant decrease of firmness occurred during 60 days of storage both in the control and AVG treated plum samples (Figure 2-a). The softening of fruit tissues is thought to be due to the physiological changes of polysaccharide constituents including pectic polysaccharides. Fruits treated with AVG retained firmness significantly better compared with fruits stored without the treatment. At the end of the storage, fruits treated with 300 mg L^{-1} AVG had the highest firmness score (34.3 N), followed by fruits treated with 200 mg L^{-1} AVG (30.8 N). The control fruits had the lowest firmness value after cold storage (21.9 N). These findings are in agreement with Fadhil and Al-Bamarny (2010) and Candir *et al.* (2017), who observed lower fruit softening in apple and tomato fruits, respectively, when they were treated with AVG after harvest. It has been widely reported that AVG prevents or delays fruit softening (Andreotti *et al.*, 2004; Greene and Schupp, 2004; Valdes *et al.*, 2009; Siddiqui, 2017). This is confirmed in the presented work by the higher firmness after cold storage of the fruit treated with AVG. Jobling *et al.* (2003) reported the reason for maintaining of fruit firmness for ‘Tegan Blue’ plums as suppression of ethylene production of fruits by pre-harvest AVG treatments. SSC and TA are two of the most important quality indices for stone fruits (Crisosto *et*

al., 1999). In the present study, fruit SSC increased during the 60-day cold storage in the control or AVG treatments, presumably due to the numerous catabolic processes taking place in fruits during ripening and senescence processes (Kader, 2002). SSC were not affected by doses of AVG treatment until the 45th day (Figure 2-b). However, at 60th day, significant differences were observed between AVG treatments. At the end of storage, 100 mg L⁻¹ AVG (14.6%) and the control treatment (14.3%) had the highest level of SSC while 300 mg L⁻¹ AVG (13.4%) and 200 mg L⁻¹ AVG (13.4%) treatments had the lowest level. It is well known that AVG has a ripening-retarding effect (Greene and Schupp, 2004), thus it might slow down starch conversion into sugar and, consequently, retard increase in SSC values (Ozturk *et al.*, 2017).

TA of plum fruits showed a declining trend during the advancement of storage period, but TA values of AVG-treated fruits were not significantly different from the control fruits (data not shown). In agreement with this study, postharvest AVG dipping treatment did not affect changes in TA content in peach (Garner *et al.*, 2001), apricot (Valdes *et al.*, 2009; Munoz-Robredo *et al.*, 2012), and tomato (Candir *et al.*, 2017) during storage or shelf life period.

Total Anthocyanin Content

Flesh reddening is a kind of senescence or chilling injury symptoms to plum fruit, which is related to the accumulation of anthocyanin in the mesocarp tissue (Crisosto *et al.*, 2004). The occurrence of flesh translucency was often accompanied by the development of flesh reddening in many Japanese plum cultivars, including 'Friar' (Crisosto *et al.*, 1999) and others (Wang *et al.*, 2016). In the present work, total anthocyanin content at harvest was 15.5 mg 100 g⁻¹ and progressively increased during storage (Figure 3-a). There was no significant increase in total anthocyanin content until the 30th day. However, at 45th day, flesh reddening began to occur in the mesocarp tissue and anthocyanins rapidly accumulated, especially in the control fruit. At the end of storage, the control fruits had the highest total anthocyanin content (39.8 mg 100 g⁻¹), while plum fruit treated with 300 mg L⁻¹ AVG (19.9 mg 100 g⁻¹) had the lowest total anthocyanin content followed by 200 mg L⁻¹ AVG treatment (21.3 mg 100 g⁻¹). It was observed that AVG treated plum fruit had much lower content of anthocyanins, compared to the control fruit. The inhibition of anthocyanin accumulation by AVG treatment may be consistent with the retard of fruit reddening and chilling

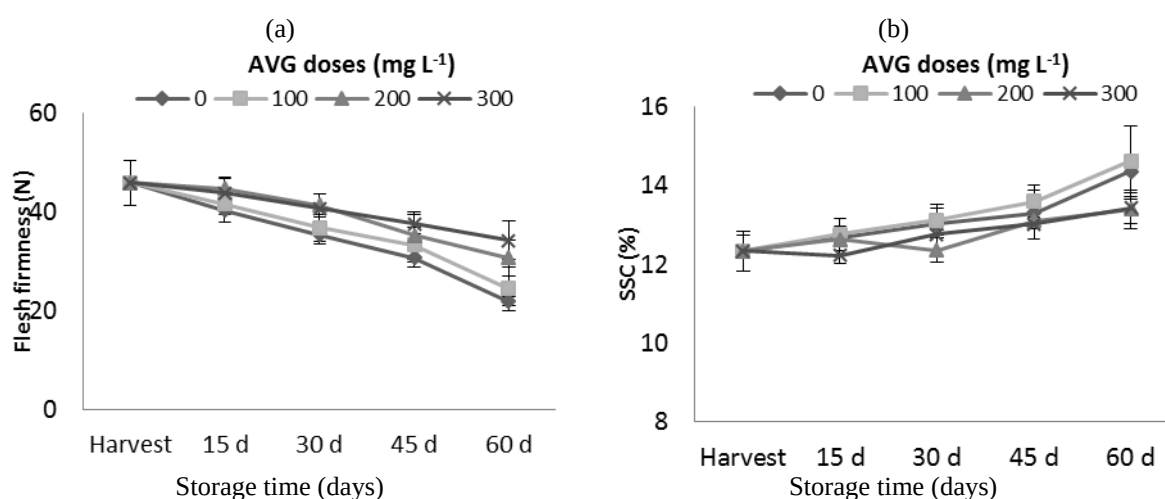


Figure 2. Changes in (a) flesh firmness (b) SSC, of plum fruits treated with AVG during cold storage. Data are mean $\pm$ SE.



injury symptoms. These results are in agreement with Ozturk *et al.* (2012) and Ozturk *et al.* (2013), who reported that pre-harvest AVG treatments in plums and sweet cherries delayed the postharvest ripening process and slowed down the anthocyanin synthesis.

Total Phenol Content

Plums and prunes are rich sources of phenolic compounds, many of them being concentrated in the exocarp (Raynal *et al.*, 1989). Changes in chemical composition of fruits including phenolic and antioxidant compounds vary based on the variety, growth period, cultural practices, ripening levels of fruits, time of harvest, post-harvest storage conditions, and postharvest fruit processing methods. (Kucuker and Ozturk, 2014). Generally, phenolic content in fruit increases after harvest for a certain period then decreases toward to the end of storage period. (Figure 3-b) shows the phenolic composition of plum fruits and changes during storage period. The total phenol content of samples at the beginning of storage was about 259.1 mg 100 g⁻¹. For all treatments, phenolic compounds tended to increase until day 45, but this increase was more pronounced in the control and 100 mg

L⁻¹ AVG treatments. These results confirmed that 200 mg L⁻¹ and 300 mg L⁻¹ AVG treatments reduced the accumulation of phenolic content. Similarly, Karaman *et al.* (2013) reported that increasing AVG doses were observed to bring about decreasing total phenolic and antioxidant activity. At the end of the storage, the increases in 300 mg L⁻¹ AVG (299.5 mg 100 g⁻¹) and 200 mg L⁻¹ AVG (286.4 mg 100 g⁻¹) treatments continued, but there was a sharp decline in the control (265.8 mg 100 g⁻¹) and 100 mg L⁻¹ AVG (279.5 mg 100 g⁻¹) treatments. It is thought that the progressively increased chilling injury in the control and 100 mg L⁻¹ AVG treated fruits led to a fast decrease in phenolic compounds. This result is in agreement with the report of Graham and Paterson (1982) and Martinez and Whitaker (1995) that the chilling injury promoted polyphenol oxidase activities and caused a decrease in total phenols of fruits.

Antioxidant Activity

Plums are known for their high content of phytonutrients and are considered a rich source of natural antioxidants necessary in our daily nutrition. (Valero and Serrano, 2010). According to literature, the

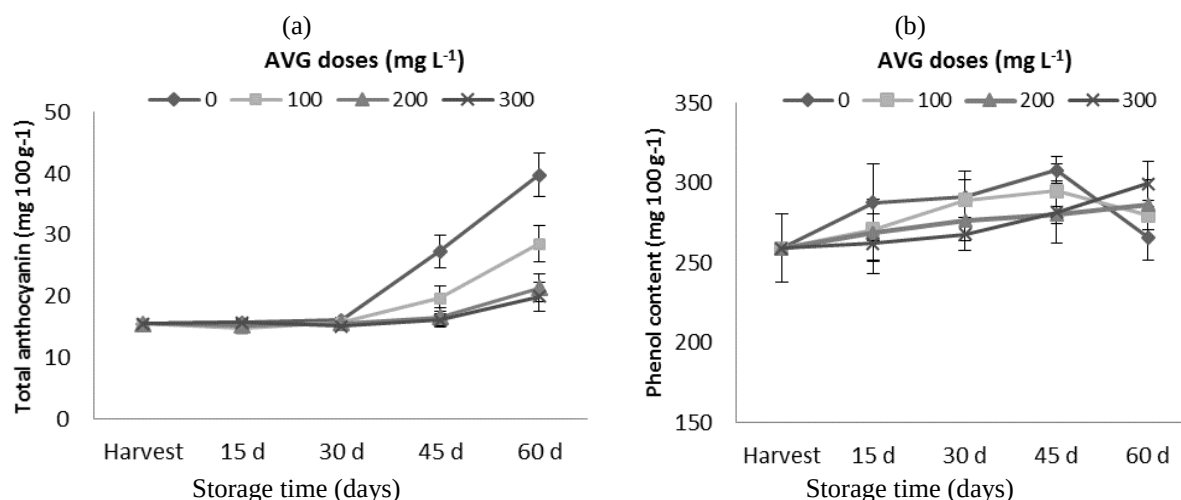


Figure 3. Changes in (a) total anthocyanin content (b) total phenol content, of plum fruits treated with AVG during cold storage. Data are mean $\pm$ SE.

antioxidant activity of plums tends to increase during cold storage and/or ripening (Kevers *et al.*, 2007; Karaman *et al.*, 2013). Similarly, in the present study, increases as well as fluctuations were determined in antioxidant activity of all treatments. At the beginning of the experiment, antioxidant activity was $16.9 \mu\text{mol g}^{-1}$ (Figure 4-a). While the highest antioxidant activity was obtained in the control treatment ($20.56 \mu\text{mol g}^{-1}$) on the 45th day, the lowest value was obtained in 100 mg L^{-1} AVG treatment ($16.2 \mu\text{mol g}^{-1}$) on the 15th day. However, on the 60th day, the decrease in the control ($17.5 \mu\text{mol g}^{-1}$) and 100 mg L^{-1} AVG ($18.1 \mu\text{mol g}^{-1}$) treatments was determined, while the 200 mg L^{-1} AVG ($19.7 \mu\text{mol g}^{-1}$) and 300 mg L^{-1} AVG ($18.9 \mu\text{mol g}^{-1}$) treatments continued to increase. At the end of storage, the increased chilling injury rate, especially in the control and 100 mg L^{-1} AVG treated plums, also reduced the antioxidant activity. The breakdown of cell structure in chilled fruits could cause the decrease in antioxidant content of the fruit during storage. The same trend was observed for total phenol content of plums. Several studies have also shown significant contributions of phenolics content to fruit antioxidant capacity and high correlations between phenolics content and antioxidant activity (Gil *et al.*, 2002; Ozturk

et al., 2015; Candir *et al.*, 2017). Wang *et al.* (2005) also reported that increased antioxidant activities and reduced active oxygen species accumulation might contribute to the delayed occurrence of chilling injury in peaches during storage.

Chilling Injury

Chilling injury refers to a syndrome that involves several physiological events, as well as the characteristic and recognizable symptoms of cold-stored fruit (Valenzuela *et al.*, 2017). Plums are susceptible to chilling injury when stored at low temperature after harvest (Crisosto *et al.*, 1999). In this study, it was observed that, irrespective of the treatments, plums did not show any chilling injury symptoms till 30 days of storage (Figure 4-b). However, chilling injury index of the control fruits progressed rapidly after 30 days and got the highest score during other analysis periods. Chilling injury symptoms in fruits were first visible as exocarp browning and flesh translucency on the 45th day for all treatments. The severity of the chilling injury incidence was reduced by AVG compared to the control treatment during storage. At the end of storage, the highest chilling injury was determined in the

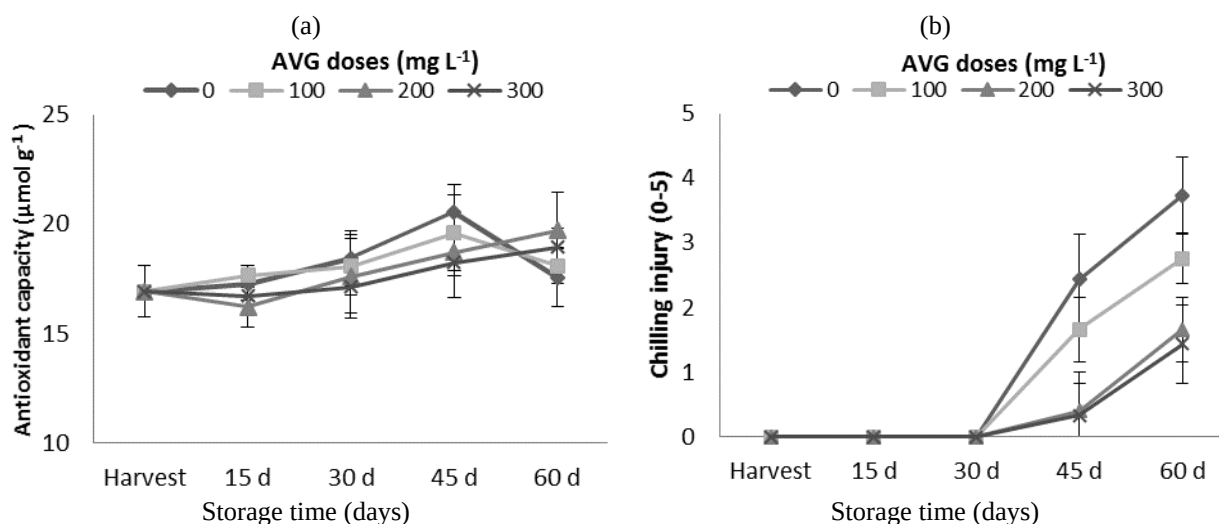


Figure 4. Changes in (a) antioxidant capacity (b) chilling injury, of plum fruits treated with AVG during cold storage. Data are mean $\pm$ SE. (0= No change; 1= Less than 10%; 2= 10-25%; 3= 25-50%; 4= 50-75%; and 5= More than 75%).



control fruits (3.7 point), while the lowest chilling injury was recorded in, respectively, 300 mg L⁻¹ AVG (1.4 point), 200 mg L⁻¹ AVG (1.6 point), and 100 mg L⁻¹ AVG (2.7 point) treatments. Chilling stress has been known to trigger the antioxidant response in fruits (Zhao *et al.*, 2009). Moreover, the lower chilling injury symptoms in plums treated with AVG may be due to slower metabolic rates and retention of various bioactive compounds in fruits. Similar results were also reported by Tavallali and Moghadam (2015) and McGlasson *et al.* (2005), who found enhanced tolerance to chilling injury by AVG treatments in mandarin and nectarine fruits. Although many trials have been carried out to study the relationship between ethylene production and ripening in plums, its role in CI development is still unknown (Candan *et al.*, 2008).

CONCLUSIONS

In conclusion, the results of the study demonstrate that postharvest treatment of AVG has potential to delay the fruit ripening in 'Friar' plum fruits. Treatments of 300 and 200 mg L⁻¹ AVG to plum fruit could be considered as a postharvest tool with good results in terms of maintenance and delay loss of flesh firmness, total anthocyanin, and total phenolic content. AVG considerably inhibited flesh reddening and the peak value of respiration rate. In addition, AVG markedly reduced the incidence of chilling injury and maintained significantly higher antioxidant activity during cold storage. The results of this study showed that postharvest AVG treatment had promising results for maintaining the quality of 'Friar' plum and extending storage life at 0-1°C and 90±5% relative humidity for 60 days.

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تیمار بعد از برداشت با آمینواتوکسی وینیل گلیسین (VAG) بر مرحله رسیدن و دوران انبارداری آلو تاثیر میگذارد

۱. بال

چکیده

هدف این پژوهش تعیین اثر تیمار کردن با غلظت های مختلف (۰، ۲۰۰، ۱۰۰۰، و ۳۰۰۰ میلیگرم در لیتر) آمینواتوکسیوینوین گلیسین (AVG) بعد از برداشت روی کیفیت میوه، صدمات سرمازدگی، و مواد بیواکتیو (زیست فعال) در آلو (*Prunus salicina* L. cv. Friar) نگهداری شده در سردخانه بود. میوه ها به مدت ۶۰ روز در حرارت $1-0^{\circ}\text{C}$ و نم نسبی $90 \pm 5\%$ انبار شد. کم شدن وزن، سفتی گوشت میوه، محتوای جامدات محلول (SSC)، اسیدیته قابل تیتراسیون، محتوای آنتوسیانین کل، محتوای فنل کل، ظرفیت آنتی اکسیدانی، نرخ تنفس، و صدمات سرمازدگی در مرحله برداشت و در طی دوره انبارداری در فواصل ۱۵ روزه تعیین شد. در مقایسه با شاهد و بر اساس جلوگیری از نرم شدن میوه، تیمار با AVG منجر به تاخیر در رسیدن محصول و طولانی شدن دوره انبارداری شد و افزایش SSC را به تعویق انداخت. تیمارهای ۲۰۰ و ۳۰۰ میلیگرم در لیتر AVG نرخ تنفس را به طور چشمگیری کاهش داد و مقدار مواد بیواکتیو را بیشتر از تیمارهای دیگر نگهداشت. در طول انبارداری، شدت صدمات سرمازدگی در تیمارهای AVG در مقایسه با شاهد کاهش یافت. نتایج چنین حکایت داشت که تیمارهای ۲۰۰ و ۳۰۰ میلیگرم در لیتر AVG بعد از برداشت می تواند برای آلو رقم Fariar اقدام موثری در طولانی کردن انبارداری باشد.