

## The effect of carboxymethyl cellulose and pistachio (*Pistacia atlantica* L.) essential oil coating on fruit quality of cold-stored grape cv. Rasheh

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### ABSTRACT

Application of natural compounds such as essential oils and plant materials as bio-products is known as an appropriately safe strategy for controlling decay and maintaining fruit quality. *Pistacia atlantica* essential oil (PAO) (ranging between 0, 200, 400, 600, 800, 1000 and 1200  $\mu\text{l l}^{-1}$ ) was tested for antifungal activity against *Botrytis cinerea* *in vitro* conditions. Carboxymethyl cellulose (CMC) (0, 1 and 2%) and PAO (0, 600 and 1200  $\mu\text{l l}^{-1}$ ) were applied to the "Rasheh" grape for improving grape cold storage to preserve fresh fruit quality during cold stored postharvest. The growth of *B. cinerea* mycelia was greatly inhibited up to 84% at 1200  $\mu\text{l l}^{-1}$  PAO under *in vitro* conditions. CMC and PAO treatments led to delaying grape weight loss and fruit decay. Titratable acidity, anthocyanin, antioxidant capacity, phenol and tannin of uncoated fruits decreased and total soluble solid contents increased during the grape storage. Fruit coating with CMC and PAO maintained higher anthocyanin, antioxidant capacity, phenol, tannin and titratable acidity. Less increase of total soluble solids was observed in coated fruits when compared to control fruits. The results indicated the beneficial effect of CMC (2%) and PAO (1200  $\mu\text{l l}^{-1}$ ) coating on maintaining of grape fruit quality 28 d after storage.

**Keywords:** Anthocyanin, antioxidant activity, decay percentage, pistachio essential oil, weight losses.

## اثر کربوکسی متیل سلولوز و اسانس بنه به عنوان پوشش خوراکی بر کیفیت میوه انگور رقم رشه نگهداری شده در انبار سرد

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### چکیده

کاربرد ترکیبات طبیعی مانند اسانس‌ها و مواد گیاهی به‌عنوان ترکیبات زیستی به صورت یک راهکار مناسب و ایمن برای کنترل پوسیدگی و حفظ کیفیت میوه شناخته شده است. اسانس *Pistacia atlantica* (PAO) (۰، ۲۰۰، ۴۰۰، ۶۰۰، ۸۰۰، ۱۰۰۰، ۱۲۰۰ میکرولیتر در لیتر) برای بررسی خاصیت ضد قارچ *Botrytis cinerea* در شرایط درون شیشه‌ای مورد آزمایش قرار گرفت. کربوکسی متیل سلولوز (CMC) (۰، ۱ و ۲ درصد) و PAO (۰، ۶۰۰، ۱۲۰۰ میکرولیتر در لیتر) بر روی انگور رقم رشه به منظور حفظ کیفیت میوه در طول دوره نگهداری در انبار سرد، به کار برده شدند. رشد میسلیم‌های *Botrytis cinerea* به میزان زیادی تا ۸۰٪ در ۱۲۰۰ میکرولیتر در لیتر PAO در شرایط درون شیشه‌ای محدود گردید. کاربرد CMC و PAO منجر به تاخیر در کاهش وزن و پوسیدگی میوه گردید. اسیدیتته قابل تیتراسیون، میزان آنتوسیانین، ظرفیت ضد اکسایشی، فنل و تانن میوه‌های پوشش داده نشده در طول دوره انبارداری کاهش یافتند و مواد جامد محلول کل افزایش یافت. پوشش دادن میوه‌ها با CMC و PAO مقدار بالاتری آنتوسیانین، ظرفیت ضد اکسایشی، فنل، تانن و اسیدیتته قابل تیتراسیون را در طول دوره انبارداری حفظ نمود. افزایش کمی در مواد جامد محلول در میوه‌های پوشش داده شده در مقایسه با میوه‌های پوشش داده نشده مشاهده گردید. نتایج اثر مفید (۲٪) CMC و (۱۲۰۰ میکرولیتر در لیتر) PAO بر حفظ کیفیت میوه انگور رقم رشه بعد از ۲۸ روز نگهداری در انبار سرد را نشان داد.

**واژه‌های کلیدی:** آنتوسیانین، اسانس بنه، درصد پوسیدگی، فعالیت ضد اکسایشی، کاهش وزن.

## Introduction

For decades, fruit postharvest management aim of maintaining of fruit quality at the highest level has been a challenge. Fruit quality is normally at a higher level in fully ripe fruits, while fruit shelf and storage life after the harvest is usually expanded when fruits are less mature or unripe at the harvest time (Toivonen, 2007). However, fruits which are commercially harvested at the commercial ripening stage (not over-ripe or senescent) show many disorderliness like fungal decays and physiological deterioration leading to a decrease in shelf life. Grape is considered as an important source of nutritional antioxidant compounds like phenolic compounds such as anthocyanins, tannins, and flavonoids (Baiano and Terracone, 2011; Lutz *et al.*, 2011). The table grape quality decreases during decreases during post-harvest storage because of dehydration, loss of weight, tissue loosening, browning of rachis, off-flavor and abnormal aromas in over-ripe fruits and high incidence of berry decay (Elmer and Reglinski, 2006). Moreover, berry fungal decay mainly caused by *Botrytis cinerea* fungi, could commercially reduce fruit quality and fruit consumption (Chervin *et al.*, 2009). Gray mold often occurs in the field at the flowering stage and consequently postharvest decay might occur by remaining conidia on the berry surfaces (Crisosto *et al.*, 2002; Guillén *et al.*, 2007).

Based on recent restrictions in using synthetic fungicides, they should be replaced by available natural alternatives such as plant based essential oils like natural oils of *Eucalyptus* and *Cinnamon* species. There are different reports which show the activity of these natural-based oils versus a wide range of plant pathogens (Ramezani *et al.*, 2002; Serrano *et al.*, 2005). Some phenolic compounds are

the main part of essential oils of plant origin with strong potential to inhibit microbial pathogens (Antunes and Cavavo, 2010). Postharvest application of *Thymus vulgaris* and *Carum copticum* oils on inoculated tomato fruit by *Penicillium digitatum* and *Alternaria alternata* have been studied (Abdolahi *et al.*, 2010). Thyme oil is used for control of *B. cinerea*, *Rhizopus stolonifer*, *Alternaria alternata* (Plotto *et al.*, 2003) and *C. gloeosporioides* (Sellamuthu *et al.*, 2013) during *in vitro* tests. *In vitro* tests exhibited that cinnamon oil effectively controlled *C. gloeosporioides* in banana (Maqbool *et al.*, 2010). Another report of an *in vitro* experimental study showed *P. atlantica* methanol extracts to be the best inhibitor against the mycelial growth of *Geotrichum candidum* (Talibi *et al.*, 2012).

Recently, different organic compounds including wax, celluloses, lipids, starch and alginate have mainly been utilized as edible coatings to properly stop fruit weight loss in storage (Cha and Chinnan, 2004). Carboxymethyl cellulose (CMC) is one of the most momentous water-soluble cellulose origin, with a great number of uses in the food, cosmetics, pharmaceuticals and detergents (Togrul and Arslan, 2003). Coating with alginate and carboxymethyl cellulose is used for preservation of grapes during storage (Yinzhe and Shaoying, 2013). Moreover, fruit edible coatings by polysaccharide materials such as carboxymethyl cellulose have been evaluated for their effects on fruit quality based on delaying fruit ripening, senescence and decay (Maftoonazad *et al.*, 2008). Hussain *et al.* (2015) recommended CMC and irradiation as a suitable combination for maintaining plum quality and delaying the fruit decay during storage.

Essential oil of *Pistacia atlantica*

*Sub sp. Kurdica* which also has antifungal properties might be effective in declining fruit decay (Hesami *et al.*, 2013). Alpha-pinene as main compound of *Pistacia atlantica* essential oil show antifungal activities (Xia *et al.*, 1999; Delazar *et al.*, 2004). Application of alpha-pinene can clearly destroy fungal cell wall and cytoplasm and alter the fungal morphology. Moreover, DNA and RNA synthesis, cell wall polysaccharide and ergosterol of cytoplasmic membrane is inhibited (Xia *et al.*, 1999). However, thus far there has been no reports on the effect of postharvest *Pistachia atlantica* essential oil (PAO) treatments in fruit quality preservation.

"Rasheh" is a main grape cultivar grown in rain fed regions in the west of Iran. Therefore, the objective of the present experiment was to determine the efficiency of PAO *in vitro* and PAO and CMC *in vivo* in controlling "Rasheh" postharvest decay and impact on the characteristics of postharvest quality. Our results might introduce this product as an interesting and innovative commercial product as well as a substitute to the synthetic postharvest fungicides.

## Materials and Methods

### Plant materials and metabolite extraction

Organically grown table grape (*Vitis vinifera* cv. Rasheh) at the ripe stage was harvested in Negel in Kurdistan province, Iran and instantly transferred to the laboratory followed by selection of grape bunches with uniform shape, size, color, and no defects. *P. atlantica* essential oil (PAO) was extracted from 100g of gum material through water-distillation with a Clevenger-type (Schot duran, Germany) apparatus for three hours. Carboxymethyl cellulose (food grade) (Sigma-Aldrich Co., USA) was Solved in sterilized double-distilled water by stirring at 80°C to acquire a final concentration of up to

1% and 2%. Carboxymethyl cellulose (CMC) is a cellulose derivative with carboxymethyl groups that has been synthesized by the alkali-catalyzed reaction of cellulose with chloroacetic acid (Baar & Kulicke, 1994).

### *In vitro* antifungal assay

Potato dextrose agar (PDA) medium was used for *in vitro* antifungal assay. Cold prepared medium was placed into the slopes on which seven different PAO concentrations: 0 (control), 200, 400, 600, 800, 1000 and 1200  $\mu\text{l l}^{-1}$  were added. After setting, 5 mm agar disk from *B. cinerea* a pure culture was placed in the PDA center plate containing PAO. Control plates included only PDA. Petri plates were then incubated for 4 d at 23 °C. For each oil concentration five replicates were prepared. *B. cinerea* growth was calculated by determining of two diameters of each colony and PAO prevention capacity was obtained as the percentage of radial inhibition of growth compared to the no treated petri plate (Nordin *et al.*, 2013; Tanovic *et al.*, 2015).

### *In vivo* assay

PAO and CMC were selected to examine their effects on rot under cold storage. Grape bunches (300 g) were dipped for 2-3 min in 600 and 1200  $\mu\text{l l}^{-1}$  PAO, 1% and 2% (w/v) CMC and sterile distilled water as a control. Subsequently, they were dried at room temperature with natural convection for 2-3 h, then stored in holed polyethylene terephalate trays under cold storage for periodic evaluation of physicochemical parameters (temperature  $4\pm 1^\circ\text{C}$ , RH85-90%). Three different bunches for each time/treatment were determined for different characteristics as explained below, at different cold storage periods at 1, 7, 14, 21 and 28 days. After each storage time, all stored samples were placed at room

temperature before the measurement of qualitative characteristics to simulate market process.

#### **Weight loss and rotted berries**

Coated and uncoated grape bunches weight loss were determined at different storage periods. Weight losses were stated as a percentage loss of the initial weight (storage time=0). The difference as a percentage from the original weight was calculated ( $\text{weight loss\%} = [\text{initial weight} - \text{weight at examined date} / \text{initial weight}] \times 100$ ). Naturally occurred rot was evaluated and grape berries that were infected with gray mold were identified and expressed as the percentage of rotted berries. Fruit-rot was estimated based on the following formula:  $\text{Incidence of fruit-rot} = (\text{Number of berries exhibiting rot} / \text{total number of investigated berries}) \times 100$  (Castillo *et al.*, 2010; Youssef & Roberto, 2014).

#### **Titrateable Acidity, pH and Total Soluble Solids**

Berries were selected from each replication and the fruit juice of 10 berries for each repetition was used to measuring total soluble solids (TSS) and titrateable acidity (TA). Fruits were homogenized and the resultant pulp was filtered. The pH of the fruit juice was measured using a digital pH meter. For measurement of TA, 10 mL of fruit juice was titrated against 0.1 N NaOH up to pH 8.2. The results were stated as % of tartaric acid. TSS was measured for each group with a digital refractometer at 20°C.

#### **Determination of Antioxidant capacity**

Activity of antioxidant was evaluated using assessment of samples free radical scavenging activity with stable radical 2,2-diphenyl-1-picrylhydrazyl (DPPH), as described by Sanchez-Moreno *et al.* (2003) and modified by Sanchez-Gonzalez *et al.*, (2011) for grape.

#### **Determination of Total Anthocyanin**

Amount of total anthocyanin was measured with pH-differential method as described by Mónica, and Wrolstad (2001). The attained grape extracts in methanol were diluted with buffer to get an absorbance at the linear range of the spectrophotometer. The diluted grape extracts pH values were 1.0 (0.025 M potassium chloride buffer) and 4.5 (0.4 M sodium acetate buffer), respectively. Absorbance was read using a spectrophotometer (S2100 SUV, U.S.A) at 520 and 700 nm. Total anthocyanin pigment (mg/kg) was determined as milligram of malvidin-3-O-glu per 1000 g fruit.

#### **Phenol analysis**

Grape total phenolic content was measured with Folin-Ciocalteu reagent as described by Slinkard and Singleton (1977) and gallic acid used as standard phenolic compound for drawing of standard curve. The absorbance was measured at 760 nm by spectrophotometer (Beckman UV-DU 520, USA).

#### **Statistical analysis**

Data for all parameters were subjected to the analysis of variance (ANOVA). Mean comparisons were carried out using LSD at  $P < 0.05$ . SAS software (Version 9.1) was used for all analyses.

### **Results and discussion**

#### **In vitro and in vivo antifungal assay**

The inhibition of fungal expansion, as a function of PAO concentration is shown in Figs 1 and 2. In comparison to control treatment, the growth of fungal mycelia (*B. cinerea* L.) was significantly ( $P = 0.05$ ) decreased, as a result of PAO application. *In vitro* inhibition was directly related to the concentrations of PAO. PAO 1200  $\mu\text{l l}^{-1}$  exhibited strong antifungal activity against *B. cinerea*, with respective inhibition values of 84%

by measuring the percentage of radial growth inhibition of molds in PDA medium (Figure 1). The PAO concentrations 200, 400, 600, 800 and 1000  $\mu\text{l l}^{-1}$  also significantly suppressed the *B. cinerea* mycelia growth compared to the control but was not as effective as 1200  $\mu\text{l l}^{-1}$ . *Pistacia Atlantica* essential oil showed antifungal activity against grey mold of *B. cinerea* (Hesami *et al.*, 2013). As Talibi *et al.* (2012) reported the methanol extract of *Pistacia atlantica*

resulted in a higher antifungal activity against *Geotrichum candidum*. The function and antimicrobial mechanism of essential oils has been characterized as the disruption of cellular membrane function and interferes with active sites of enzymes and cellular metabolism (Marino *et al.*, 2001). Essential oils may change the permeability of microbial membranes for captions and altered ion gradients lead to the impairment of vital cell processes and eventually cell death (Ultee *et al.*, 1999).

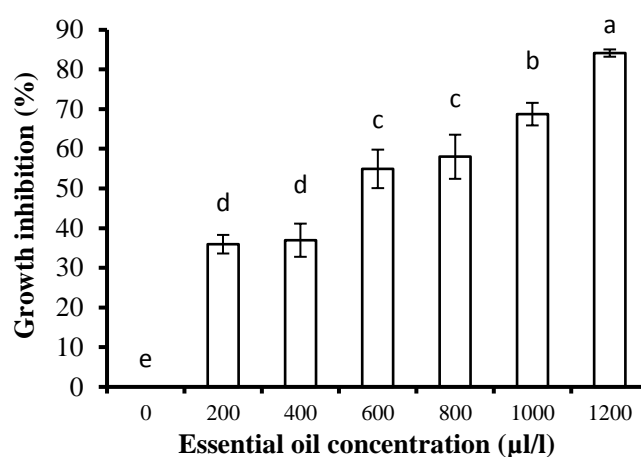


Figure 1. The effect of pistachio (*Pistacia atlantica* L.) essential oil ( $\mu\text{l l}^{-1}$ ) on percent of inhibition *Botrytis cinerea* inhibition (percentage) culture. Different letters stand for statistically significant differences at  $P < 0.05$ . Bars in each mean symbol indicate the standard error.

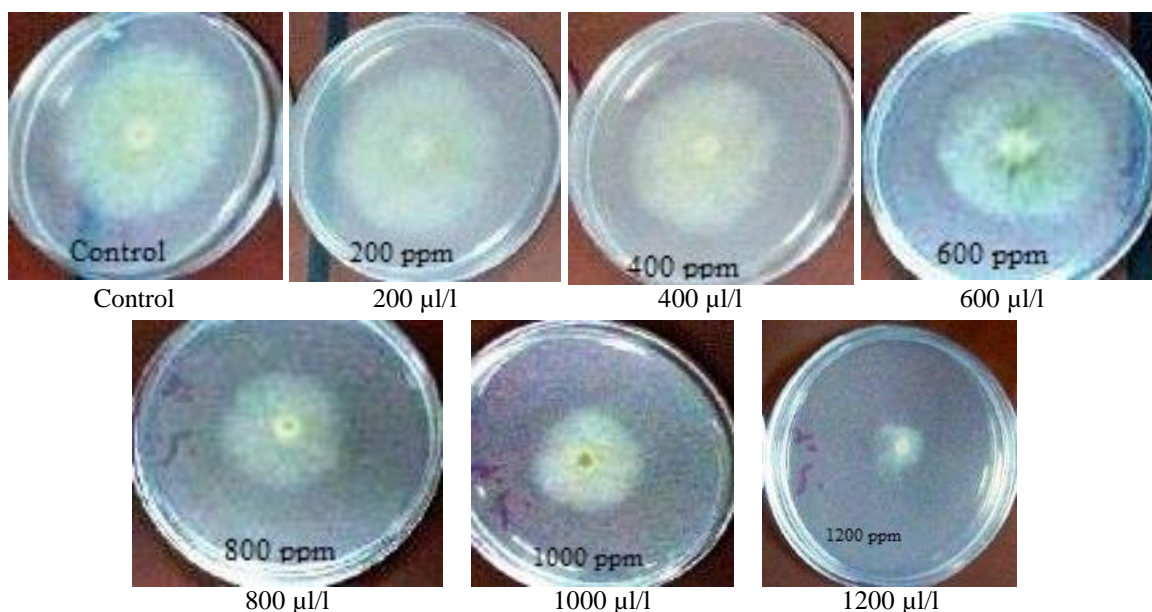


Figure 2. Effect of different pistachio (*Pistacia atlantica* L.) essential oil concentrations on inhibition of *Botrytis cinerea* in vitro culture

As compared to the untreated fruit, all tested PAO and CMC concentrations reduced the number of infected fruits (Figure 3). The results showed that PAO and CMC could significantly reduce the decay incidence on the grape compared to the control. Data on decay percentage indicated that control samples started decaying after 7 days while fruits coated by CMC 1 and 2% and PAO started decaying after 14 days of storage. This indicates that the organic extracts of *Pistacia atlantica* contain active compounds, with some antifungal activity. Previous reports indicate that fruit decay in raspberry was reduced during postharvest treatments with essential oil of *Melaleuca alternifolia* (Wang, 2003). The effect of carboxymethyl cellulose coating has also been investigated for delaying decay and its positive effect on maintaining peach fruit quality (Maftoonazad *et al.*, 2008). Our results are in accordance with the results of other researchers, who have reported antifungal activities of peppermint and sweet basil volatile oils (Ziedan and Farrag, 2008), cinnamon (*Cinnamomum zeylanicum* L.) oil (Tzortzakis, 2009; Xing *et al.*, 2010) and ajowan fennel essential oils (Abdolahi *et al.*, 2010). Coating made with CMC and PAO in the present study was effective in reducing grape berry decay compared to the uncoated fruits. These results were in agreement with those obtained by Hussain *et al.*, (2015) who recorded the extension of plum shelf life by CMC coating at 1 and 2%.

#### **Weight loss, Titratable Acidity, pH and Total Soluble Solids**

Weight loss of fruit in all treatments increased during the storage time, but was significantly greater in control than treated fruits (Figure 4). Weight loss occurred mainly during the first 7 days of storage and was more pronounced for controls than coated samples with PAO

and CMC which showed the smallest weight losses. The uncoated samples experienced an accelerated weight loss during storage, which can be attributed to an increase in the fruit's metabolic activity, associated with tissue senescence over long storage times. This process is slowed down after coating application, as reported in previous studies for the Muscate cultivar (Sánchez-González *et al.*, 2011a; Pastor *et al.*, 2011). Lower reduction of weight loss was obtained in the coated grape than the uncoated samples and essential oil improved this property, as might be expected from its hydrophobic nature (Sánchez-González *et al.*, 2009, 2010a, 2010b, 2011a). In agreement with present study, Hassani *et al.* (2015) showed that weight loss in essential oil treated apricots was lower than control. PAO coating had a strong effect in maintaining higher weight at the end of storage period. These results are in agreement with the results reported by Tian *et al.* (2011) which indicated coatings with eucalyptus and cinnamon oils significantly reduced weight loss compared to the control. The respiration rate was clearly affected by the essential oil concentration and the infection degree (Cristescu *et al.*, 2002). Similarly, in our experiment, it could be concluded that the tested essential oil had a positive influence on weight loss of grapevine fruits by reducing respiration rate. Our results illustrate that CMC has a potential as an edible coating for postharvest as well. Lower level of weight loss through the use of CMC coatings on grape has previously been observed by Yinze and Shaoying (2013). The positive effect of polysaccharide coatings as a water barrier in citric fruit and the efficient effect of CMC coating on preserving fruit quality have been reported (Valencia-Chamorro *et al.*, 2009; Hussain *et al.*, 2015).

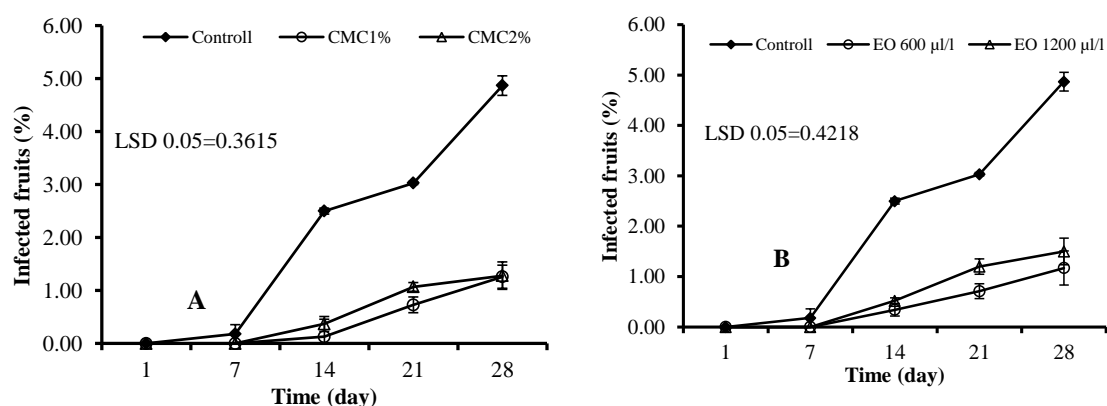


Figure 3. Effect of Carboxymethyl cellulose (CMC) (A) and pistachio (*Pistacia atlantica* L.) essential oil (EO) (B) on infected fruit of cold-stored grapes 28 days after storing. Bars in each mean symbol indicate the standard error.

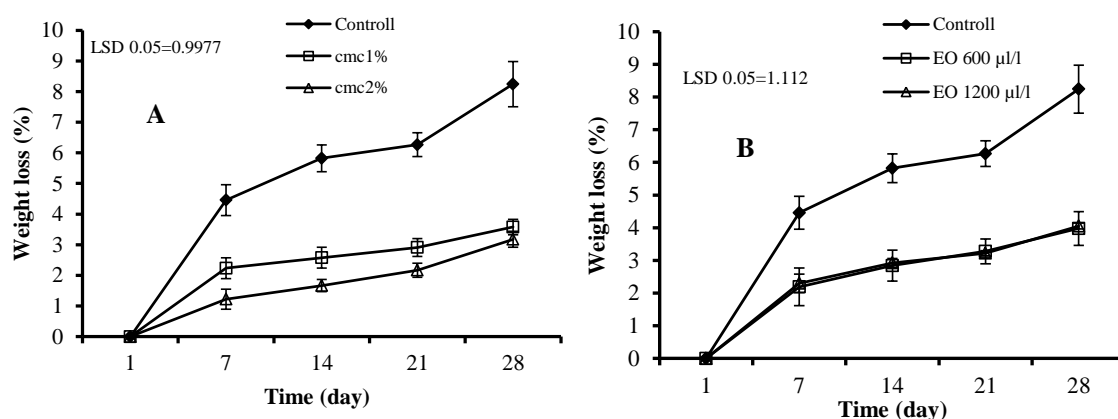


Figure 4. Effect of Carboxymethyl cellulose (CMC) (A) and pistachio (*Pistacia atlantica* L.) essential oil (EO) (B) on weight loss (%) of cold-stored grapes. Bars in each mean symbol indicate the standard error.

Change in total soluble solids contents (TSS), pH and titratable acidity (TA) of different samples as a function of storage time is shown in Figure 5 and Figure 6. TSS content and pH increased while TA decreased during storage with the progress of the ripening process. This behavior indicates gradual ripening of grapes during the storage period as reported by other researchers for other grape cultivars (Valero *et al.*, 2006; Meng *et al.*, 2008; Sánchez-González *et al.*, 2011a). The coating of grapevine berry with CMC and PAO had a strong impact on preserving of TSS and pH values at the storage period end. The highest amount of TSS was obtained for untreated samples, followed by those

coated with PAO and CMC. In agreement with present study previous study showed that using of *C. copticum* and *E. caryophyllata* oils for apricot fruit coating has retained TSS and TA during storage (Hassani *et al.*, 2012). Malmiri *et al.* (2011) has shown that coatings with cellulose has maintained TSS content considerably.

TA changes were lower in EO coated berries with CMC and PAO. It is possible that the rise in respiration of defective fruits stimulate usage of organic acids and leads to a reduction of fruit TA (Zokaee Khosroshahi *et al.*, 2007). Higher levels of TA in CMC and PAO coated table grape could be attributed to protective O<sub>2</sub> barrier or

oxygen supply decline on the surface of fruit which inhibits respiration (Jiang and Li, 2001). Therefore, our results

confirmed PAO beneficial effects on complex biochemical changes associated with ripening.

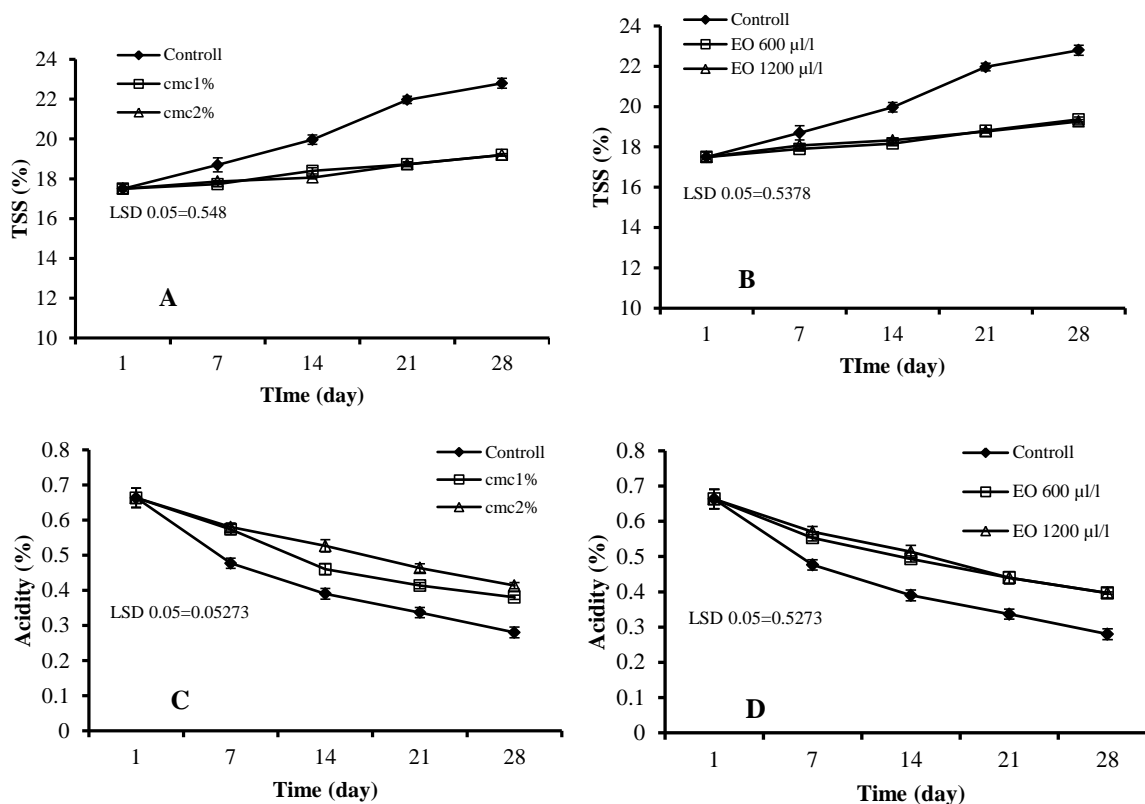


Figure 5. Effect of Carboxymethyl cellulose (CMC) (A, C) and pistachio (*Pistacia atlantica* L.) essential oil (EO) (B, D) on total soluble solid (TSS) and titratable acidity (TA) of cold-stored grapes. Bars in each mean symbol indicate the standard error.

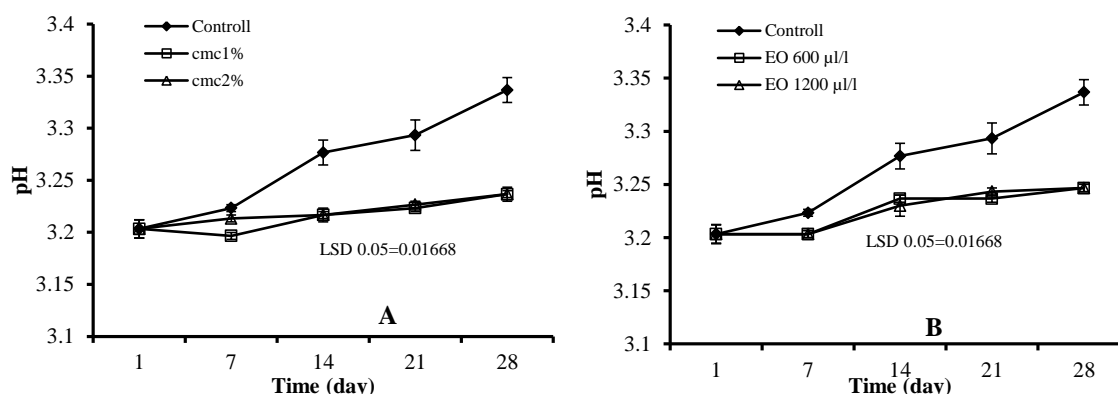


Figure 6. Effect of Carboxymethyl cellulose (CMC) (A) and pistachio (*Pistacia atlantica* L.) essential oil (EO) (B) on pH of cold-stored grapes. Bars in each mean symbol indicate the standard error.

#### Anthocyanin, antioxidant activity and phenol content

The anthocyanin, antioxidant activity, phenol and tannin content of the

samples in all treatments were declined during the storage period (Figure 7, 8, 9, 10). Anthocyanin of berry in all samples were reduced during the



storage time, but was significantly lower in uncoated than in coated fruits at the storage end (Figure 7). Therefore, CMC and PAO-coated fruits retained higher anthocyanins and phenolics content than uncoated fruits. Moreover, anthocyanin level in fruits coated with 2% CMC and  $1200 \mu\text{l l}^{-1}$  PAO was higher than the fruits coated with 1% CMC and  $600 \mu\text{l l}^{-1}$  PAO. Cellulose coatings have been widely employed to retardation of ripening and fruit quality loss (Yaman and Bayoindirli, 2001, Zhou *et al.*, 2008). CMC as edible cellulose coating could extend storage life and inhibit fruit decay due to their capacity to alter the tissues internal atmosphere (Tien *et al.* 2000). Edible coatings showed a positive effect on preserving higher content of total phenolic compounds and anthocyanin, which were declined in untreated fruits because of senescence processes and over-ripening (Gol *et al.*, 2013). Decrease of anthocyanin and phenolic content in present study in treated fruits could in all probability be related to the delay of fruit senescence in CMC and PAO coating treatments. Similar results were also reported by Hussain *et al.* (2016) who found that after 28 days of refrigerated storage, the anthocyanin content was higher in cherries treated with CMC. In addition, essential oils improved anthocyanins contents in raspberries after 7 days of storage (Jin *et al.*, 2012).

For the coated as well as control grape berries, total phenolic content was progressively decreased over the entire storage period, but control fruits had greater decrease (Figure 8) which has previously been observed in grapes (Valero *et al.* 2006, Meng *et al.*, 2008, Sánchez-González *et al.*, 2011a) and strawberries (Ferreira *et al.*, 2007). Activity of phenylalanine ammonia-lyase (PAL) decreases in the maturation and postharvest stages and reduction of

PAL activity leads to a reduction in phenolic accumulation in grape (Meng *et al.*, 2008). During storage, the fruit coated with PAO and CMC exhibited higher levels of phenols with respect to that of control. Previous work showed that postharvest putrescine and chitosan treated grape berries and chitosan treated tomato fruits exhibited higher total phenolic and antioxidant activity compared with control (Liu *et al.*, 2007; Shiri *et al.*, 2013). Phenolic compounds might be contributed to antioxidant activity and fruit quality (Díaz-Mula *et al.*, 2012); coated fruits with higher phenolic content would have higher quality than controls. As previous study demonstrates, the fruit coating created a semi-permeable barrier that restricted gas exchange, reduced water loss, delayed ripening and senescence by modifying endogenous  $\text{CO}_2$  and  $\text{O}_2$  (Wang and Gao, 2013; Aloui *et al.*, 2014). It seems that coatings were able to delay the loss of phenolic compounds during storage, by acting as an effective barrier against oxygen transmission.

Fruit tannin in all treatments decreased markedly during the storage time, but tannin concentration in coated fruits was higher than control (Figure 9). Khademi *et al.* (2013) showed that tannin content decreased as the storage day increased in persimon. Reduction of soluble tannin could be related to the polyphenol oxidase enzyme activity which oxidizes tannin to phenol (Bello and Henry, 2015) and complex formation between the pectin derived from cell wall, and tannins (Taira *et al.*, 1997). Coating with CMC and PAO might lead to reduction of reaction with oxygen and higher rates of tannin is preserved under these conditions. As tannins interact with oxygen this compound further polymerizes and becomes less astringent (Conde *et al.*, 2007). In agreement with the present study Barman *et al.* (2014) reported

coating of pomegranate aril with putrescine + carnauba maintained significantly higher tannin content than control on 60<sup>th</sup> day of the storage period.

The sample antioxidant capacity was highest during the first seven storage days, but afterwards strongly decreased in all treatments (Figure 10), which could be attributed to the reduction of phenolic compounds and tannin (Figure 8,9,10). In our study, the application of CMC (2%) and PAO (1200  $\mu\text{l l}^{-1}$ ) resulted in preserving the higher antioxidant activity after 28 days in coated fruits than uncoated fruits. The main cause for higher maintenance of total antioxidant activity might be explicated by lowering losses of anthocyanin, phenolic contents and tannins in treated fruits with CMC and PAO during storage period. Grape is

considered as great sources of phenolic compounds, which are mostly responsible for the grapes antioxidant attributes (Baiano and Terracone, 2011). Therefore, preservation of phenolic content in fruits coated with CMC and PAO has led to maintenance of higher antioxidant activity. Another reason for higher antioxidant retention in PAO treated berry might be due to PAO capacity to acting as scavenger of free radicals. There have been numerous studies on antioxidant activity of *P. lentiscus*, which is a considerable source of natural antioxidants (Hatamnia *et al.*, 2014; Gourine *et al.*, 2010). Similar to the present study, application of cinnamon leaf oil inhibited fungal decay and preserved antioxidant activity of grape during storage period (Melgarejo-Flores *et al.*, 2013).

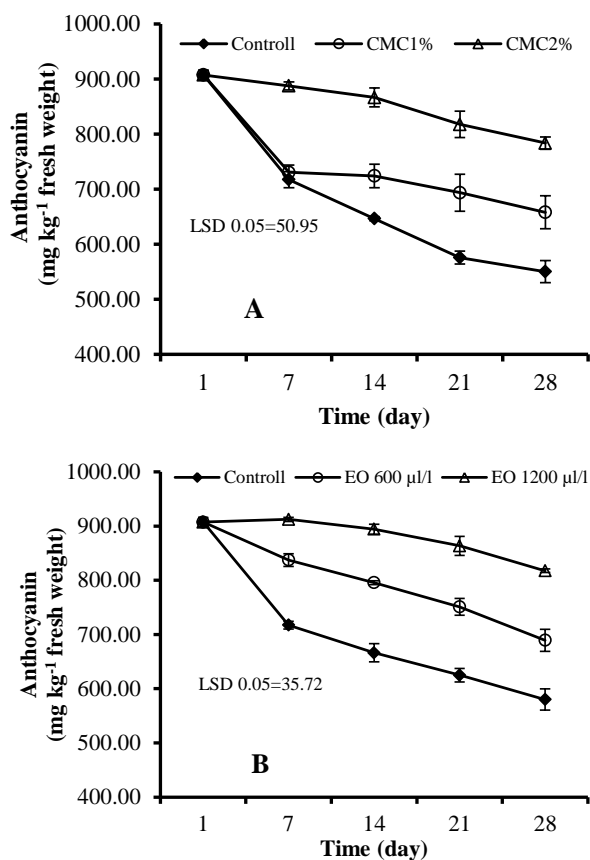


Figure 7. Effect of Carboxymethyl cellulose (CMC) (A) and pistachio (*Pistacia atlantica* L.) essential oil (EO) ( $\mu\text{l l}^{-1}$ ) (B) on anthocyanin of cold-stored grapes. Bars in each mean symbol indicate the standard error.

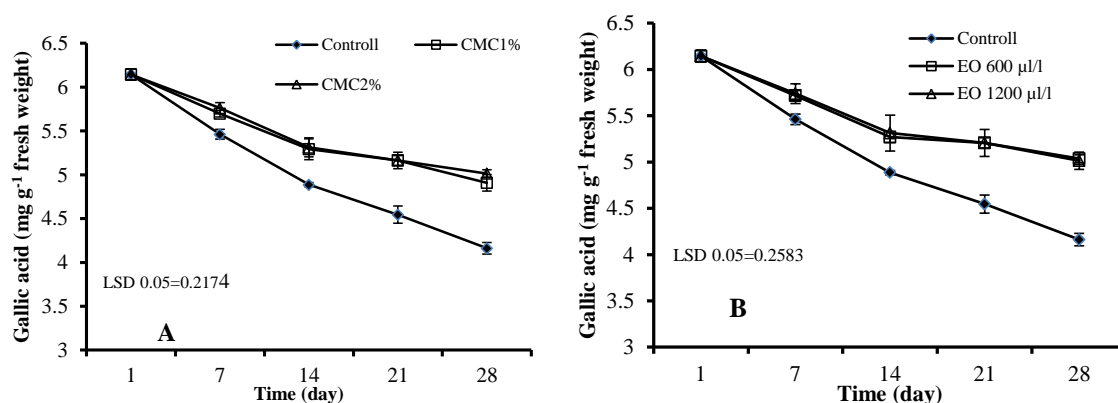


Figure 8. Effect of Carboxymethyl cellulose (CMC) (A) and pistachio (*Pistacia atlantica* L.) essential oil (EO) ( $\mu\text{l l}^{-1}$ ) (B) on phenol (Gallic acid) of cold-stored grapes. Bars in each mean symbol indicate the standard error.

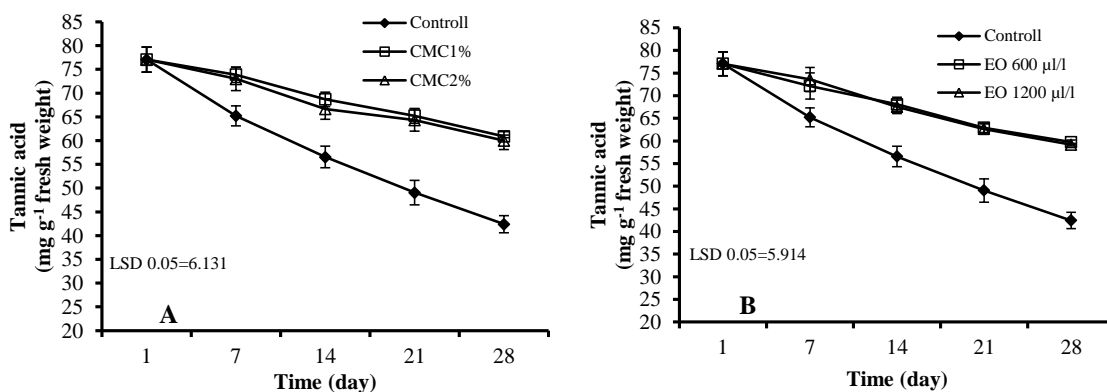


Figure 9. Effect of pistachio (*Pistacia atlantica* L.) essential oil (EO) ( $\mu\text{l l}^{-1}$ ) and Carboxymethyl cellulose (CMC) on tannin (Tannic acid) of cold-stored grapes. Bars in each mean symbol indicate the standard error.

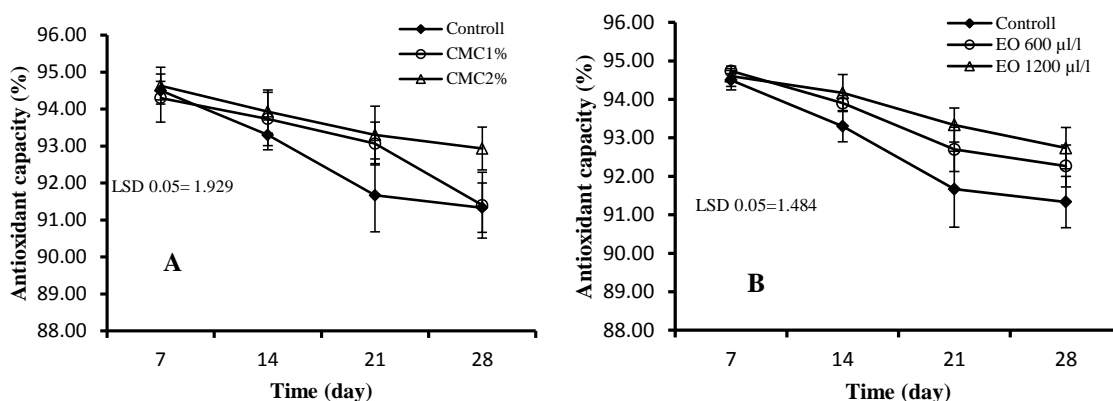


Figure 10. Effect of Carboxymethyl cellulose (CMC) (A) and pistachio (*Pistacia atlantica* L.) essential oil (EO) (B) on antioxidant capacity of cold-stored grapes. Bars in each mean symbol indicate the standard error.

## Conclusions

Our results indicated that postharvest PAO treatment and CMC can be effective tools for extended storage and shelf life of "Rasheh" grape. The highest

antifungal effect was observed in 1200  $\mu\text{l l}^{-1}$  PAO. PAO as a coating material resulted in fruit decay reduction due to the PAO antifungal properties. CMC and PAO coating prevent weight losses

and the degradation of TSS and TA. This edible coating had a positive effect on preservation higher concentrations of total phenols and anthocyanin, which were decreased in uncoated fruits. Coatings with 2% CMC and 1200  $\mu\text{l l}^{-1}$  PAO showed greatest preservation of antioxidant activity. The use of this

useful coating enhances the benefits based on preservation of qualitative parameters and limiting of the microbial spoilage. According to our results the application of *P. Atlantica* essential oil might be a groundbreaking and valuable tool, and a substitute for synthetic fungicides during grape storage.

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