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Preharvest Application of Sulfur as Pesticide on Fresh Hull and Kernel of Pistachio (Pistacia vera L.)

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ABSTRACT

In recent years, sulfur has been shown to be effective in controlling pistachio psylla. In this study, the effect of sulfur foliar application as psylla pesticide on fresh fruit and kernel of three cultivars of pistachio was determined and compared with two commercial pesticides. Refinery micronized and mineral sulfur were applied at two concentrations of 30 and 60 kg/1000 L of water. The quality of fresh pistachio hulls and kernels were investigated. In 'Fandoghi' cultivar, all sulfur treatments reduced the kernel moisture. The firmness and water activity of hulls and kernels were significantly affected. All sulfur treatments decreased hull lightness in 'Fandoghi', while it increased in 'Akbari' and 'Ahmadaghaei' cultivars. Reduction of hull chroma was observed in some cases in all cultivars. Hue of hulls was not affected by the treatments. Lightness, chroma and hue values of the testa of kernels were not affected in 'Fandoghi' and 'Akbari'. In 'Ahmadaghaei', refinery micronized sulfur increased the testa lightness and hue and decreased chroma probably because of the softer texture of hull in 'Ahmadaghaei' which breaks more easily during harvest practice. Sulfur treatments positively influenced appearance and general acceptance of 'Ahmadaghaei' fruits. The taste and aroma of pistachios were preserved in most of treatments. High concentration (60 kg/1000 L) of both sulfur types enhanced the ethylene production in 'Ahmadaghaei' which was almost same as the unripe fruit because sulfur treatments reduced the percentage of unripe fruit. In 'Ahmadaghaei', refinery micronized sulfur treatments increased the respiration rate.

Introduction

The genus *Pistacia* belongs to Anacardiaceae family that has several species, among them *Pistacia vera* has commercially valuable and edible seeds (Gheysarbigi et al., 2020). Pistachio tree is native to west of Asia, however, today it is also cultivated in south of Europe, north of Africa, China and west of USA. According to Food and Agriculture Organization (FAO), with the production of 440,000 tons of pistachios, Iran was the first producer and exporter of this product in the world in 2014. The pistachio psylla, *Agonoscena pistaciae*, is an important pest of pistachio orchards. This insect is a native pest of pistachio cultivars in Iran and neighboring countries and is known among pistachio growers as dry sap. It causes irreparable damage to pistachio crop, sometimes destroying the crop for three consecutive years (Hosseini Naveh and Abbasi, 2020). Psylla is active in early spring with the onset of vegetative growth of pistachio trees until late fall. It attacks buds, young leaves and stems, and reproduces rapidly. It causes significant economic losses, and the control of this pest relies almost exclusively on pesticides (Mehrnejad, 2002).

Movento is a widely used pesticide by pistachio growers which has a relatively long durability and effectiveness (TahamiZarandi et al., 2018). Imidacloprid or confidor is another poison used for controlling pistachio psylla (Rouhani and Samih, 2013). Despite the application of various poisons, psylla is still considered a serious challenge in pistachio orchards because it has become quite resistant to the applied poisons (Afrousheh and Hasheminasab, 2018).

Sulfur is a macronutrient playing important roles in normal plant growth. It is involved in the composition of amino acids and secondary metabolites, in nitrogen metabolism, and causes resistance to pests and diseases (Rodrigues and Zoffoli, 2016). The effective application of sulfur as a pesticide has been one of the strategies used in agriculture for years. Its effect is non-systemic contact. Sulfur in the commercial form was produced as a byproduct of coal, natural gas and petroleum refinement and mining process. It is applied as dust (powder), granular (colloidal formulations) or liquid (wettable) forms. Generally, sulfur is compatible with other pesticides. It can be used pre and postharvest in various forms and through several ways such as foliar spraying, fumigation, SO_2 generating pads inside the packages and etc. It is used for several hundred kinds of foods and crops, ornamental plants, turfs, and residential sites (Afrousheh and Hasheminasab, 2018). The effectiveness of sulfur for controlling pistachio psylla is reported in literature. For example, the average population of psylla nymphs decreased up to 63 days after sulfur spraying on each leaflet comparing control (Mohammadi Nia and Emami, 2018). In another study, sulfur in combination with kaolinite was even up to 98% efficient on reducing pistachio psylla (Bakhtiari et al., 2016). Besides pistachio psylla, sulfur is widely used for controlling brown rot of peaches (Holb and Schnabel, 2008), powdery mildew of apples (Holb and Kunz, 2016), peanut leaf spot (Culbreath et al., 2019), mildew on roses (Hosseininia et al., 2008) and etc.

Various forms of sulfur as a pesticide have been extensively used in pistachio orchards in the recent years, but it is important for a pesticide having no negative affect on the quality of the product. The reports by growers contain negative and positive reflections in relation to this pesticide. In this study, sulfur spraying was applied as a useful, low-cost tool to reduce the losses in pistachio caused by psylla and it was compared with two chemical pesticides including movento and confidor. Refinery micronized and mineral sulfur were applied separately in two different concentrations. The quality and sensory attributes of hull and kernel were determined. Because of varietal differences within the pistachio species, three commercial cultivars including 'Fandoghi', 'Ahmadaghaei' and 'Akbari' were chosen and compared.

Materials and Methods

Plant material and treatments

Preliminary studies of this project were began in May 2019 in the orchards of Anar city, Iran. Three pistachio cultivars including 'Fandoghi', 'Ahmadaghaei' and 'Akbari' were selected and sprayed with movento (0.5 kg/1000 L), confidor (0.4 kg/1000 L) and two types of sulfur, including refinery micronized sulfur (30 and 60 kg/1000 L in water) and the mineral sulfur (30 and 60 kg/1000 L in water). The concentrations were chosen based on farmer's recommendations. Control trees were not sprayed. The spraying was repeated after 50 days. The experiment was performed in the form based on a complete randomized block design with 4 replications. Each treatment consisted of three rows of trees, the replicates of which were selected in the middle row. Six neighboring trees were considered as one replication and the sampling (at least 10 fruits) was done from four sides of each tree (Mohammadi Nia and Emami, 2018; Hosseini et al., 2020). The spraying operation was carried out in the morning and ended before the weather warmed up. In September and early October, at the same time for pistachio harvest, the fruits were sampled and transferred to laboratory.

Moisture content

A number of 5 kernels or 15 hulls (the exomesocarp or the soft skin) were used for moisture content determination. The weight of each replicate was recorded on the first day and after drying for two days at 40 °C. The cumulative weight loss was obtained from the difference between the weight of the first day and the weight after drying and was expressed as a percentage of the original fresh weight (Cantin et al., 2012).

Unripe fruit

For each replicate, the entire package was counted. The percentage was assessed by dividing the number of unripe fruits over the total number of fruits multiplied by 100 (Cantin et al., 2012).

Firmness

Tissue firmness was evaluated using a Digital Force Tester (Lutron fg5020, Taiwan), fitted with a sharp 11 mm probe. Two different measurements were carried out on two opposite sides of the central zone of kernels and hulls. The values were expressed as kilogram-force (KgF) (Wang et al., 2006).

Water activity

One g of kernels or hulls were used. Water activity was directly measured by a water activity meter (Novasina, Switzerland) and expressed as a_w (Rossello et al., 1994).

Color indicators

Color values of hulls and kernels (with testa) were directly measured with a color meter (Minolta Chroma Meter Model CR-400, Minolta, Japan). The color was measured as the lightness (L*), red-green (a*) and blue-yellow (b*). The chroma value and hue angle were calculated by the fallowing equations (Equations 1 and 2) (Gheysarbigi et al., 2020).

Equation 1: Chroma $=\sqrt{(a*)^2 + (b*)^2}$

Equation 2: hue angle $= \tan^{-1}(\frac{b*}{a*})$

Sensory evaluation
The sensory analysis included a panel test constituted of ten semi-trained panelists. The panelists evaluated the taste, aroma, appearance and general acceptance of fruit. Excellent was shown with five, very good with four, good with three, moderate with two, poor with one (Wang et al., 2006).

Ethylene production and respiration rate
15 g of fresh pistachio fruits of 'Ahmadaghaei' cv. was obtained from each treatment and placed in 1.6 L airtight respiration jars at 20 °C and allowed to equilibrate for 2 h. 1 mL of head-space atmosphere was withdrawn using plastic syringes and injected into a gas chromatograph (Agilent 7890B, USA) equipped with a packed column and a thermal conductivity detector (TCD) for quantification of CO_2 or C_2H_4. Injector, detector and oven temperature of gas chromatograph was set at 100, 120, 80°C respectively. Nitrogen was used as a carrier gas at the flow rate of 73.7 mL min^{-1}. The respiration rate and ethylene production were expressed as ng kg^{-1} s^{-1} (Gheysarbigi et al., 2020).

Statistical analysis
The study was a factorial experiment based on a completely randomized design with four replications. Sources of variation were cultivars ('Fandoghi', 'Ahmadaghaei' and 'Akbari'), treatments (control, movento, confidor, R30, R60, M30, and M60) and their interactions. Mean values were calculated and reported as the mean $\pm$ standard error of means. Data were analyzed by SAS 9.1 statistical software package, and the least significant difference (LSD) test at P = 0.01 or 0.05 was used to compare the means. The graphs were plotted in MS-Excel software package.

Results

Moisture content
Cultivar, treatments and their interaction had significant effects on moisture content of kernels but not hulls (Table 1). 'Ahmadaghaei' and 'Akbari' cultivars were similar to each other, from this point that none of the treatments influenced the moisture of kernels compared to the control. But in 'Fandoghi', confidor and all sulfur treatments reduced the moisture of kernels (Fig. 1).

Table 1. Analysis of variance of some traits of pistachios under the effects of pesticides (movento (0.5 kg/1000 L), confidor (0.4 kg/1000 L), refinery micronized sulfur at two concentrations (30 and 60 kg/1000 L) and mineral sulfur at two concentrations (30 and 60 kg/1000 L)), cultivar ('Fandoghi', 'Ahmadaghaei' and 'Akbari') and their interaction

Sources of variation	df	Mean squares						
		Hull moisture (%)	Kernel moisture (%)	Unripe fruit (%)	Hull firmness (KgF)	Kernel firmness (KgF)	Hull water activity (a_w)	Kernel water activity (a_w)
Treatment	6	17.73^{ns}	124.31**	188.17**	1.40*	0.13^{ns}	0.000007^{ns}	0.00002**
Cultivar	2	7.10^{ns}	391.06**	2855.17**	59.80**	10.19**	10.000170**	0.00014**
Treatment × cultivar	12	5.53^{ns}	72.13**	199.92**	1.60**	0.68**	0.000025**	0.00003**
Error	42	13.98	12.30	23.42	0.59	0.13	0.000006	0.000006
Coefficient of variation		4.84	8.82	23.52	21.93	12.32	0.26	0.25

Sources of variation	df	Mean squares					
		Hull lightness	Kernel lightness	Hull chroma	Kernel chroma	Hull hue	Kernel hue
Treatment	6	119.47**	275.81**	141.83**	130.95**	0.17^{ns}	0.08**
Cultivar	2	2523.73**	52.5.06**	729.16**	337.76**	1.70**	26.56**
Treatment × cultivar	12	320.02**	201.38**	94.93**	124.41**	0.19^{ns}	0.16**
Error	42	1.36	16.40	1.08	2.14	0.16	0.01
Coefficient of variation		2.17	7.04	3.75	4.87	0.22	0.06

Sources of variation	df	Mean squares					
		Taste	Aroma	Appearance	Acceptance	Ethylene (ng kg^{-1} s^{-1})	Respiration (ng kg^{-1} s^{-1})
Treatment	6	0.91**	0.34**	1.22**	0.45**	0.17**	190.90**
Cultivar	2	3.73**	0.94**	11.38**	1.77**	-	-
Treatment × cultivar	12	0.46**	0.21*	1.60**	0.67**	-	-
Error	42	0.10	0.09	0.07	0.12	0.007	4.97
Coefficient of variation		7.89	7.30	6.59	7.96	9.10	14.85

* and ** show significance at the 5% and 1% levels, and ns means no significant difference.

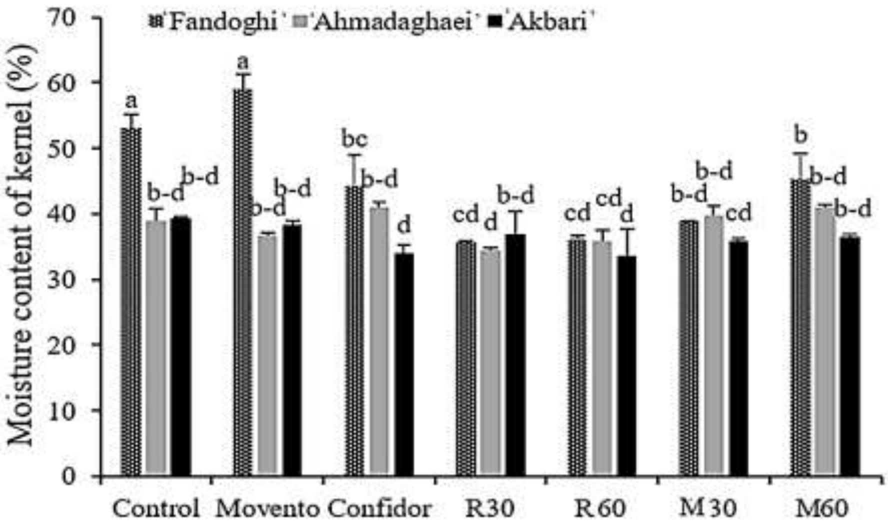

Fig. 1. Moisture content of kernels of three pistachio cultivars ('Fandoghi', 'Ahmadaghaei' and 'Akbari') that were treated preharvest with movento (0.5 kg/1000 L), confidor (0.4 kg/1000 L), refinery micronized sulfur at two concentrations (30 and 60 kg/1000 L) (R30 and R60) and mineral sulfur at two concentrations (30 and 60 kg/1000 L) (M30 and M60)

Unripe fruit

Cultivar, treatments and their interaction had significant effects on unripe fruit percentage (Table 1). 'Fandoghi' had very low percentages of unripe fruit because it normally is a faster cultivar regarding fruit ripening. 'Akbari' seemed not to be very much affected by the treatments but, in 'Ahmadaghaei' the treatments stimulated the ripening process (Fig. 2).

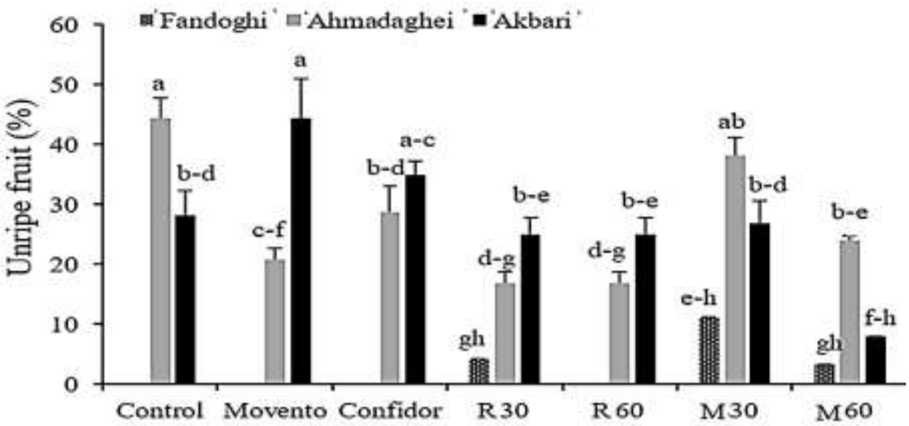

Fig. 2. Unripe fruit percentage of three pistachio cultivars ('Fandoghi', 'Ahmadaghaei' and 'Akbari') that were treated preharvest with movento (0.5 kg/1000 L), confidor (0.4 kg/1000 L), refinery micronized sulfur at two concentrations (30 and 60 kg/1000 L) (R30 and R60) and mineral sulfur at two concentrations (30 and 60 kg/1000 L) (M30 and M60)

Firmness

The interaction of treatments and cultivar had significant effects on hull and kernel firmness (Table 1). 'Akbari' and 'Ahmadaghaei' cultivars had the highest and the lowest hull firmness. The treatments did not affect hull firmness except confidor which reduced it in 'Akbari' cv. (Fig. 3 A). 'Fandoghi' showed the least kernel firmness and the treatments were not effective on this trait in most cases (Fig. 3 B).

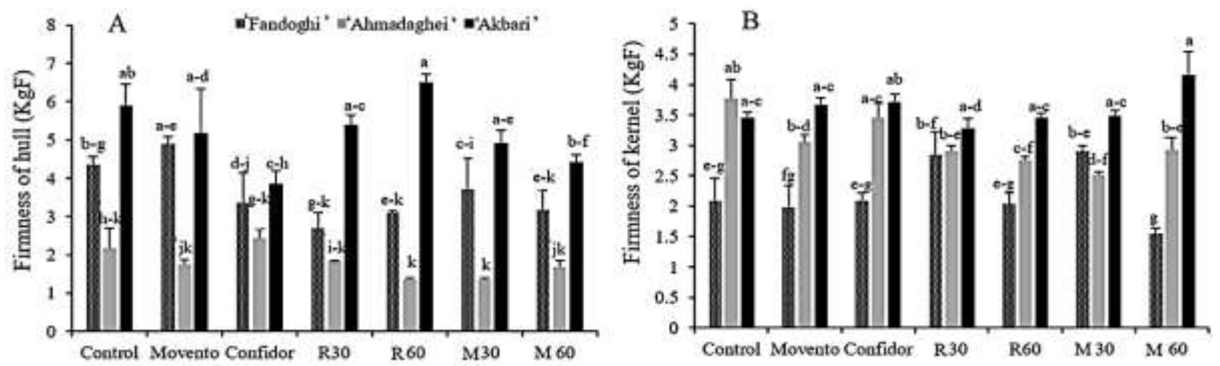

Fig. 3. Firmness of hulls and kernels of three pistachio cultivars ('Fandoghi', 'Ahmadaghaei' and 'Akbari') that were treated preharvest with movento (0.5 kg/1000 L), confidor (0.4 kg/1000 L), refinery micronized sulfur at two concentrations (30 and 60 kg/1000 L) (R30 and R60) and mineral sulfur at two concentrations (30 and 60 kg/1000 L) (M30 and M60)

Water activity

The interaction of treatments and cultivar had significant effects on hull and kernel water activity (Table 1). Sulfur treatments did not cause a significant effect on water activity of hulls and kernels in most cases. M60 increased the water activity of 'Ahmadaghaei' hulls and kernels (Fig. 4A,B).

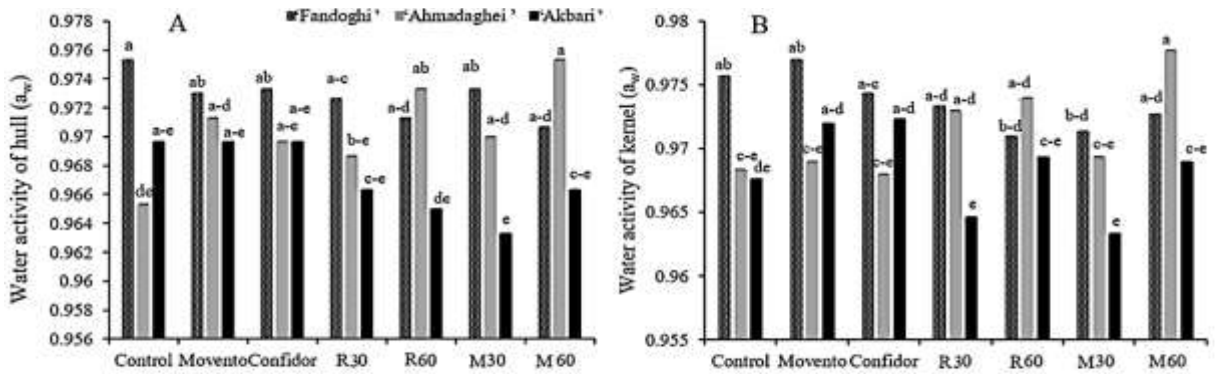

Fig. 4. Water activity of hulls (A) and kernels (B) of three pistachio cultivars ('Fandoghi', 'Ahmadaghaei' and 'Akbari') that were treated preharvest with movento (0.5 kg/1000 L), confidor (0.4 kg/1000 L), refinery micronized sulfur at two concentrations (30 and 60 kg/1000 L) (R30 and R60) and mineral sulfur at two concentrations (30 and 60 kg/1000 L) (M30 and M60)

Color indicators

Cultivar, treatments and their interaction had significant effects on color indicators of hulls and kernels except hue of hulls which was only affected by cultivar (Table 1). 'Fandoghi' had the highest lightness and hue values of hulls while 'Ahmadaghaei' had the highest chroma value of hulls. Sulfur influenced the lightness and chroma of hulls in some cases, so that all sulfur treatments decreased hull lightness in 'Fandoghi' although, some of them increased this trait in 'Akbari' and 'Ahmadaghaei' (Fig. 5 A). Reduction of hull chroma by sulfur was also observed in some cases in all cultivars (Fig. 5 B). Hue value of hulls which is the basic color of them was not affected by treatments but only by cultivar (Fig. 5 C).

The lightness, chroma and hue values of the testa of kernels were not affected by treatments in 'Fandoghi' and 'Akbari'. But, in 'Ahmadaghaei', refinery micronized sulfur at both concentrations increased the testa lightness and hue values and decreased chroma value (Fig. 5 D,E,F).

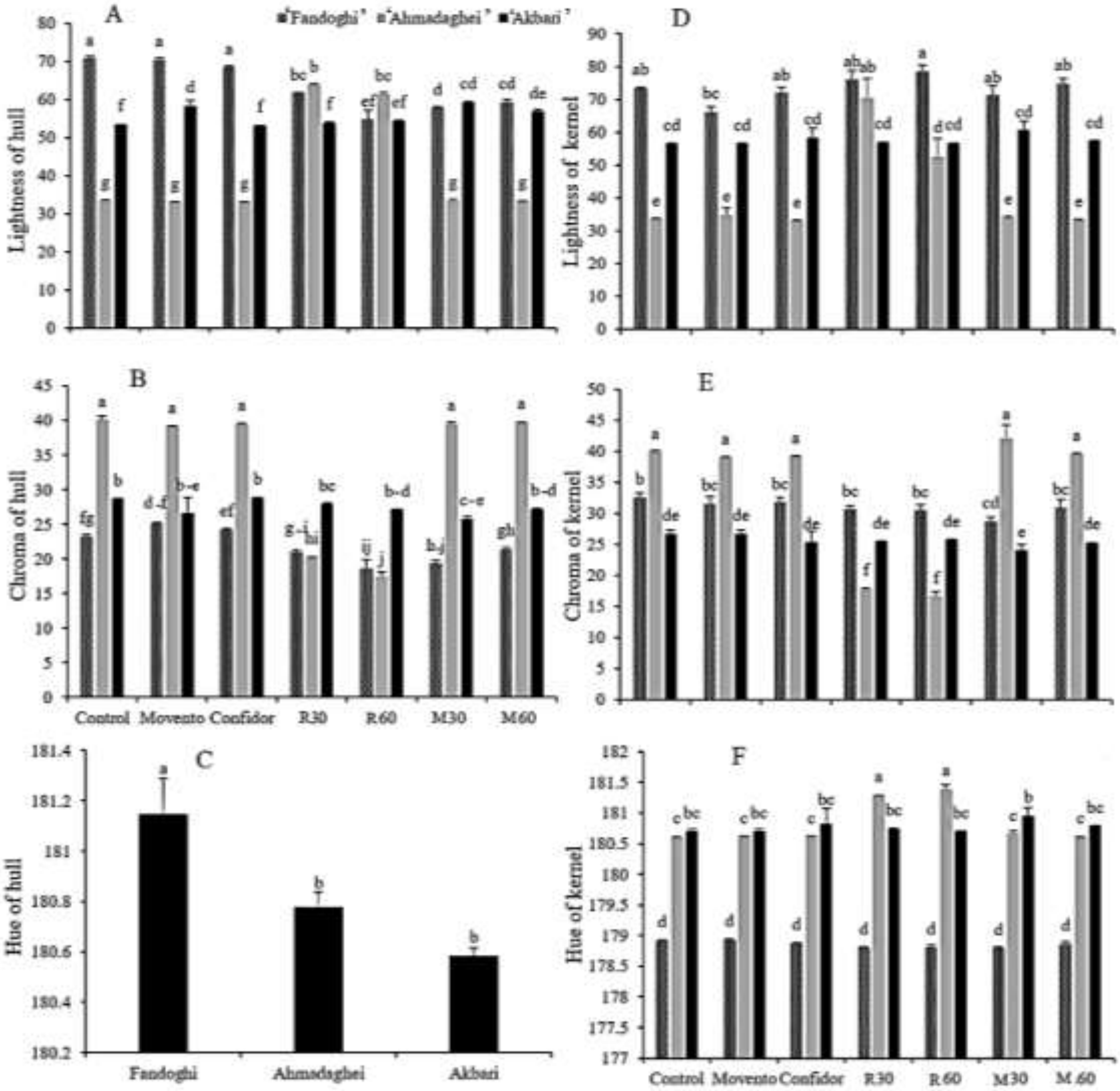

Fig. 5. Color indicators of hulls and kernels of three pistachio cultivars ('Fandoghi', 'Ahmadaghaei' and 'Akbari') that were treated preharvest with movento (0.5 kg/1000 L), confidor (0.4 kg/1000 L), refinery micronized sulfur at two concentrations (30 and 60 kg/1000 L) (R30 and R60) and mineral sulfur at two concentrations (30 and 60 kg/1000 L) (M30 and M60)

Sensory evaluation

Cultivar, treatments and their interaction had significant effects on taste, aroma, appearance and general acceptance of pistachios (Table 1). 'Fandoghi' and 'Akbari' cv. had a good degree of fruit appearance and general acceptance which were not very much affected by treatments in most cases but, 'Ahmadaghaei' seemed not to be desirable in control and movento and confidor treated fruits. Although, sulfur treatments had a good impact on appearance and general acceptance of 'Ahmadaghaei' (Fig. 6 A,B). The taste and aroma of pistachios were preserved in most cases of treatments. Some exceptions were M30 and 60 that lowered the taste of 'Akbari' cv. A similar result was seen in R60 for 'Fandoghi' cv. (Fig. 6 C). R60 influenced the aroma in 'Akbari' cv. (Fig. 6 D).

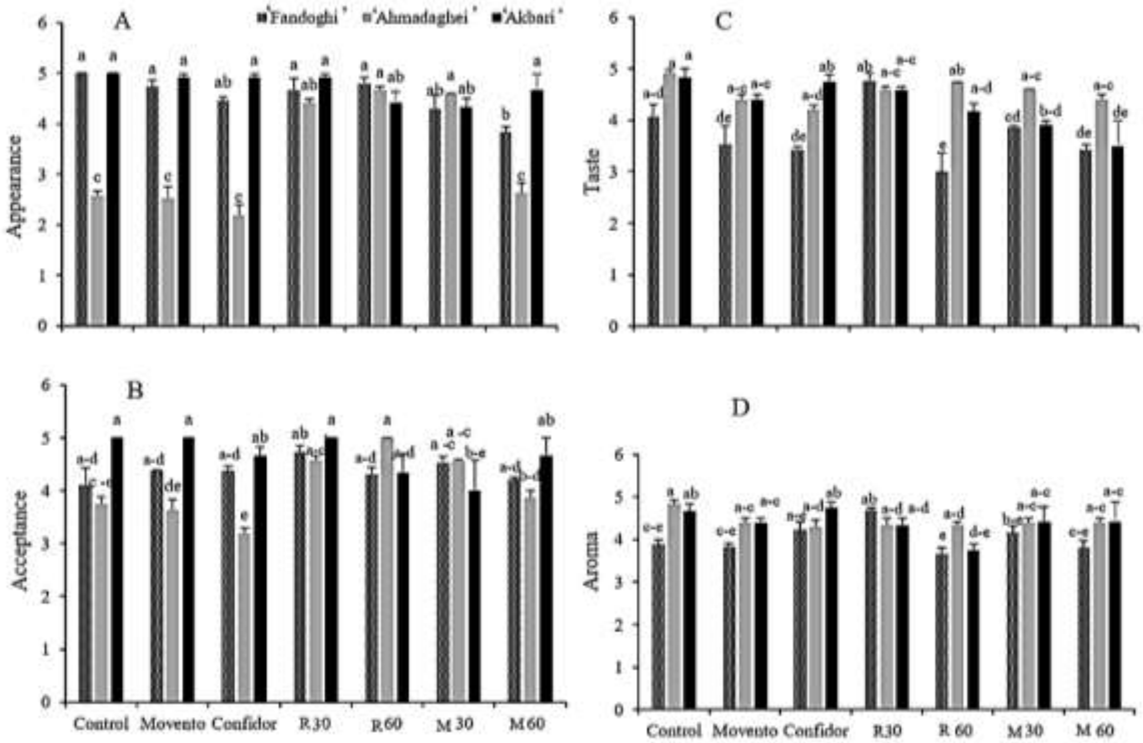

Fig. 6. Sensory indicators of three pistachio cultivars ('Fandoghi', 'Ahmadaghaei' and 'Akbari') that were treated preharvest with movento (0.5 kg/1000 L), confidor (0.4 kg/1000 L), refinery micronized sulfur at two concentrations (30 and 60 kg/1000 L) (R30 and R60) and mineral sulfur at two concentrations (30 and 60 kg/1000 L) (M30 and M60)

Ethylene production and respiration rate

Based on the results of unripe fruit, only 'Ahmadaghaei' cv. was tested for ethylene production and respiration rate. The treatments had significant effects on these traits (Table 1). Sulfur treatment enhanced the ethylene production in two cases including R60 and M60 (high concentrations) (Fig. 7 A), which is somehow in agreement with the results of unripe fruit because sulfur treatments reduced the percentage of unripe fruit (Fig. 2). Both refinery micronized sulfur treatments increased the respiration rate while other treatments did not affect this trait (Fig. 7 B).

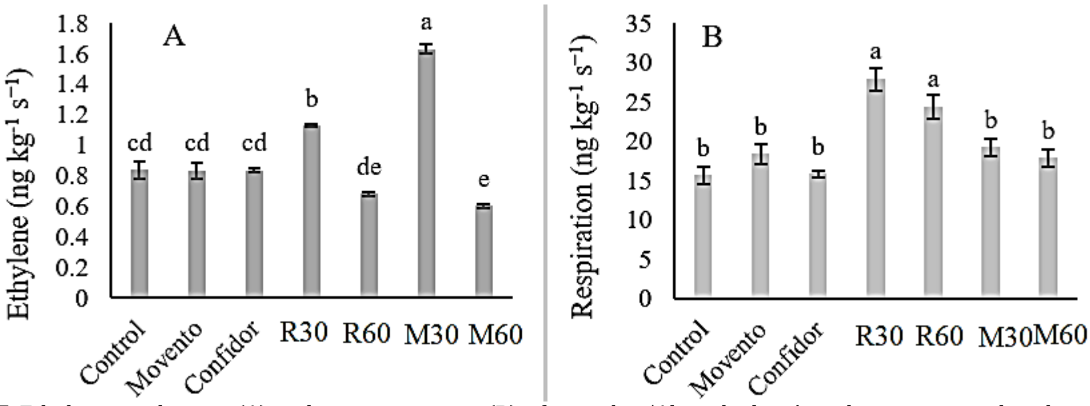

Fig. 7. Ethylene production (A) and respiration rate (B) of pistachio 'Ahmadaghaei' cv. that was treated preharvest with movento (0.5 kg/1000 L), confidor (0.4 kg/1000 L), refinery micronized sulfur at two concentrations (30 and 60 kg/1000 L) (R30 and R60) and mineral sulfur at two concentrations (30 and 60 kg/1000 L) (M30 and M60)

Discussion

The difference in moisture content among cultivars may reflect the differences in cuticle permeability or surface to volume ratios (Cantin et al., 2012). In blueberries, SO_2 fumigation did not affect the fruit moisture of Brigitta and O·Neal cultivars. But in cultivar Legacy, high concentrations of SO_2 (250-350 μL L^{-1}) kept the moisture during storage, although fumigation with 50-200 μL L^{-1} did not affect the moisture of fruit (Rivera et al., 2013). In another study on blueberries, when SO_2 was used in combination with a perforated polyethylene bag, lower water loss was observed postharvest comparing bag alone (Rodrigues and Zoffoli, 2016).

In higher plants, sulfur plays critical roles in the catalytic or electrochemical functions of the biomolecules in cells. Sulfur is an essential component for the synthesis of the nutritionally important amino acids cysteine and methionine, as well as for a wide range of sulfur-containing metabolites (Saito, 2004). Methionine converts to S-adenosyl methionine which is an ethylene precursor (Asgher et al., 2018). Since ethylene has an important role in plant senescence and fruit ripening, the acceleration of fruit ripening in 'Ahmadaghaei' cv. can be related to the effect of sulfur in methionine production. This is in agreement with our results on ethylene production. In peach cv. Cresthaven, the fruits treated with sulfur-based products were slightly more mature comparing fruits treated with fungicides (Schnabel and Rayne, 2004).

Firmness is an important characteristic of fruits that increases the potential for storage and confers resistance to disease and mechanical damage (Fisher and Bennett, 1991). Therefore, it is essential for pre and postharvest treatments to keep the firmness of plant tissues and have no adverse effect on it. In our study, the treatments did not influence the firmness except R60 and M30 which caused a slight decrease in kernel firmness in 'Ahmadaghaei' cv. A reduction in firmness of blueberry fruits cv. Liberty fumigated by SO_2 has been reported (Rivera et al., 2013). When blueberry cv. Legacy is stored in modified atmosphere packaging or in perforated bags in combination with SO_2 a reduction in firmness can be seen (Rodrigues and Zoffoli, 2016). But in another study, the firmness of figs was not consistently affected by different SO_2 treatments postharvest (Cantin et al., 2011).

Water activity is the ratio of the vapor pressure of water in a food at a specified temperature to the vapor pressure of pure water at same temperature. The growth of pathogens in foods is markedly affected by different factors especially water activity and temperature (Romero et al., 2007). In all samples of this study, the water activity of hulls and kernels was above 0.9 a_w which is a high value and suitable for the growth of most pathogens. Our treatments did not have a significant effect on water activity in most cases. It can be seen in literature that sulfur is used in food technology as a preservative because it helps the quality of food to remain constant, meeting market requirements (Rossello et al., 1994). Consistently, any influence of sulfur treatments on water activity was not expected.

One of the important features of fruits is their varied and attractive colors. The color in many products is a sign of quality and marketability. Color change is one of the most prominent changes that occurs in many fruits and is often used as the main criterion by the consumer to distinguish fresh, ripe, or decayed fruit (Hernandez-Munoz et al., 2008). The CIELAB color space is a color space defined by the International Commission on Illumination (CIE) in 1976. It expresses color as three values: L* for the lightness from black (0) to white (100), a* from green (−) to red (+), and b* from blue (−) to yellow (+) (Goncalves et al. 2007). In the Munsell color system, hue (basic color), chroma (color intensity), and L* value (lightness) are three properties of color (Kuehni, 2002). Pistachio is presented in markets mainly as shelled dried kernels, but for a short time of the year, it can also be presented as whole fresh fruit after harvest. Therefore, the color of both the hulls and kernels are important.

Pistachio hulls have a color spectrum from red to green. The main difference between red and green hulls is the presence of anthocyanins, especially cyanidin derivatives, in the former ones (Ersan et al., 2016). Pistachio cultivars show specific hull colors. In this study, 'Fandoghi' had the highest lightness and hue values of hulls while 'Ahmadaghaei' had the highest chroma value of hulls. All sulfur treatments decreased hull lightness in 'Fandoghi' although, some of them increased this trait in 'Akbari' and 'Ahmadaghaei'. Reduction of hull chroma by sulfur was also observed in some cases in all cultivars. Hue value of hulls which is the basic color of them was not affected by treatments but only by cultivar. High concentrations of SO_2 can cause bleaching or discoloration in fruits which is an undesirable effect. SO_2 interacts with the membranes, making it leaky to solutes. Also, SO_2 directly reacts with anthocyanins rendering them colorless (Lichter et al., 2000). In figs for example, 25 μL/L SO_2 affected the color of fruits to the lowest extent but, higher concentrations caused fruit bleaching (Cantin et al., 2011). Sulfur-treated Washington peach fruits appeared to have dramatically inferior color and eye appeal when compared with fruit treated with combinations of chlorothalonil, captan, and benomyl (Drake, 1984). Blake peach fruit from sulfur-treated plots seemed to have inferior color compared with fruit in captan-treated plots (Scherm and Savelle, 2001). Increased hull lightness in 'Ahmadaghaei' and 'Akbari' is probably the result of bleaching but, its reduction in 'Fandoghi' may be the result of membrane damage and browning.

Color indicators of the testa of kernels were not affected by treatments in 'Fandoghi' and 'Akbari'. This result shows that pesticides and sulfur do not efficiently reach the kernels. But, in 'Ahmadaghaei', refinery micronized sulfur at both concentrations increased the testa lightness and hue values and decreased chroma value (similar to hulls of this cultivar). It can be assumed that R30 and 60 could penetrate to testa in 'Ahmadaghaei' and bleached it making it lighter with lower color intensity (chroma) and changed its basic color (hue). The reason is probably the softer texture of hull in

'Ahmadaghaei' which breaks more easily during harvest practice. Sulfur penetrates probably through hull cracks but not through its intact texture.

lower degree of fruit appearance and general acceptance of 'Ahmadaghaei' in control and fungicide treated samples is probably due to the softer texture of hulls in this cultivar which makes it more sensitive to mechanical and microbial damages. It can be assumed that sulfur treatments helped keeping its appearance through prevention of microbial disorders. Some research has reported the negative effects of SO_2 on the taste and smell of fruits such as litchi (Sivakumar et al. 2010). But in blueberries, no off-flavor was developed after SO_2 fumigation (Cantin et al., 2012). The effect of sulfur on sensory indicators is dependent on factors such as concentration, time of exposure and the texture of plant tissues. If pistachio hulls are intact, sulfur most likely does not pass to the kernel. We think that the loss of taste and aroma caused by some treatments is due to the hulls not the kernels. Because our experiment was done by fresh pistachio with sulfur residues on their hull surface, we speculate that this off-flavor will be resolved after drying of kernels.

Ethylene produced from sulfur dust-treated bean plants showed two stages of stimulation in shoots, one preceding full bloom and one preceding fruit-set. Likewise, the earliness of flowering was enhanced. The authors concluded that a slow oxidation of elemental sulfur in air produces SO_2 which greatly enhances ethylene evolution from leaf tissues (Recalde-Manrique and Diaz-Miguel, 1981). Alfalfa seedlings exposed to 0.7 μL/L SO_2 for 8 hours by fumigation, elevated ethylene production 10 times greater than control plants (Peiser and Yang, 1979). Ethylene production in tomato was enhanced by 1 h SO_2 fumigation and more increased by prolonged exposure up to 4 h. Ethylene synthesis induced by SO_2 fumigation was by way of ACC pathway. The authors claimed that ethylene might play an important role in SO_2-induced plant injuries at relatively short terms of SO_2 fumigation (Gong Young et al., 1995).

In wheat, the respiration rate increased in in seedlings fumigated by 0.06 ppm SO_2 for 8 h daily from germination to grain maturity. The authors concluded that the series of reactions leading to detoxification of SO_2 are ATP mediated which is provided by respiration and hence respiration rate increased at SO_2 treated plants (Agrawal and Deepak, 2003). We can assume that R30 and 60 caused a probable slight toxicity to hulls, which resulted in respiration elevation. In the contrary, It has been reported that SO_2 dramatically inhibits the respiration rate of longan fruit during storage (Pang et al., 2007). At a SO_2-polluted site, all species of lichen show a decrease in respiration rates with time (Haffner et al., 2000). But, in the toxitolerant lichen species (*Lecanora conizaeoides*) respiration was unaffected by SO_2 pollution (Gilbert, 1970).

Conclusion

Since pistachio psylla has become relatively resistant to chemical pesticides, the growers have started to use alternative ways, one of them is sulfur spraying. The efficacy of sulfur on controlling pistachio psylla has been reported in literature and is also documented by empirical evidence. This is critical for a pesticide to have no negative effect on the food quality and human health. Our results indicated that sulfur does not efficiently penetrate into pistachio kernel which is the edible part of fruit. Because sulfur did not affect the color of kernel testa in two cultivars. Sulfur application resulted in good effects on fruit appearance in one cultivar but, some negative effects on taste and aroma in the same cultivar, although two other cultivars were not affected. The effect of sulfur on acceleration of fruit ripening was obvious in one cultivar which could be related to the elevation of ethylene production caused by high concentration of sulfur in the cultivar. Refinery micronized sulfur enhanced the respiration rate in one cultivar. Chemical pesticides had some adverse effects on fruit quality in some cases, for example, confidor reduced kernel moisture in 'Fandoghi', hull firmness in 'Akbari' and fruit appearance in 'Ahmadaghaei' cultivars. In general, both kinds of sulfur did not influence the quality of fruit severely and can be used as an alternative to chemical pesticides which are not as effective as they were in the past.

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Conflict of interest

The authors indicate no conflict of interest for this work.

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