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Application of *Ocimum basilicum* Essential Oil as Vapor on Postharvest Storage of Plum Fruit cv. 'Golden Drop'

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Abstract

Increased interest in the use of natural compounds instead of chemicals is due to concerns about the effect of synthetic ingredients on humans' health and over environment. Therefore, in this study essential oil from *Ocimum basilicum* as a natural and safe compound, was applied at three levels (100, 200 and 300 µl/l) as vapor and its effects on postharvest quality and storage life of 'Golden Drop' plums was evaluated. After application of treatments, the fruits were stored at +1 °C and 80-85% relative humidity for 42 days. During the storage period, samplings were carried out every week and to simulate market condition, they were kept at room temperate for 24 h. Then some of the qualitative and quantitative traits, such as total soluble solids (TSS), titrable acidity (TA), TSS/TA ratio, weight loss, firmness, ascorbic acid, total antioxidants, as well as color (L*, hue angle) were measured. Results showed that the basil essential oil contributed to a better maintenance of TSS, TA, TSS/TA ratio, firmness, ascorbic acid, antioxidant and delayed weight loss and color changes compared to control. However, the fruit treated with 300 µl/l concentration had essence flavor compared to control. In conclusion, the use of basil essential oil is an effective tool on maintaining postharvest quality and storage life of plum fruits.

Keywords: antioxidant, color, firmness, natural compounds, weight loss

Introduction

Ripening is the process by which fruits attain their desirable quality, flavor, color, palatable nature and other textural properties. Ripening is associated with changes in composition i.e. conversion of starch to sugar. On the basis of ripening behavior, fruits are classified as climacteric and non-climacteric fruits. Ripening of climacteric fruits, of which plums are included, is handled by ethylene which causes morphological, physiological and biochemical changes regarding ripening process, such as skin color, sugar and organic acid metabolism and fruit softening (Valero *et al.*, 2005). During fruit ripening and softening, storage and shelf life of plum fruits decreases. Plum is a highly perishable fruit and will soon deteriorate after ripening. Its storage life is also limited even at low temperatures.

In recent years, consumer demands for food products that are free of synthetic chemicals residues, or products that are grown and supplied in an organic way, are significantly increasing. By using natural compounds such as plant essential oils, as non-destructive method, controlling decays and increasing storage life of plums is possible. Numerous papers have been published which report the use of natural compounds instead of chemicals, which in most cases have been associated with good results.

Essential oils are secondary metabolites that are produced in various parts of aromatic plants. They are volatile compounds that are called volatile oils or essential oils due to evaporation at normal temperatures. They are colorless compounds that get a dark color over time due to oxidation. Lipophilic characteristic of the essential oils is very important in inhibiting the growth of pathogens (Lanciotti *et al.*, 2004). Antioxidant characteristic of essential oils causes the reduction of enzymatic browning and increase storage life of fruits and vegetables, without loss of quality (Lanciotti *et al.*, 2004; Ponce *et al.*, 2004).

Combination of mentol, eugenol or thymol with modified atmosphere packaging (MAP) maintained the quality of table grape during storage, since reduced caused weight loss and color change, delayed the increase in TSS/TA ratio and reduced fruit softening (Valverde *et al.*, 2005). These treatments caused reduction of rachises and berries decay rate. Also, yeasts and fungi were significantly reduced in packages containing grape and natural antimicrobial compounds. The results contributed to maintenance of table grape quality and safety for longer storage period. Also, the use of methyl jasmonate treatment prevented fungal growth in grapefruit (Droby *et al.*, 1999), reduced decay and maintained the postharvest quality of papaya (Gonzalez-Aguilar *et al.*, 2003) and prevented microbial contamination of fresh-cut celery and peppers (Buta and Moline, 1998).

Thus, based on the above mentioned points, the aim of this study was to assay a potential use of basil essential oil as vapor on maintaining qualitative and quantitative characters and storage life of 'Golden Drop' plums.

Materials and methods

Plum fruits (*Prunus salicina* Lindl. var. 'Golden Drop') were harvested at commercial maturity from horticulture research center of the University of Tehran, located at Karaj, Iran. Plum fruits of uniform size and free from visual symptoms of disease or blemishes were used for the experiment. The fruits were transported to the laboratory immediately after harvest and were randomly selected for different treatments. In the laboratory, 6 plums (average mass of approximately 270 g) were placed in separate plastic bags. *Ocimum basilicum* essential oil (purchased from commercial company) at concentrations of 100, 200 and 300 µl/l was applied on filter papers and placed in each plastic bag for expose plums to vapor (without any direct contact between plums and filter paper). Control samples were handled similarly with the exception of the volatile treatment. Treated and untreated fruits were stored at +1 °C and 80-85% relative humidity for 6 weeks. During the storage period, 12 sampling bags were carried out every week to simulate market conditions, being kept at room temperate for 24 h.

Gas Chromatography-Mass Spectrometry analysis of the essence

The analysis of the volatile constituents of the essential oil were run on a Hewelt-Packard GC/MS system (GC: 6890; MS: 5973). The fused- silica hp INNOWAX capillary column (30 m × 0.25 mm ID, film thickness of 0.32 µm) was directly coupled to the MS. The carrier gas was helium, with a flow rate of 1 mm/min. Oven temperature was programmed (60 °C for 3 min, then 60-220 °C at 5 °C/min) and subsequently, held isothermal for 2 min. Injector temperature: 250 °C, detector temperature: 300 °C. Split ratio 1:20.

Volume injected was 0.1 µl of 1% solution (diluted in hexane). The mass spectrometer was hp recording at 70 eV; scan time 1.5 sec; mass range 40-300 amu. The components of the oil were identified by comparison of their mass spectra with those of a computer library (Wiley 275 library). Retention indices were calculated using retention times of n-alkanes that have been injected to the same instrument (Adams, 1995; Shibamoto, 1987).

Total soluble solids content, acidity and TSS/TA ratio determination

The soluble solids content (TSS) of juice was determined using a digital refractometer and was expressed as percent soluble solids content. Titrable acidity (TA) was determined by titration against 0.1N NaOH up to pH 8.2 by using 5 ml of juice diluted to 50 ml with distilled H_2O.

The results were expressed as g of malic acid per 100 g fresh weight. The TSS/TA ratio was calculated by dividing TSS with the corresponding TA value.

Weight loss and fruit firmness

Weight loss percent was determined by the following formula: (A – B)/A x 100, in which A is the fruit weight just before storage and B is the fruit weight after storage period. Fruit firmness was determined by using a penetrometer fitted with a 5 mm tip. A small slice of fruit skin was removed from each side of a fruit, three fruits from each replicate were used for firmness measurements and results were expressed as Newton (N).

Fruit color

The changes in fruit color parameters including L^*, a^* and b^* were recorded at opposite sides of skin surface using three fruits from each replicate with a Minolta Chromameter CR400 then expressed as L^*, hue angle ($h° = 180 + \tan^{-1} b^*/a^*$, if $a^* < 0$) (Pek *et al.,* 2010; Fernando *et al.,* 2007).

Ascorbic acid

The level of ascorbic acid was determined using 2,6-dichlorophenol indophenol method as described by A.O.A.C. (1994).

Total antioxidant activity

Total antioxidants in fruits' pulp tissue were estimated by using the method of Faniadis *et al.* (2010) with some modifications. One gram of fruit pulp which had already been lyophilized with liquid nitrogen and stored at -80 °C was homogenized in a glass pestle and mortar, using 8 ml of 80% methanol. Then the mixture was shaken slowly for 10 min in the cold chamber and centrifuged for 15 min at 12000 rpm in 4 °C. Thereafter the supernatant was filtered through filter paper at cold chamber. Stock solution of 2,2-diphenyl-1-picrylhydrazyl (DPPH) 100 µM was made by dissolving 0.00394 g DPPH in 100 ml of methanol. In the dark room and inside the glass cuvette, 1700 µl of DPPH solution, 1000 µl of distilled water and 100 µL of methanol extract were combined and put in the darkness for 2 h. Then the absorbance was measured at 520 nm against methanol (as blank). Different concentrations of ascorbic acid (as standards) were used for determining the antioxidant capacity of samples.

Ascorbic acid stock solution (1 mM, 0.0176 g ascorbic acid in 100 ml 80% methanol) was prepared and from this stock other concentrations (0.125 mM, 0.25 mM and 0.5 mM) were produced to determine a standard curve. The absorbance was measured at 520 nm by using a spectrophotometer. According to standard curve and based on absorbance of the samples, the antioxidant capacity was determined and expressed as mg of ascorbic acid equiv. 100/g FW (A.O.A.C. 1994).

Statistical analysis

Statistical analysis of the data obtained in the present study was carried out using split factorial method in a completely randomized design layout, with two factors, including basil concentrations and storage period. All treatments were replicated for three times.

Means comparison was performed using Duncan's multiple range test to examine if differences between treatments and storage time were significant at $p \leq 0.05$. All analyses were performed with SAS software package 9.1 for Windows.

Results and discussion

Results obtained by GC-MS analysis of the essential oil of *O. basilicum* are presented in Tab. 1. Eighteen compounds were identified in the essential oil of *O. basilicum*. As a result of GC-MS analysis, *O. basilicum* contained estragol (71.85%) as the major compound.

TSS, pH, TA and TSS/TA

During the storage, amount of TSS decreased gradually (Tab. 2). Using basil oil had a significant effect on TSS, so that the TSS was lower in treated fruits than in control fruits

Tab. 1. Chemical composition of the *O. basilicum* essential oil

No.	Chemical composition	Percentages of compounds
1	Estragol (Methyl Chavicol)	71.85
2	Linalool	22.24
3	Eugenol	1
4	α-Thujene	0.11
5	Myrcene	0.7
6	β-Pinene	0.6
7	Methyl Eugenol	0.6
8	Cis-Bergamotene	0.40
9	Caryophyllene oxide	0.40
10	Linalool oxide	0.24
11	1,8- Cineol	0.21
12	α-Pinene	0.2
13	Citronellol	0.20
14	Dihydro Linalool	0.18
15	α-Terpinene	0.1
16	ρ-Cymene	0.1
17	γ-Terpinene	0.1
18	β-Gurganene	0.08

(Fig. 1). Similarly, Ju *et al.* (2000), on the Chinese pear, showed that essential oil maintained the TSS compared to control by retarding the ripening process.

Among the metabolic activities, respiration is aN important process in which the organic acids may be used as substrate and following that TA to be decreased during the storage (Tab. 2). The most important organic acid in plum fruit is malic acid and it decreased during storage. Essential oil caused the maintenance of TA with respect to control (Fig. 2). Serrano *et al.* (2005) reported that essential oils treatments were effective on retarding the TA loss of cherry compared to control.

Over storage time TA decreased with the process of ripening, while TSS increased and subsequently TSS/TA ratio increased (Tab. 2). The lower TSS/TA ratio in treated plums was due to lower accumulation of sugars and higher level of TA. The treated fruits had less ripening degree, which was due to lower TSS and retarded ripening, compared to control fruits (Fig. 3). It seems that essential oil by lowering respiration process and retarding of ripening has caused reduction in consuming of carbohydrates and organic acids.

Weight loss and fruit firmness

Moisture loss is driven by a difference in water vapor pressure between the product surface and the environment. Weight loss is the result of water evaporation from the surface of fruits during the storage (Tab. 2). The product surface may be assumed to be saturated, and thus, the water vapor pressure at the commodity surface is equal to the water vapor saturation pressure evaluated at the product's surface temperature. However, dissolved substances in the moisture of the commodity tend to slightly lower the vapor pressure at the evaporating surface (Sastry *et al.,* 1983).

In this research, weight losses for treated fruits were lower than in control and the lowest weight loss was found with the concentration of 300 μl/l of basil oil (Fig. 4). This shows that essential oil has caused reduction in the weight loss process, yet its mechanism is unclear (Valverde *et al.,* 2005).

Reduction in respiration might have been the reason for this effect of essence.

Fruit firmness deceased over time (Tab. 2). Changes in firmness are due to changes in the chemical structure of the cell wall, because during the ripening process polygalacturonase and pectin methylesterase enzymes cause dimetylation of galacturonic acid from cell wall pectin and Ca^{2+} gets released in the polymer chains, resulting the softening of the cell walls (Perasanna *et al.,* 2007; Wei *et al.,* 2010). Fruit firmness in treated fruits was better maintained during storage compared to control fruits (Fig. 5).

Softening contributes to quality loss by reducing shelf life, but the addition of basil essential oil resulted in higher flesh firmness during cold storage. Similar effect has been reported for sweet cherry (Serrano *et al.,* 2005) and table grape (Valverde *et al.,* 2005; Valero *et al.,* 2006) which might be due to lower respiration rate.

Ascorbic acid and total antioxidants activity (TAA)

The ability of antioxidants to scavenge reactive oxygen species (ROS) is important to protect tissues from light-induced oxidative damage. The content of ascorbic acid and TAA decreased during storage (Tab. 2). Fruits treated with basil essence better maintained the ascorbic acid content compared to control (Fig. 6). Ascorbic acid is antioxidant and there is positive correlation between its amounts and the level of TAA, which was better maintained in treated fruits than control (Fig. 7).

The rate of senescence process, which is the most important factor in reducing the quality and storage life of the fruit, is a result of free radicals activities. High levels of TAA in the product causes reducing of senescence process (Arora *et al.,* 2002). This result is similar to that reported for table grape (Valero *et al.,* 2006).

Color changes

Color is a very important indicator of the quality of fresh fruit. It also serves for estimating the stage of maturity of fruits. Among plant pigments responsible for the color of fruits are anthocyanins. L^* shows the darkness or brightness of the fruit color, in a way that reduced L^* coincides with the darkening of fruits (James *et al.,* 2002). L^* decreased with the time and the fruits get darker during the ripening process in the storage (Tab. 2). L^* in treated fruits was higher than control during the storage and there was no significant difference in L^* between different basil concentrations (Fig. 8). Although the essential oil mechanism in maintaining L^* is unknown (Serrano *et al.,* 2005), however retarding of ripening might be the main factor. Maintenance of L^* in treated fruits can also be related to reduction of weight loss (Valverde *et al.,* 2005). Different pattern of surface discoloration of fruits (such as strawberries) is due to differences in the concentration and ratio of various phenolic compounds (Zhang *et al.,* 2008).

The hue gradually decreased during storage (Tab. 2). Fruits treated with essential oil had the highest hue angle and were greener than control fruits and with increasing the concentration, fruit color change occurred slower (Fig. 9). This has also been reported in other fruits, such as table grape (Martínez-Romero *et al.,* 2003), loquat (Amorós *et al.,* 2008) under MAP conditions and sweet cherry with essential oils (Serrano *et al.,* 2005).

Tab. 1. Chemical composition of the *O. basilicum* essential oil

Treatment	Mean of Parameter								
Storage period (week)	TSS (%)	TA (%)	TSS/TA ratio	Weight loss (%)	Firmness (N)	Ascorbic acid (mg/100g)	Antioxidant activity (mg/100g)	L*	Hue angle
0	8.00g	2.42a	3.31g	0.00g	3.73a	7.93a	49.86a	67.86a	110.17a
1	8.30f	2.24b	3.71f	0.70f	3.54a	7.50b	47.31b	65.01b	108.44b
2	8.43e	2.08c	4.08e	1.03e	2.59b	7.17c	45.67c	63.61c	107.02c
3	8.56d	1.94d	4.48d	1.25d	2.48bc	6.86d	43.79d	62.40d	105.60d
4	8.73c	1.81e	4.93c	1.44c	2.39bc	6.60e	42.65e	60.19e	103.65e
5	8.87b	1.62f	5.60b	1.62b	2.20cd	6.24f	39.75f	58.26f	102.66e
6	9.05a	1.43g	6.41a	1.81a	2.05d	5.93g	38.09g	55.70g	100.91f

Note: Means of treated and untreated fruits at columns with at least one common letter did not show significant difference at 5% level, using Duncan test

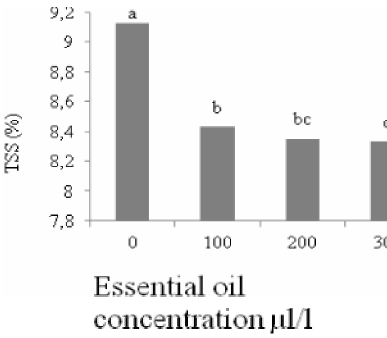

Fig. 1. The effect of basil essential oil treatment on TSS of 'Golden Drop' plums

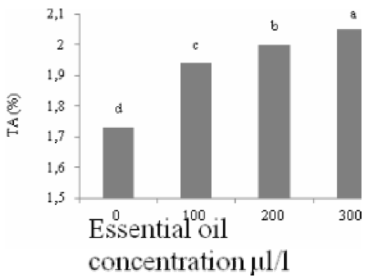

Fig. 2. The effect of basil essential oil treatment on TA of 'Golden Drop' plums

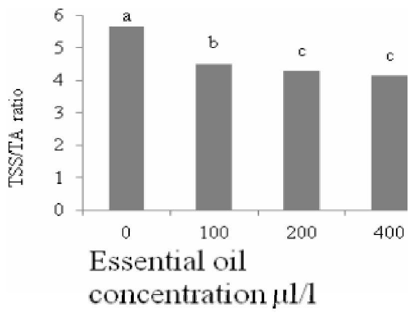

Fig. 3. The effect of basil essential oil treatment on TSS/TA of 'Golden Drop'

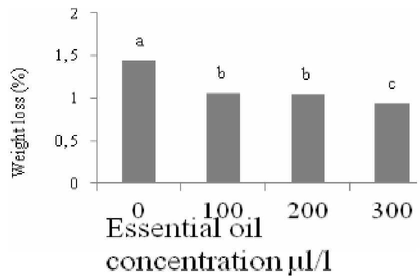

Fig. 4. The effect of basil essential oil treatment on weight loss of 'Golden Drop'

Note: All data in figures are the mean of all the measurements per factor in all sampling dates

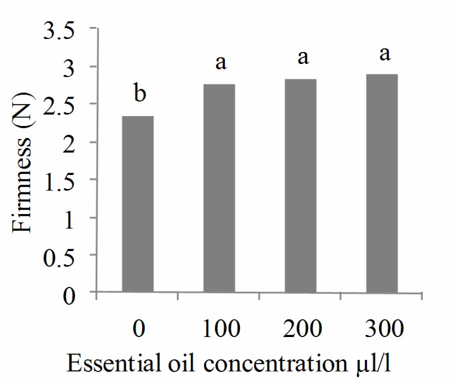

Fig. 5. The effect of basil essential oil treatment on firmness of 'Golden Drop' plums

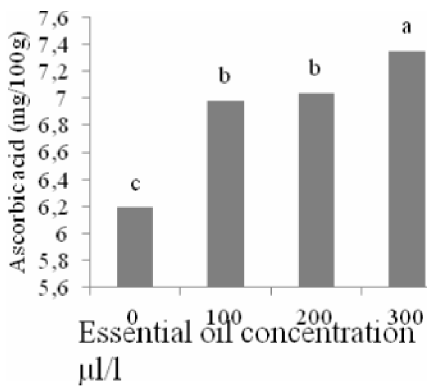

Fig. 6. The effect of basil essential oil treatment on ascorbic acid of 'Golden Drop' plums

Note: All data in figures are the mean of all the measurements per factor in all sampling dates

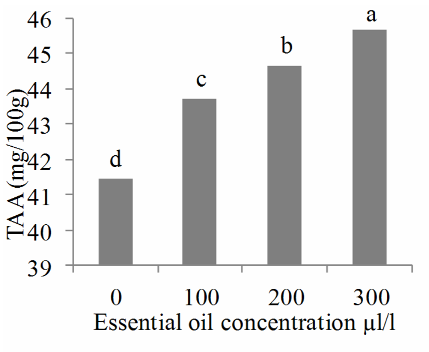

Fig. 7. The effect of basil essential oil treatment on TAA of 'Golden Drop' plum

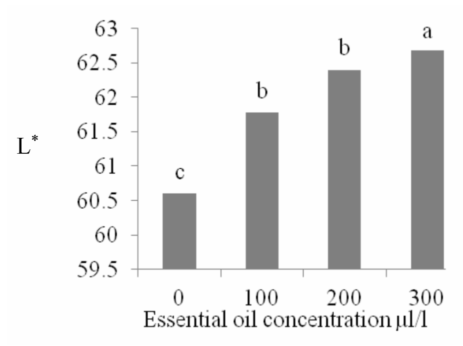

Fig. 8. The effect of basil essential oil treatment on L^* of 'Golden Drop' plum

Note: All data in figures are the mean of all the measurements per factor in all sampling dates.

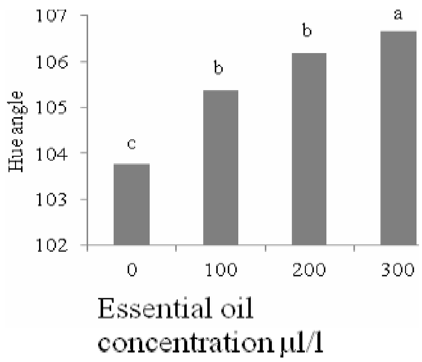

Fig. 9. The effect of basil essential oil treatment on hue angle of 'Golden Drop' plum

Note: All data in figure are the mean of measurement hue angle in all sampling dates

Conclusions

Many efforts in the storage, distribution and sale of horticultural products have leaded to the global supply of high-quality fresh products to consumers, to respond to the consumers' demands for healthy products, avoiding the application of chemicals as a mean of preservation. In this study, basil essential oil was used as a safe and natural compound for increasing the storage life and maintaining the qualitative and quantitative characteristics of *Prunus salicina* var. 'Golden Drop'. Treatments with 100, 200 and 300 µl/l of basil essence resulted in improved quality of the plum fruits during storage. However, the fruit treated with 300 µl/l concentration had essence flavor compared to control. Further studies are needed for a better understanding of the essential oils mechanism on physiology of the fruits and their ripening process.

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