Effects of Ozonated Water on Storage Life and Postharvest Quality of Iranian Table Grape (cv. Bidaneh Qermez)

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Abstract

Grape as a perishable fruit is susceptible to fungal infection especially *Botrytis cinerea,* as an agent of gray mold, and berry dehydration. There are limitations in use of synthetic chemical compounds to control postharvest losses in grape and other fruits. The aim of this study was the evaluation of non-chemical and safe methods for this purpose. Therefore, ozone soluted at 0.3 ppm concentration with three treatment times (5, 10 and 15 min), in water was applied on 'Bidaneh Qermez' grape cultivar. Qualitative characteristics (decay, vitamin C, reducing sugars, weight loss, berry abscission, dehydration, cracking and sensory analyses) were evaluated. The results showed that treated fruits with ozone had lower decay percentage, weight loss, berry dehydration, abscission, cracking and higher reducing sugars and storage quality compared to controls. As a general result, ozone treatment caused lower decay incident and longer storability.

Keywords: Ozone treatment, Maintain quality, Shelf-life, Table grape

1. Introduction

Quality loss is a normal process during storage, but the level of degradation depends on storage conditions and the technologies used (Hulea, et al., 1982; Mari, et al., 2003). Currently, arrays of chemical treatments are used to preserve fresh produce. However, there are growing health and environmental concern over current practices, due mainly to the risk of generating potentially harmful by-products and residues (Spotts and peters, 1980) which are potential carcinogens; therefore it has created the need to investigate the efficacy of non-traditional sanitizers and other alternative technologies (Selma et al., 2008). One candidate is ozone (Palou et al., 2003; Rice, 1999), a tri atomic oxygen. It is effective in killing microorganisms through oxidation of their cell membranes (Cataldo, 2003). An expert advisory panel asserted the determination that ozone qualified for listing as a Generally Recognized As Safe (GRAS) material (Graham et al., 1997). Ozone has a wide range and a long history as a water disinfectant. Short-term fumigation with 2500 or 5000 $\mu ll^{-1} \times$ h ozone reduced postharvest gray mold of grapes in storage by approximately 50% (Mlikota Gabler, et al., 2010). Ozone storage treatment method decreased *Botrytis Cinerea* contamination of kiwifruits (Barboni, et al., 2010). Comparing with gaseous ozone, treatment of ozonated water was more effective on reduction of aerobic mesophilic bacteria count. Total mold

count was decreased by 1.7 mgl⁻¹ in ozonated water at 7.5 min. and molds were completely inactivated at 15 min (Zorlugene, et al., 2008). The purposes of this study were to evaluate the effects of ozonated water on the storability and quality-related attributes of 'Bidaneh Qermez', the famous Iranian table grape.

2. Material and Methods

2.1 Plant material

Table grapes (*Vitis vinifera* L.), cv. 'Bidaneh Qermez' were harvested randomly from a commercial vineyard located in Takestan, Qazvin, Iran at commercial maturity stage and transported to the laboratory. Grape clusters were divided into small clusters of approximately 300g each, those with defects were discarded. Samples were taken every 15 days up to the end of experiment (105 days) and fruits were evaluated at each sampling time after keeping them a day at room temperature.

2.2 Ozone treatment

Ozone generator (Model MHP1H, France) was used to produce 0.3 ppm ozone concentration in water. The current Threshold Limit Value-Short Term Exposure Limit (TLV-STEL) for ozone is 0.3 ppm for 15 min as recommended by the American Conference of Governmental Industrial Hygienists (ACGIH) and approved by federal Occupational Safety and Health Administration (OSHA) (Xu, 1999). The output of ozone generator in water was monitored with ozone test kits for ozone concentration. Grape clusters (300g) were dipped into ozonated water for 5, 10, 15 min prior to storage and air dried. Then were placed in polyethylene bags and stored for 105 days at 4°C and 75% Relative Humidity (RH). Control fruits were dipped into non-ozonated water for 5, 10, 15 min and stored in the same conditions as above samples. All the experiments with O_3 were made in the laboratory of Science and Research Branch at Azad University (Tehran, Iran). Measurements were made at room temperature.

2.3 Weight loss percentage

The effect of ozone exposure on table grape weight loss was also investigated. Weight of individual bunches was recorded at the beginning of harvest and different sampling times and expressed as percentage of original weight (Valero, et al., 2006).

2.4 Color changes

Color changes of berries were analyzed on the skin surface with a Minolta Chromameter CR400, JAPAN. Color was measured at the further two points apart on the equator of each berry and expressed as Chroma (C = [(a2+b2)0.5]) and Hue angle (H° = tan -1 (b*/a*)) (Valero, et al., 2006).

2.5 Vitamin C content

The content of vitamin C was determined using indophenol procedure. 10 ml of samples were filterated and titrated against sodium 2, 6-dichlorophenol indophenol dye to a faint pink color which persisted for 5-10 seconds. It was expressed as mg vitamin C/100g fruit weight (Titer × dye equiv. × dilution ×100/ Wt. of sample) (Saini et al., 2006).

2.6 Reducing sugars content

Reducing sugar was measured with the procedure of Lane-Eynon. Mixed Fehling's solution was made by transferring 5 ml of solution A and 5 ml of solution B to a 250 ml conical flask and mixed well. Diluted fruit juice added to mixed Fehling's solution drop by drop. Then heated over Bunsen burner and boiled for two minutes and then three drops of methylene blue indicator was added (Saini et al., 2006).

2.7 Decay analysis

Percent of decay was scored on a 1-9 scale, where: 0= intact fruit, 1= less than 10 % Decay, 2= between 10-20 % decay, 3= between 20-30 % decay, 4= between 30-40 % decay, 5= between 40-50 % decay, 6= between 50-65 % decay, 7= between 65-80 % decay, 8= more than 80% decay (Nigro, et al., 2000).

2.8 Berry abscission, cracking and dehydration of rachis

Berry abscission was scored on a 1-5 scale, where: 1= very low, 2= low, 3= moderate, 4= high, 5= very high. For the rachis, symptoms of dehydration and browning for primary and secondary branches were evaluated on a ranked scale of 1-5, where= 1= green and fresh, 2= green, 3= semi-dry, 4= 50% dry, 5= absolutely dry (Valero, et al., 2006; Xu, et al., 2007). Number of cracked berries in 1kg were evaluated (Xu, et al., 2007).

2.9 Sensory evaluation

Sensory analyses to compare the quality of treated and control table grapes were carried out by a 10 trained adults aged 25-40 years. It was about aroma, taste, firmness, appearance and texture. Panelists scored grapes

between 1-10. 10 being the best total quality and 1 being the worst (Martinez-Romero, et al., 2007). Samples were scored for overall quality by using an interval hedonic scale. Assessments were continued until fruits condition were considered unacceptable.

2.10 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 3 replicates. Data obtained were subjected to analysis of variance (ANOVA). Means were separated by SNK and LSD test.

3. Results and Discussion

3.1 Decay analysis

During the storage period, the incident of berry decay was increased (Table 1). The controls showed 2.56 % of decay while it was just 1.12% for ozone treated samples. Increasing of treatment time with ozone from 5 to 15 min resulted in an increased resistance to decay (Fig. 1). Use of ozone with 2500 and 5000 μLL^{-1} concentrations could control *Botrytis cinerea* in grapes up to 50% (Mlikota Gabler, et al., 2010). Use of ozone with 5000 mgL^{-1} concentration in strawberry has been effective in reducing fungal and yeast infections (Allende, et al., 2007). Ozone can be effective in stimulating the production of phytoalexins within the plant, which increase the plant's resistance to decadence accordingly and have high antioxidant role (Sarig, et al., 1996).

3.2 Reducing sugars content

While time elapsed, reducing sugars rate was increased form 13.92% at the harvest time to 17.05 in the 105th day (Table 1). A significant increase in reducing sugars rate was experienced in ozone treated samples compared to the controls. The mean difference between these two groups was 0.76. Result showed that increase in ozone treating period led to an increased reducing sugars rate (Fig. 2). Glucose and fructose (reducing sugars) of carrots which were treated by ozone 50 ± 10 nLL^{-1} increased slowly and linearly during a month storage (Hildebrand, et al., 2008). Sugars in tomatoes which were treated with ozone with concentrations of 0.15 and 1.0 $\mu molmol^{-1}$ increased significantly compared with controls (Tzortzakis, 2007). The amount of glucose in strawberries treated by ozone (156 μgm^{-3}) did not have any significant changes, but the amount of fructose in fruits decreased (Keutgen and Pawelzik, 2008). Increase in reducing sugars is a physiological response which works as a signal (Rolland, et al., 2006). Sugars have little antioxidant action but control antioxidant systems. It can be concluded that the increase in reducing sugars can enhance plant resistance against pathogens, because it can affect the total antioxidant content of fruit. In general, it has been shown that due to the high oxidative capacity of O3 and its ability to generate toxic agents, they elicit plant defense reactions such as the production of phytoalexins including resveratrol in grapes (Sarig et al., 1996; Allende et al., 2007).

3.3 Weight loss

Ozone treatment decreased the weight loss of samples noticeably. Weight loss of ozone treated samples was 0.06% less than that of control samples. Meanwhile the use of 15 min ozone treatment had a significant effect on samples water loss (Fig. 3). It seems the life of fresh grapes is considered under the influence of berries and cluster's weight loss. Sample weight loss happens due to water loss and the reduction in stored materials in the process of respiration. Ozone leads to increased dehydration of samples only if it is used in high concentrations which cause damage to epidermis and cuticle tissues of the fruit (Palou, et al., 2002).

3.4 Color changes

The Hue angle decreased with increasing storage duration (Table 1). The ozone treated samples had a lower H˚ compared to control samples (Fig. 4). The Chroma reduced in the storage period (Table 1). Ozone treatment led to a lower Chroma compared to that of the control samples (Fig. 5). It appears that when the samples experience weight loss and dehydration during the storage period, the phenomenon of browning berries occurs and cause more color changes in them. Reduction in the amount of green pigment over time is the main index of berry color purity reduction (Artes-Hernandez and Aguayo, 2004). It seems that in ozone treated samples more green pigments were reduced.

3.5 Berry abscission and dehydration of rachis

Ozone treated samples had a lower berry abscission in comparison to the controls. The most berry abscission in ozone treated samples was observed with 5 and 10 min ozone treatment (Fig. 6). Results showed that dehydration of rachis increased during the storage period (Table 1).   This increase was lower in ozone treated samples compared to controls. The 15-min ozone treated samples preserved the freshness of their rachis much better than other samples (Fig. 7). Grapes which were treated by 0.1 mgg^{-1} ozone for 20 to 40 min experienced

no damage to the rachis during the storage period (Sarig, et al., 1996). Table grapes, which were treated by 0.3 ppm ozone, had no damage to their rachis in the storage period (Palou, et al., 2002).

3.6 Berry cracking and fruit quality

Ozone treated samples experienced a less berry cracking compared to control samples, so that their mean difference was 212.2 kg. Berry cracking of the 15-min ozone treated samples was 20.5 kg less than other ozone treated samples (5 and 10 min) (Fig. 8). Because polyethylene package performs as a barrier against water vapor escape, it condenses the water vapor in the package atmosphere on the surface of the fruit and leads to re-absorption of water, this re-absorption could cause berry cracking. It seems ozone treating led to reduction in water loss. Appearance of ozone treated berries was free from all damages (Mlikota Gabler, et al., 2010).

The quality of the fruit was affected in the storage period, controls were infected in the first 15th days, accordingly we failed to compare them with the other samples in the 15th day, but those samples which experienced 15 min of ozone treatment preserved a better quality compared to other samples, but the difference was not statistically significant (Fig. 9). Most panelists chose the ozone treated (with concentration of 0.15 $\mu molmol^{-1}$) tomatoes regarding appearance and sensory evaluation (Tzortzakis, 2007).

According to our results, ozone treatment did not affect the vitamin C content in a significant manner (Fig. 10). Similar findings are reported in tomato fruits treated with ozone (Shalluf et al., 2007; Tzortzakis, 2007).

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Table 1. The mean of evaluated parameters during the storage period in table grape cv. 'Bidaneh Qermez'

| Time	Decay	Weight loss	Reducing sugars	Chroma (C)	Hue angle(H°)	Berry abscission	Berry dehydration	Berry cracking	Fruit quality
0	0e	0d	13.92f	9.47a	44.9a	0e	1d	0c	10a
15	1.17c	0.05b	14.37e	9.46a	41.5c	1.83c	2.5b	0c	8.6b
30	2.67b	0.13a	15.80d	8.28b	43.5a	2.78a	3.11a	6.4a	7.7c
45	0.78cd	0.09a	15.26c	7.19cd	42.7bc	1.67c	2.22bc	5.6c	7.3cd
60	2.7b	0.1a	15.96b	7.26c	41.9c	2.11bc	3.22a	10c	7c
75	2.83b	0.1a	15.95b	7.02cd	43.2ab	2.5ab	2.5b	28b	6.9d
90	0.67d	0.02c	16.85a	6.98cd	43.4ab	0.67d	2c	0c	-
105	4.33a	0.05b	17.05a	6.66d	39.7d	0.67d	2.33bc	0bc	-
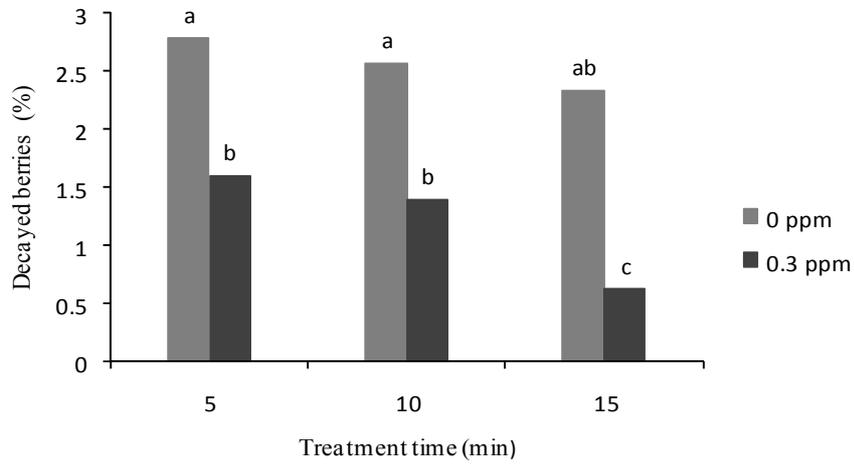
Figure 1. The effect of ozone treatment and treatment time on decayed berries of 'Bidaneh Qermez' table grape
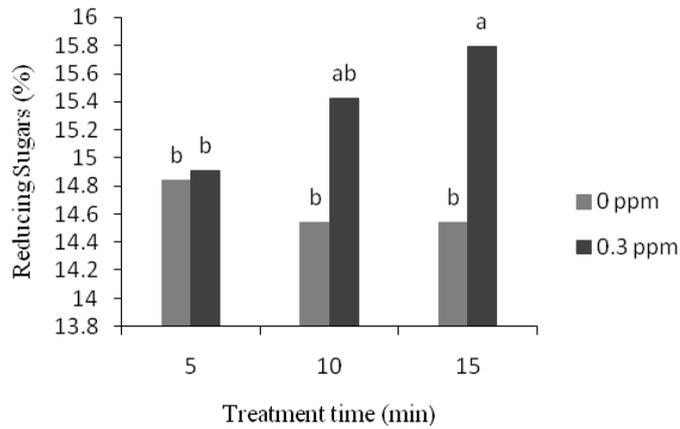

Figure 2. The effect of ozone treatment and treatment time on reducing sugars of 'Bidaneh Qermez' table grape
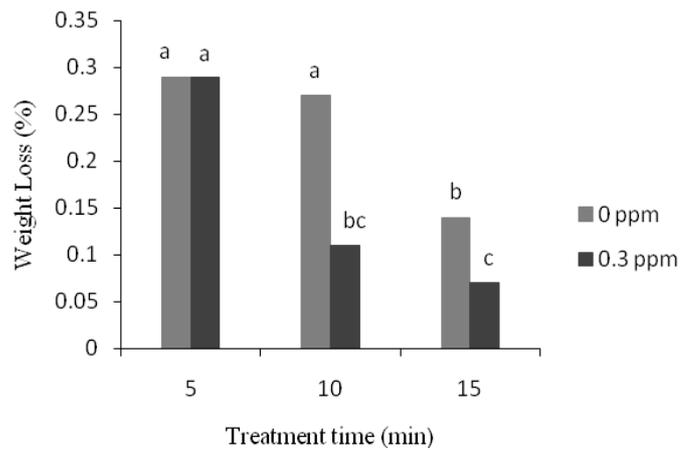

Figure 3. The effect of ozone treatment and treatment time on weight loss of 'Bidaneh Qermez' table grape
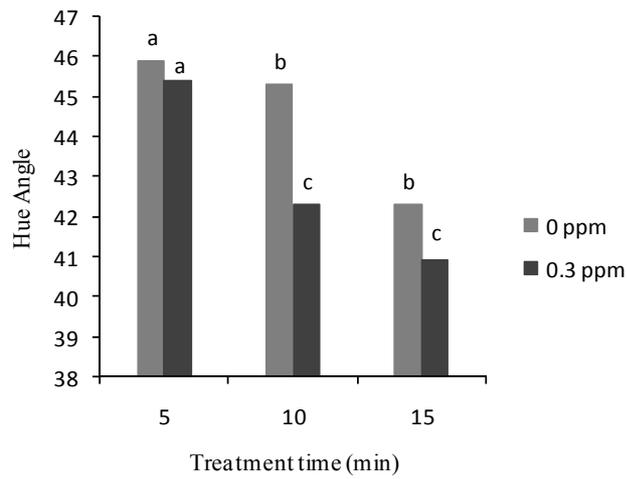
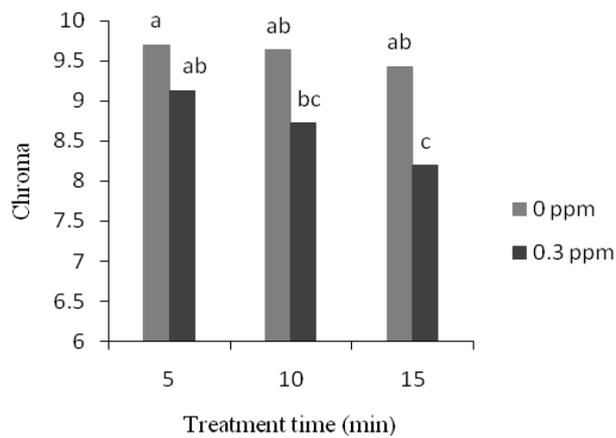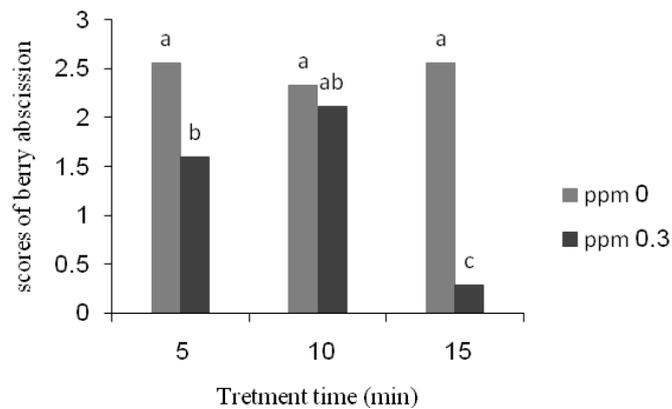Figure 4. The effect of ozone treatment and treatment time on hue angle of color in 'Bidaneh Qermez' table grape

Figure 5. The effect of ozone treatment and treatment time on chroma of color in 'Bidaneh Qermez' table grape

Figure 6. The effect of ozone treatment and treatment time on berry abscission of 'Bidaneh Qermez' table grape
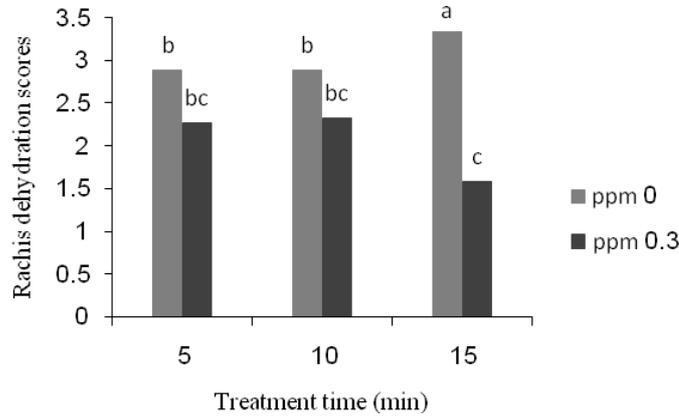
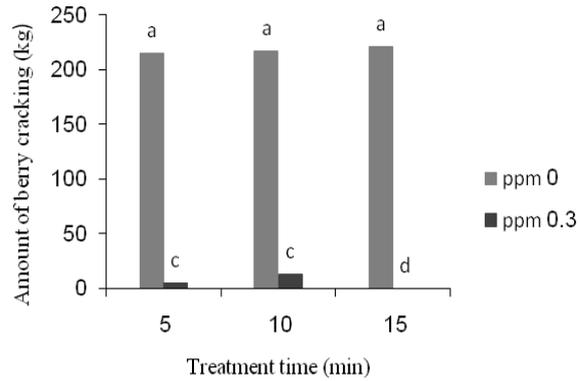Figure 7. The effect of ozone treatment and treatment time on rachis dehydration of 'Bidaneh Qermez' table grape

Figure 8. The effect of ozone treatment and treatment time on berry cracking of 'Bidaneh Qermez' table grape
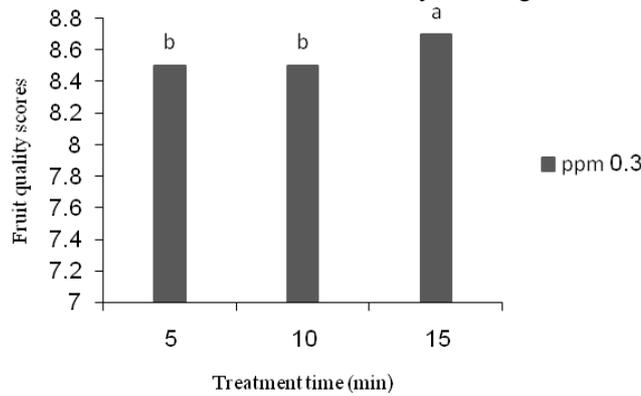

Figure 9. The effect of ozone treatment and treatment time on sensory analysis of 'Bidaneh Qermez' table grape
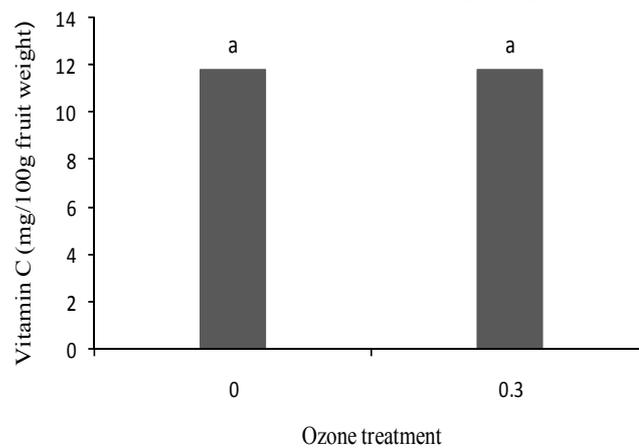

Figure 10. The effect of ozone treatment on vitamin C of 'Bidaneh Qermez' table grape