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RESEARCH ARTICLE

Impact of postharvest salicylic acid and jasmonic acid treatments on quality of "Crimson Seedless" grapes during cold storage and shelf life

Bassem N. Samra[1,2]

1. Department of Pomology, Faculty of Agriculture, Mansoura University, Mansoura 35516, Egypt

2. Department of Biology, Faculty of Science, Taif University, P.O. Box 888, Taif 21974, Kingdom of Saudi Arabia

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Corresponding Author

Bassem N. Samra

Abstract

The present study aimed to investigate the impact of postharvest on quality of Crimson Seedless" grapes by dipping in salicylic acid (SA) at 2, 4mM and jasmonic acid (JA) solutions at 5, 10 mM for 5 minutes at 22°C during cold storage and shelf life. The results showed that both of SA and JA treatments at different concentrations were significantly effective in reducing weight loss, berry decay, berry shatter and total loss in cluster weight percentages during cold storage period and shelf life as compared to the control. Also, the pervious treatments significantly reduced rachis browning, improved visual appearance of berries and increased berry firmness during cold storage period and shelf life incomparisonwith control. On the other hand, soluble solid content (SSC%), titratable acidity (TA%), SSC/acid ratio, berry separation force and total anthocyanin in berry skin were not significantly affected by SA and JA treatments during the previous period. Furthermore, the lowest values of weight loss, berry shatter, berry decay, total loss in cluster weight percentages, rachis browning and superior berry appearance were presented by SA at 4 mM and JA at 10 mM at the end of shelf life in comparison with other treatments. It could be concluded that SA at 4 mM and JA at 10 mM as a postharvest treatments are applicable for improving the storability of "Crimson Seedless" grapes and maintaining their quality during cold storage and shelf life.

INTRODUCTION

Grapevine (*Vitis vinifera*, L) is one of the most important fruit crops grown in the world. The cultivated area of grapevine in Saudi Arabia recently increased since, it reached about 13282 hectares producing 149847 tons according to FAO (2013). Crimson Seedless cultivar is a late-season, attractive, red seedless grape cultivar with firm berries. Also, "Crimson seedless" grape has superior eating characteristics, berry texture firm and crisp, and its flavor is excellent (Dokoozlian and Peacock, 2001). Table grape is one of the moderately susceptible fruits to decay and subject to serious water loss during postharvest handling, rachis browning, which occurs as a consequence of water loss (Crisosto et al., 2001). However, postharvest life of table grapes is relatively short due to water loss, skin browning, rachis dehydration and browning, berry shatter and fungal decay. In this respect, gray mold (*Botrytis cinerea*) is the most postharvest disease of table grapes especially in late season. Previously, using SO_2 during cold storage as a fumigant or generator is the most universal method to control fungal decay and also to maintain the table grape quality (Ga et al., 2003). Although SO_2 excellently controls fungal decay and prevents rachis browning, its residues are toxic and dangerous to human health. Recently, acetyl salicylic acid (ASA) is a derivative of salicylic acid (SA) play an important role on reducing fungal decay in harvested fruits, preventing postharvest

disease and disorders in horticultural crops (Ranjbaran et al., 2011). In addition, salicylic acid (SA) is a simple phenolic compound with a phytohormone–like function in plant growth and development. Also, SA activates the expression of several defense–related genes (Loake and Grant, 2007) and SA has been shown to inhibit ethylene biosynthesis and their action (Srivastava and Dwivedi, 2000; Zhang et al., 2003). Postharvest application of SA prolong storage life and reduced fruit softening rate of kiwifruit (Zhang et al., 2003), delayed ripening of harvest fruits (Srivastava and Dwivedi, 2000) and delayed discoloration with the inhibition of browning on grapes (Ranjbaran et al., 2011). Jasmonic acid and its volatile methyl ester, methyl jasmonate (MeJA), are a class of cyclopentanone compounds regarded as endogenous regulators that play an important role in regulating the stress response, plant growth, and development (Creelman and Mullet, 1997). Furthermore, MeJA has been applied to reduce the development of chilling injury symptoms on mango (Gonzalez-Aguilar et al., 2000). Moreover, MeJA has been shown to reduce decay and maintain the postharvest quality of papayas (Gonzalez-Aguilar et al., 2003).
In raspberry, Wang and lin (2000) demonstrated that MJ increased the resistance of tissues against decay by enhancing their antioxidant system and their free radical scavenging capability and there is a positive correlation between antioxidant activity and total phenolic or anthocyanin content. Little or no attention has been given to apply salicylic acid and jasmonic acid treatments on quality of crimson seedless grapes during cold storage and shelf life. Therefore, the aim of this study was to investigate the impact of postharvest salicylic acid (SA) and jasmonic acid (JA) on quality of crimson seedless grapes during cold storage at 0°C and shelf life at 20°C.

Materials and Methods:

Plant materials and experimental procedure:
Harvest date was carried out on the 15th of September during season 2014 when berries reached full colour and soluble solids content in berry juice were about 16-18 %, from vines received common horticultural practices, undamaged berries, free from any obvious pathogen infection and the existence of healthy greenish rachis, then transported to the laboratory of Biology Department, Taif University. Clusters were sorted to remove any infected and damaged berries. At the beginning of the experiment, samples of 12 clusters were taken to determine the initial berry and cluster properties, then 60 clusters were randomly divided into five groups and each group dipping for five minutes at 22°C in one of the salicylic acid (SA) at (2 or 4mM) and jasmonic acid (JA) at (5 or 10 mM) and distilled water was used as control. All solutions contained tween-20 (2 ml L-1) and were air–dried at room temperature for 2 h, and then each cluster packed using ventilated bags. All bags with clusters were weighted and each four bags were put in ventilated box (50x30x12) cm. After treatments, carton boxes were taken and stored under cold storage at 0^{o}C and 90-95 % relative humidity (R.H.) for 60 days followed by 3 days shelf life at 20^{o}C. The treated clusters were evaluated in each treatment at 30, 60 and 60+3 days of treatment.

Cluster weight loss and berry shatter percentages:
Cluster weight was recorded several times during the storage period and calculated as percentage of water loss in comparison with initial weight. Berry shatter was determined by weighting the berries per cluster which separate from cap stem after moderate shaking and then percent of berry shatter was estimated.

$$\text{Berry shatter \%} = \frac{\text{Weight of berry shatter}}{\text{Intial weight}} \text{x}100$$

Berry decay and total loss in cluster weight percentages:
Percent of berry decay was recorded several times during the storage period and calculated as percentage of decayed berries in comparison with initial weight. Also, total loss in cluster weight percentage was calculated by adding the percentage of loss in cluster weight, berry shatter and decayed berries.

Berry separation force and berry firmness:
Berry separation force was determined by measuring the separation force from samples of 10 berries for each cluster (replicate) for each treatment and the average was estimated (gm_f). Berry firmness was measured on 10 berries for each replicate were taken randomly for each treatment to determine berry firmness and the results were expressed as Newton's units. Berry separation force and firmness were determined by using PHSH-Pull (Dynamometer Model DT 101) with 3/16 inch plunger).

Soluble solids content (SSC%), Titratable acidity (TA%) and SSC/acid ratio :
Soluble solids content in berry juice were measured as Brix % by using digitalrefractometer (DR 6000, A. Kruss Optronic GmbH, Hamburg, Germany).Ten ml of berry juice was titrated against 0.1 N sodium hydroxide solution using phenolphthalein as indicator. Total acidity was expressed as gram tartaric acid/100 ml juice according to (A.O.A.C., 1980). SSC/acid ratio was calculated from the results recorded from juice SSC and titratable acidity.

Total anthocyanin content:

Total anthocyanin content was measured according to (Mazumdar and Majumder, 2003) by extracting half gram of fresh berry skin in 10 mL of ethanolichydrocholric acid 1.5 N. Samples put overnight at 40^{o}C then centrifuged for 3 min and filtered through filter paper (Whatman No.1). The filtered aliquot was maintained in darkness for about 2 h with cover of the container. The optical density (OD) value of the solution was measured through 535 nm wave length in a spectrophotometer against blank. The amount of total anthocyanin in berry skin was calculated using the following equations:

Total absorbance value for the berry skin (per 100g) $=\frac{e.b.c}{d.a}$

Where:

a= Weight of sample

b= Volume made for color measurement

c= Total volume made

d= Volume of aliquot taken for estimation

e= Specific OD value at 535 nm wavelength.

The 1 mg mL-1 of the solution is equivalent to the absorbance of 98.2. Therefore, the amount of total anthocyanin present in the sample (mg/100g) = Total absorbance for the sample /98.2.

Rachis browning and berry appearance:

Rachis browning development was evaluated using the following scoring system: (1) healthy, entire rachis including the cap stems (merging point between berries and rachis) green; (2) slight, only cap stems showing browning; (3) moderate, cap stems and secondary rachis showing browning and (4) severe, cap stems, secondary and primary rachis completely brown (Crisosto et al., 2001). All brown berries were removed and weighed, and berry browning was expressed as a percentage of cluster weight. Berry appearance was evaluated from visual inspection of berries and assignment of score, i.e., 1: excellent, 2: good, 3: slightly dull, 4:<50% brownish and soft berries and 5: >50% brownish and soft berries (Xu et al., 2007).

Statistical analysis of data:

The data were statistically analyzed as a factorial experiment in a complete randomized design with four replicates by analysis of variance (ANOVA) using the statistical package software SAS (SAS Institute Inc., 2000, Cary, NC., USA).

Results and Discussion

Weight loss, berry decay, berry shatter and total loss in cluster weight percentages:

Total loss in cluster weight were mainly due to weight loss, berry shatter and berry decay percentages. In our study weight loss, berry shatter (Table 1), berry decay and total loss in cluster weight percentages (Table 2) were significantly increased gradually as storage period advanced . Moreover, SA and JA treatments at all concentrations significantly decreased previous parameters during cold storage period and shelf life as compared to the control. Since, SA might have reduced respiration and transpiration which concomitantly delayed senescence. In addition, salicylic acid as an electron donor produces free radicals which prevents normal respiration (Wolucka et al., 2005). In this respect, weight loss is due to metabolic activity, respiration and transpiration. Also, SA can be decreased respiration rate and fruit weight loss by stomata closing (Zheng and Zhang, 2004). However, the current results are in agreement with those obtained by (Shafiee et al., 2010) who reported that dipping strawberry fruits in salicylic acid solution had less weight loss than control. Furthermore, SA treatments significantly decreased weight loss of grapes during cold storage (Khalil, 2014 and Ranjbaran et al., 2011). The effect of SA on berry shatter can be attributed to a suppression of both ethylene production and its action in the abscission layer (Srivastava and Dwivedi, 2000). Moreover, SA treatments significantly reduced fungal decay of grapes confirming the fact that SA leads to induce the expression of many defense genes against pathogens (Khalil, 2014). Exogenous application of SA might be improved resistance of treated fruit against fungal attack (Xu and Tian, 2008). The inhibitory effects of postharvest treatment with SA on fungal decay confirm the previous reports about its anti-fungal effects (Lu and Chen, 2005). Furthermore, Xu and Tian (2008) reported that the resistance of SA-treated fruits against fungal attack can be attributed to the consequence of increases in activities of anti-oxidant enzymes. Likewise, Jiankang et al. (2006) showed that higher defensive enzymes (chitinase, peroxidaseand phenylalanine ammonia-lyase (PAL) in young pear fruitswhich SA has been sprayed on them. These results are in agreement with those observed by (Ranjbaran et al., 2011) who presented that SA treatments reduced fungal decay of grapesduring cold storage. Similarly, Ding et al. (2001) showed that methyl jasmonate induced the synthesis and expression of some stress proteins such as heat shock proteins which lead to increase resistance of the pathogens and decreased incidence of

decay. It is observed in strawberry that the suppression of fungus decay may be induce the chemical defense mechanisms of plant tissues by low concentration jasmonate (Ayala-zavala et al., 2005). The present study demonstrated that the lowest significant percent of weight loss, berry shatter, berry decay and total loss were achieved by SA at 4 mM and JA at 10 mM at the end of shelf life in comparison with other treatments.

Table (1): Impact of salicylic acid (SA) and jasmonic acid (JA) on weight loss and berry shatter percentages of "Crimson Seedless"grapes during cold storage and shelf life

Treatments	weight Loss % Storage period (days)				Berry shatter% Storage period (days)			
	0	30	60	60+3*	0	30	60	60+3*
Control	0m	2.93i	4.8e	7.18a	0h	1.81ef	4.04d	8.09a
SA 2 mM	0m	2.56jk	4.42fg	6.24b	0h	0.36g	1.99e	5.15b
SA 4 mM	0m	2.41l	4.27h	5.96c	0h	0.38g	1.89e	3.99d
JA 5 mM	0m	2.64j	4.50f	6.12b	0h	0.61g	2.00e	5.18b
JA 10 mM	0m	2.46kl	4.34gh	5.59d	0h	0.40g	1.72f	4.51c

Means within and between columns followed by the same letter are not significantly different at level p = 0.05 means * 60 days after cold storage at 0°C plus 3 days at shelf life at 20°C.

Table (2): Impact of salicylic acid (SA) and jasmonic acid (JA) on berry decay and total loss percentage of "Crimson Seedless"grapes during cold storage and shelf life

Treatments	Berry decay % Storage period (days)				Total loss % Storage period (days)			
	0	30	60	60+3*	0	30	60	60+3*
Control	0i	1.25g	4.00e	8.47a	0k	5.99h	12.85e	23.73a
SA 2 mM	0i	0.56h	2.35f	5.64c	0k	3.48ij	8.77fg	17.00c
SA 4 mM	0i	0.53h	2.28f	5.19d	0k	3.31j	8.43g	15.13d
JA 5 mM	0i	0.66h	2.43f	6.19b	0k	3.91i	8.94f	17.49b
JA 10 mM	0i	0.60h	2.41f	5.50c	0k	3.46ij	8.47g	15.58d

Means within and between columns followed by the same letter are not significantly different at level p = 0.05 means * 60 days after cold storage at 0°C plus 3 days at shelf life at 20°C.

Berry separation force and berry firmness:

The present study showed that berry separation force and berry firmness were gradually reduced with storage period advanced till 60 days of cold storage and at the end of shelf life (Table 3). These results are in agreement with previously found that fruit firmness significantly decreased with increasing storage period (Samra et al., 2014; El-Metwally et al., 2014 and Ranjbaran et al., 2011). However, SA and JA treatments had insignificant effect on berry separation force as compared to the control during cold storage period and shelf life. Otherwise, the best significant value for berry firmness after 60 days of cold storage and shelf life was achieved by SA and JA treatments at all concentrations. In this respect, Zhang et al., (2003) reported that the influence of SA applications on improving kiwifruit firmness are due to inhibit ethylene production and its function. Moreover, SA inhibits cell wall and membrane degrading enzymes such as polygalacturonase (PG), lipoxygenase (LOX), cellulase and pectin methylesterase (PME) leading to decrease the fruit softening rate. In addition, Srivastava and Dwivedi (2000) explained that in climacteric fruits SA delays fruit softening by affecting major reduction of ethylene production. Furthermore**,** Lolaei (2013) showed that application of (Methyl Jasmonate) MJ had higher effects on delayed fruit repining and increased firmness of strawberry fruit.

Table (3): Impact of salicylic acid (SA) and jasmonic acid (JA) on berry separation force and berry firmness of "Crimson Seedless"grapes during cold storage and shelf life

Treatments	Berry separation force (gm_f) Storage period (days)				Berry firmness (N) Storage period (days)			
	0	30	60	60+3*	0	30	60	60+3*
Control	850a	771b	660d	618e	835a	759d	666g	604j
SA 2 mM	850a	775b	668d	625e	835a	766d	690f	623i
SA 4 mM	850a	737c	676d	631e	835a	793bc	720e	643h
JA 5 mM	850a	788b	683d	628e	835a	783c	694f	624i
JA 10 mM	850a	788b	675d	620e	835a	799b	703f	630hi

Means within and between columns followed by the same letter are not significantly different at level p = 0.05
means * 60 days after cold storage at 0°C plus 3 days at shelf life at 20°C.

Soluble solids content (SSC%), Titratable acidity (TA%) and SSC/acid ratio:

The present data showed that SSC was significantly increased during cold storage period and shelf life in all treated and untreated clusters. Otherwise, TA and SSC/acid ratio were not significantly affected by storage period (Table 4). In this respect, Ranjbaran et al. (2011) found that SSC increased with storage period advanced in all treated and untreated clusters but TA reduced in all clusters during cold storage and shelf life. Furthermore, the present work investigated that SSC, TA and SSC/Acid ratio were not significantly affected by SA and JA treatments during cold storage period and shelf life.Also, SA application had insignificant effect on mango SSC during cold storage (Ding et al., 2007) as has been observed in the present study. In this respect, Asghari and Aghdam (2010) observed that kiwifruits treated with MeSA of 32 ml L-1 maintained a lower SSC content than the control at the end of cold storage. Thus, MeSA reduced ethylene production may results to decrease SPS enzyme activity leading to decrease in sucrose synthesis. Moreover, Ghasemnezhad and Javaherdashti (2008) showed that raspberry fruit had the highest titratable acidity, but its content gradually decreased during storage. Declining both SSC and TA resulted in decreasing SSC to TA ratio, although no differences were found after 3 days of storage at 4°C. Moreover, the titratable acidity of berries treated with MJ was almost similar to untreated fruits after 7 days of storage.

Table (4): Impact of salicylic acid (SA) and jasmonic acid (JA) on SSC, acidity and SSC/acid ratio of "Crimson Seedless"grapes during cold storage.and shelf life

Treatments	SSC (Brix %) Storage period (days)				Acidity % Storage period (days)				SSC/acid ratio Storage period (days)			
	0	30	60	60+3*	0	30	60	60+3*	0	30	60	60+3*
Control	17.5i	17.7hi	18.3fg	19.0b-d	0.54e	0.59a-d	0.60a-c	0.63a	32.4ab	30.2ab	30.5ab	30.2ab
SA 2 mM	17.5i	18.1f-h	18.3e-g	19.4ab	0.54e	0.59a-d	0.61ab	0.61ab	32.4ab	30.7ab	30.2ab	31.6ab
SA 4 mM	17.5i	17.8g-i	18.6d-f	19.2a-c	0.54e	0.56c-e	0.58b-e	0.62a	32.4ab	32.3ab	32.6ab	30.8ab
JA 5 mM	17.5i	18.1e-h	18.7c-e	19.0b-d	0.54e	0.55de	0.60a-c	0.63a	32.4ab	33.0a	31.4ab	30.4ab
JA 10 mM	17.5i	18.2e-h	18.6d-f	19.4ab	0.54e	0.56c-e	0.59a-e	0.62ab	32.4ab	33.0a	32.0ab	31.3ab

Means within and between columns followed by the same letter are not significantly different at level p = 0.05
means * 60 days after cold storage at 0°C plus 3 days at shelf life at 20°C.

Total anthocyanin content:

The present data showed that total anthocyanin content in berry skin was continuously decreased significantly during cold storage period and shelf life (Table 5). These results are in agreement with (Samra et al., 2014 and El-Metwally et al., 2014). Otherwise, total anthocyanin content of grape berries displayed an increasing trend up to 45 days and then declined gradually (Harindra-Champa et al., 2014). The current work presented that total anthocyanin content was not significantly affected by all treatments during cold storage period and shelf life, but both of SA and JA treatments had somewhat increment in total anthocyanin at the end of shelf life in comparison with control. In this respect, Harindra-Champa et al.(2014) reported that SA at different concentrations had significant higher of total anthocyanin content in comparison with control. Moreover, Shafiee et al. (2010) found that addition of SA to nutrient solution was not effective on fruit color in comparison with control, postharvest treatments were not effective on lightness and the effect of SA may be due to decrease respiration which prevents fruit senescence during storage. It seems that these compounds prevent enzymatic activities which have a role in anthocyanin synthesis. In addition, Ghasemnezhad and Javaherdashti (2008) found that total anthocyanin content on raspberry

fruit was significantly affected by MJ treatment and storage period. Treated berries with24µl.l-1 maintained the highest level of anthocyanin as compared with other treatments at the end of thestorage period. Furthermore, Ayala-Zavala et al. (2005) revealed that strawberries treated with MJ showed the highest anthocyanin pigment at the end of storage period.

Table (5): Impact of salicylic acid (SA) and jasmonic acid (JA) on total anthocyanin of "Crimson Seedless"grapes during cold storage and shelf life

Treatments	Total Anthocyanin mg/100g (f.w.) Storage period (days)			
	0	30	60	60+3*
Control	85.0a	79.0bc	71.3de	63.8f
SA 2 mM	85.0a	80.0b	73.8d	65.3f
SA 4 mM	85.0a	80.5ab	75.0cd	66.8ef
JA 5 mM	85.0a	81.5ab	72.3d	65.0f
JA 10 mM	85.0a	79.8b	74.0d	66.3f

Means within and between columns followed by the same letter are not significantly different at level p = 0.05 means * 60 days after cold storage at 0°C plus 3 days at shelf life at 20°C.

Rachis browning and berry appearance:

Rachis browning was significantly increased during cold storage period and shelf life. Otherwise, berry appearance was significantly reduced during the previous period (Table 6). In this respect, the development of rachis browning during grape storage is associated with polyphenol oxidase activity (Carvajal-Millan et al., 2001). The present study showed that both of SA and JA treatments were significantly reduced rachis browning and visual appearance of berries as compared to the control during cold storage period and shelf life. These results are in agreement with (Ranjbaran et al., 2011) who reported that the effect of SA concentration on delaying rachis browning must be through inhibition of polyphenol oxidase activity. Moreover, SA treatments had significant positive effect on visual appearance of berries after 45 days of cold storage. In this investigation the lowest values of rachis browning and superior berry appearance were presented by SA at 4 mM and JA at 10 mM at the end of shelf life in comparison with other treatments. There is not any report on the effect of JA on rachis browning and berry appearance on grapes.

Table (6): Impact of salicylic acid (SA) and jasmonic acid (JA) on rachis browning and berry appearance of "Crimson Seedless" grapes during cold storage and shelf life

Treatments	Rachis Browning Storage period (days)				Berry Appearance Storage period (days)			
	0	30	60	60+3*	0	30	60	60+3*
Control	1.00k	2.88e	3.38c	3.88a	1.00j	2.38ef	3.38b	3.94a
SA 2 mM	1.00k	1.50j	2.44f	3.38c	1.00j	2.13gh	2.50e	3.00c
SA 4 mM	1.00k	1.44j	2.13h	2.94de	1.00j	1.88i	2.19fg	2.75d
JA 5 mM	1.00k	1.88i	2.38fg	3.63b	1.00j	1.94hi	2.50e	3.38b
JA 10 mM	1.00k	1.38j	2.19gh	3.13d	1.00j	2.00g-i	2.38ef	3.13c

Means within and between columns followed by the same letter are not significantly different at level p = 0.05 means * 60 days after cold storage at 0°C plus 3 days at shelf life at 20°C.

Conclusion:

It might be concluded that SA at 4 mM and JA at 10 mM applications had a positive effect in reducing berry decay, maintaining cluster and berry quality, improving rachis condition and enhancing berry appearance during cold storage period and shelf life. Therefore, postharvest SA at 4 mM and JA at 10 mM applications could be suggested for improving the storability of "Crimson Seedless" grapes and maintaining their quality characteristics during cold storage conditions and shelf life.

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