

POSTHARVEST QUALITY MAINTENANCE OF ‘Crimson Seedless’ GRAPES BY PREHARVEST APPLICATIONS

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ABSTRACT

The present study was carried out during both 2018 and 2019 growing seasons on ‘Crimson Seedless’ grape vine grown on sandy soil. Vines received water, salicylic acid 200 mg/L, chitosan 10 g/L, putrescine 5 mM, phenylalanine 400 mg/L, ascorbic acid 500 mg/L, citric acid 700 mg/L, gibberellic acid 20 mg/L, active dry yeast 15 g/L at véraison (change of color) stages and at 10 days before harvesting time. Fruit clusters were harvested when total soluble solids (TSS) reached 16%, stored at 0°C and 85–90 relative humidity for 35 days then kept 2 days at shelf life (25 ± 2°C and air humidity 70 ± 2). Results at harvest time showed that, salicylic acid, putrescine, gibberellic acid and yeast increased average berry weight and juice volume. Salicylic acid, chitosan, putrescine, citric acid and gibberellic acid maintained the berry adherence strength and firmness. Salicylic acid, chitosan, putrescine and phenylalanine increased berry red color, as well as berry anthocyanin, TSS and sugars content. Highest acidity content was obtained by salicylic acid, chitosan, citric acid and gibberellic acid spraying. Results after cold storage and shelf life showed that, berry weight loss decreased significantly by all chemical sprays, except phenylalanine. High maintenance of berry adherence strength and firmness occurred by gibberellic acid followed by putrescine. All chemicals reduced the unmarketable grapes except the phenylalanine.

Key words: agrochemicals, grape, quality, sprays, storagability

INTRODUCTION

The production, marketing and consumption of fresh horticultural crops are continuously increasing in the world. This increase is not only linked to the growth of the world population and to the consequent demand of fruits and vegetables but also to the continuous improving of the produce quality. Obtaining high yield is the main object of fruit growers, however, nowadays this should be correlated to a high appearance quality and nutritional value that should be maintained until reaching hands of the consumer.

Grape (*Vitis vinifera* L.) is considered one of the most important fruits produced in Egypt. According to FAO [2019] the area of grape vines in Egypt increased

recently reaching 73,351 ha which produces 1,626,259 Ton. However, it faces problems of quality losses either before or after harvesting which lowers its marketing and storage ability. Such losses would include poor coloring, clusters weight loss, stem drying and berries shattering, wilting and shriveling, as well as fungal infections [Dokoolian and Peacock 2001]. Accordingly, constant attention is required when dealing with grape growing, harvesting and storing in order to reduce such losses and to obtain maximum profit.

Foliar spraying with agrochemicals is one of the practices contribute to improve the grape berries quality at harvest time and after that by delaying ripening

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and reducing diseases infection. This practice is essential for grape producers when clusters are to be stored or shipped to distant markets. In the recent years, fruit consumer don not only seeks for the fruit appearing quality such as size and color, but also for its health and nutritive value. At the same time, most international markets are demanding fruit crops that are treated with least amounts of chemicals of any types. Therefore, several studies investigated the use of agrochemicals not only affecting fruit quality and its maintenance after harvest, but also the ones that are environmentally friendly and health safe. Salicylic acid (SA) chitosan, putrescine, phenylalanine, ascorbic acid, citric acid, gibberellic acid, active dry yeast and ascorbic acid are examples of safe chemicals used for such purpose. Yeasts are suitable as a bio-control agent of postharvest diseases [Freimoser et al. 2019]. Pre- or postharvest chitosan application has been considered as an alternative treatment to the use of synthetic fungicides to prevent berries postharvest decay and extend the storage life as well as retaining the overall berry quality [Reglinski et al. 2005, Liu et al. 2007]. Also, several studies indicated the important role of gibberellic acid, salicylic acid, putrescine and phenylalanine in enhancing the physico-chemical quality of the grape berries such as berry diameter and length, total soluble solids (TSS), acidity, TSS/acid ratio, sugars and anthocyanin content, as well as it keeping its firmness, adherence strength after harvest and during shelf life [Marzouk and Kassem 2011, El-Sayed 2013].

Additionally, Bhaskara Reddy et al. [2000] reported that the chitosan preharvest foliar sprays significantly reduced strawberry fruits deterioration and maintained the keeping quality during cold storage (fruit were firmer and ripened at a slower rate). Sun et al. [2010] suggested that ascorbic acid and chitosan showed positive roles in maintaining litchi fruit membrane integrity. As a result, total soluble solids, soluble sugars, and acidity in treated fruits were significantly increased, thus resulting in a lowered fruit decay rate. They also reported that ascorbic acid and chitosan showed positive effects on inhibiting pulp browning and decay and extending the shelf life by increasing anti-oxidation capacity while avoiding dehydration and microbial attack simultaneously. Also, citric acid preharvest foliar spray presented a higher skins berry

anthocyanin and soluble solids content (SSC) [Hegazi et al. 2014].

'Crimson Seedless' is one of the late ripening grape cultivars that recently entered Egypt and its cultivation was spread along new reclaimed areas such as El-Noubaria region. It is characterized with superior eating quality including firm texture, crisp and flavor. However, as any other grape variety, it faces quality losses at harvest time and during cold storage. Therefore, in accordance to the above discussed, the present study was conducted in order to study the effect of different preharvest foliar spraying with salicylic acid, chitosan, putrescine, phenylalanine, ascorbic acid, citric acid, gibberellic acid, active dry yeast and ascorbic acid on the retention of fruit quality attributes and nutritional value of fresh Crimson seedless grapes at harvest and after cold storage.

MATERIALS AND METHODS

Experimental site and plant materials. The present study was carried out during both 2018 and 2019 growing seasons on 'Crimson Seedless' grape vine. Vines were grown in a private orchard located at El Noubaria, El-Behira Governorate in a sandy soil under drip fertigation system with Nile water. The vines were planted at 3.5×4 m spacing and pruned by retaining a maximum of 60–67 nodes/vine. In November of both years vines were fertilized with organic manure and calcium super phosphate (15% P_2O_5) at a rate of 15 and 1.7 kg per vine, respectively. Also, 1.5 kg ammonium sulphate (20.5% N) and 1.0 kg potassium sulphate (48% K_2O) per vine were added in 3 equal doses at the beginning of March, April and May.

Preharvest treatments and experimental design. The experiment was designed as randomized complete block design (RCBD) and the following 9 foliar spray treatments were obtained with 3 replicates for each treatment (1 replicate = 2 vines): water, salicylic acid 200 mg/L, chitosan 10 g/L, putrescine (1,4-diaminobutane) 3 mM, phenylalanine 400 mg/L, ascorbic acid 500 mg/L, citric acid 700 mg/L, gibberellic acid 20 mg/L, active dry yeast 15 g/L. The chemical sprays were obtained at véraison (change of color) stages and then 10 days before harvesting time. The surfactant Nourfilm (produced by Alam Chemica) was added at the rate of 45 cm³/100 L water to all sprayed chemicals

in order to obtain best penetration results. The chemicals were applied directly to the clusters with a hand-held sprayer until runoff in the early morning.

Clusters stored conditions. Berry clusters were harvested at commercial harvest time (during the second half of September). Clusters of each replicate were packed in liners including boxes. Each box contained 8 kg (12–13 cluster) and every 3 boxes represented 3 replicates for each treatment. A fruit sample of 1.5 kg (approximately 2.5 clusters) of each replicate was taken for measuring the fruit physico-chemical quality characteristics at harvest. In the meantime, the rest of clusters were cold stored at 0°C and 90–95% relative humidity (RH) for 35 days, then kept for two days at shelf life (25 ±2°C and air humidity 70 ±2%) in order to investigate clusters storage ability.

Clusters quality at harvest. Average berry weight (g) and juice volume (mL) of 100 berries were measured. Berry adherence strength (g) was measured. Berry firmness (g/cm²) was recorded by a pressure tester texture analyzer instrument using a penetrating cylinder to a constant 1 mm inside the berry and by a constant speed and the peak of resistance was recorded per gram.

Cluster red color uniformity by counting the number of red berries to the total number of berries in each cluster. Red berries through the average number of red fruits to the number of total fruits within the clusters. In the juice, the percentage of TSS was measured by a hand refractometer (A.S.T., Japan), acidity as tartaric acid determined by titration with 0.1 N NaOH according to A.O.A.C [2019]. Total sugars were determined calorimetrically using phenol sulphuric acid method.

Total anthocyanin content (mg/100 g fresh weight): 0,5 g of fresh skin berries was ground with 10 mL of acidified alcohol solution, centrifuged for 3 min. and then filtered. The extract was measured at 535 nm using Spectra color meter according to Ranganna [1979].

Clusters quality after cold storage (clusters storage ability). In a sample of 6.5 kg clusters from each replicate, fruit storage ability was detected by measuring berry firmness, adherence strength, cluster weight loss. Also, the percentage of unmarketable clusters was calculated by the following equation after considering decayed, shriveled, shattered berries as defected.

$$\begin{aligned} \text{unmarketable clusters berries \%} &= \\ &= \frac{\text{weight of defected berries}}{\text{initial cluster weight}} \times 100 \end{aligned}$$

In addition, clusters fruit storage life was estimated as number of days from zero time of storage until reaching 6% clusters fruit weight loss and/or 20–25% unmarketable clusters fruit.

Statistical analysis

All data were tested for the effects of treatments on analyzed parameters by the one-way analysis of variance (ANOVA). Treatments means were separated and compared using the honest significant differences (HSD) at 0.05 level of significance.

RESULTS

Clusters quality at harvest

The effect of the different preharvest sprays on the clusters physical characteristics is presented in Table 1. Obtained results of both seasons showed that salicylic acid, putrescine, gibberellic acid and yeast increased the average berry weight and berry juice volume compared to the water sprayed clusters. Also, significant differences among treatments in the berry's adherence strength were recorded in both seasons. Salicylic acid, chitosan, putrescine, ascorbic acid, citric acid and gibberellic acid significantly increased the berries adherence strength, with putrescine and gibberellic acid resulting in the highest values. In addition, salicylic acid, chitosan, putrescine, gibberellic acid, yeast and citric acid sprays increased berry firmness during both seasons, with gibberellic acid and yeast resulting in the highest values. However, no significant differences in berry firmness occurred among clusters treated with phenylalanine, ascorbic acid or citric acid. Furthermore, results of both seasons also showed that salicylic acid, chitosan, putrescine, phenylalanine and yeast sprays resulted in increasing the percentage of cluster red color uniformity, with phenylalanine indicating the highest value followed by yeast.

Moreover, the effect of the different agrochemical's sprays on the chemical characteristics of 'Crimson' berries are presented in Table 2. Results showed that clustered treated with salicylic acid, chitosan, putrescine and phenylalanine during both seasons, and

Table 1. Effect of agrochemicals foliar spray on berry weight, juice volume, adherence strength and firmness of 'Crimson Seedless' grapes at harvest time of 2018 and 2019 seasons

Treatments	Berry Wright (g)		Juice volume (mL)		Adherence strength (g)		Firmness (g/cm ²)		Cluster red color uniformity (%)	
	2018	2019	2018	2019	2018	2019	2018	2019	2018	2019
Water	3.0	3.2	11.2	12.3	300.4	278.7	204.8	234.1	55	51
Salicylic acid	3.4	4.1	13.2	13.4	319.1	309.6	229.5	263.3	68	62
Chitosan	2.8	3.4	10.9	11.8	338.8	313.9	235.9	276.6	71	60
Putrescine	3.7	4.0	13.3	13.6	387.7	344.4	277.8	271.6	72	68
Phenylalanine	3.3	3.4	12.3	12.5	311.5	285.0	211.2	224.9	82	82
Ascorbic acid	3.2	3.6	12.7	12.8	325.5	315.1	207.2	248.3	63	58
Citric acid	3.3	3.6	11.9	12.5	325.0	323.9	209.9	204.9	60	58
Gibberellic acid	3.8	4.3	13.8	13.6	401.1	362.1	255.0	293.8	47	47
Yeast	3.6	3.9	13.2	13.4	314.5	306.6	231.0	264.9	78	66
HSD _{0.05}	0.4	0.7	1.8	1.1	17.7	25.8	19.7	23.9	10	9

HSD – honest significant differences

Table 2. Effect of agrochemicals foliar spray on berry TSS, acidity, total sugars and anthocyanin content of 'Crimson Seedless' grapes at harvest time of 2018 and 2019 seasons

Treatments	Total soluble solids (%)		Acidity (%)		Total segars (%)		Anthocyanin (mg/100 g)	
	2018	2019	2018	2019	2018	2019	2018	2019
Water	16.6	16.8	0.44	0.50	13.3	15.6	3.1	3.0
Salicylic acid	20.9	19.4	0.80	0.63	15.6	17.7	6.3	6.2
Chitosan	21.1	20.0	0.77	0.77	15.9	19.2	7.6	6.4
Putrescine	21.6	19.4	0.64	0.54	15.1	16.4	6.2	6.3
Phenylalanine	20.2	21.3	0.50	0.42	17.7	18.4	6.3	6.8
Ascorbic acid	19.5	19.6	0.60	0.61	14.7	16.6	4.2	6.2
Citric acid	18.6	18.1	0.78	0.66	14.3	16.3	5.0	6.1
Gibberellic acid	17.4	17.3	0.81	0.78	14.6	16.4	4.3	3.8
Yeast	18.1	18.5	0.68	0.23	14.6	16.2	4.3	3.5
HSD _{0.05}	3.3	2.1	0.32	0.11	1.8	0.89	2.1	2.6

HSD – honest significant differences

ascorbic acid in the second season only had significantly higher TSS content than the water treated ones. Obtained data also revealed that salicylic acid, chitosan, citric acid and gibberellic acid in both seasons, and ascorbic acid in the second season only resulted in the highest berry acidity content. As for the berry anthocyanin content, obtained data showed that berry anthocyanin content was significantly increased under the salicylic acid, chitosan, putrescine and phenylalanine treatments during both seasons and under ascorbic acid and citric acid sprays in the second season only.

Clusters storage ability

The effect of the different foliar spraying on cluster weight loss, berries adherence strength, berries firmness and unmarketable grapes after cold storage and keeping at shelf life is presented in Table 3. Obtained results showed significant decrease in the percentage of cluster weight loss during cold storage by all sprayed agrochemicals, except phenylalanine, in comparison with the control. In the meantime, chitosan and gibberellic acid sprays did not significantly differ in their influence on cluster weight loss, as well as, no

significant effect differences occurred among salicylic acid, chitosan, putrescine, ascorbic acid, citric acid, yeast and gibberellic acid. However, gibberellic acid sprays recorded the least weight loss values followed by chitosan during both seasons.

In addition, obtained data of both seasons revealed that all sprayed agrochemical, except phenylalanine resulted in significantly higher adherence strength than the water spraying. The highest value was obtained by the gibberellic acid treatment followed by putrescine. Regarding berry firmness, results of both seasons showed that, salicylic acid, chitosan, putrescine and gibberellic acid maintained berry firmness during cold storage, with no significant differences occurring among them. In addition, the highest firmness values were measured in clusters treated with gibberellic acid and chitosan. Regarding the percentage of unmarketable clusters, obtained data revealed that in comparison with the water sprayed clusters, all spraying treatment except phenylalanine reduced the percentage of unmarketable clusters after cold storage and keeping for two days at shelf life. Gibberellic acid, ascorbic acid and citric acid indicated the lowest per-

Table 3. Effect of agrochemicals foliar spray on cluster weight loss, adherence strength, berry firmness and unmarketable clusters of 'Crimson Seedless' grapes after 35 days of storage at 0°C and two days keeping at shelf life in 2018 and 2019 seasons

Treatments	Cluster weight loss (%)		Adherence strength (g)		Berry firmness (g/cm ²)		Unmarketable clusters (%)	
	2018	2019	2018	2019	2018	2019	2018	2019
Water	8.4	9.4	224.1	223.6	172.5	191.2	27.5	32.6
Salicylic acid	5.3	5.6	242.0	259.8	207.0	213.0	12.5	11.2
Chitosan	4.8	4.2	243.1	257.5	214.2	225.0	13.4	11.6
Putrescine	5.8	5.7	278.9	264.1	197.2	210.8	15.3	9.6
Phenylalanine	6.4	5.5	238.1	234.7	167.2	198.3	19.3	20.3
Ascorbic acid	5.7	7.7	243.7	262.4	186.8	200.2	13.1	12.1
Citric acid	5.8	6.5	255.0	261.0	188.5	173.9	8.2	9.6
Gibberellic acid	3.1	4.5	298.5	321.6	221.3	226.6	7.0	6.7
Yeast	5.4	5.1	256.8	295.9	187.1	185.5	12.8	6.4
HSD _{0.05}	2.1	1.9	16.8	18.7	15.2	9.8	9.7	15.4

HSD – honest significant differences

cent of unmarketable clusters, followed by salicylic acid, chitosan and yeast.

DISCUSSION

The above-mentioned results indicated a positive enhancement in all studied quality properties of the grape clusters by all sprayed agrochemicals especially salicylic acid, chitosan, putrescine, gibberellic acid and phenylalanine. Similar improvement in the physico-chemical characteristics of grape berries at harvest and during storage by different agrochemical applications was recorded in previous investigations. High values of fruit TSS, sugars and vitamin C as a result of chitosan application is recorded by Sun et al. [2010]. Marzouk and Kassem [2011] found that berry firmness, shelf life and adherence strength was better by putrescine, salicylic acid and gibberellic acid treatments and the percentage of unmarketable berries decreased after cold storage or after keeping at ambient temperature. Also, Kassem et al. [2011] reported increases in berry weight, firmness and acidity content at harvest by putrescine, salicylic acid and gibberellic acid applications. El-Sayed [2013] found that the phenylalanine preharvest treatment increased berry TSS and anthocyanin. He added the putrescine preharvest applications decreased berry unmarketable and weight loss percent and increased firmness. Never the less, chitosan is reported to maintain grape berries TSS, anthocyanin content, acidity and sugars content, as well as, delaying berries firmness change during storage [Plácido et al. 2016]. Similarly, working on jujube fruits, Qiuping and Wenshui [2007] recorded better retention of chlorophyll, total soluble solids and ascorbic acid content in the fruits as a result of chitosan treatments. Also, Plainsirichai et al. [2014] working on Rose apples found that chitosan resulted in high fruit firmness values. Moreover, obtained results of the present study showed an increase in berries acidity content at harvest by chitosan, citric, gibberellic and ascorbic acids [Kassem et al. 2011 and Al-Obeed 2011].

Weight loss of berries is mostly a result of losing water vapor loss during respiration causing berries dehydration. Symptoms of dehydration are not shown until the damage is quite evident in clusters' stems and wrinkles starts to appear in the berry skin above 5% weight loss [Nelson 1978]. However, in some

table grapes cultivars, water losses of 2 to 3% based on the initial weight, are sufficient to cause browning symptoms in stems [Crisosto et al. 1994]. It is well confirmed that decreasing weight loss delays clusters deterioration [Lo'ay and El-Boray 2018]. Results of the present study showed significant reduction in weight loss by all sprayed agrochemicals. Similarly, Sharma and Shafaat [1995] found that fruits of Thompson seedless grape treated with citric acid at 100 ppm and kept in sealed polyethylene bags for 11 days had the lowest percentage of weight loss and shriveling. Also, Chien et al. [2007] confirmed the ability of chitosan to maintain fruit weight as a result of its applications. In the meantime, Al-Obeed [2011] recorded enhancement in berry firmness, shelf life, adherence strength and storability, and reduction in berry shattering and weight loss by putrescine, gibberellic acid and salicylic acid treatments. Similar findings were obtained by Kassem et al. [2011]. Never the less, Khalil [2020] reported the significant influences of gibberellic acid in reducing weight loss, shattering and unmarketable percent of Flame Seedless grapes. In addition, pre- and post-harvest applications of salicylic acid are reported to effectively reduce fruit softening rate, delay ripening process and thus maximizing fruit quality maintenance and storage life [Zhang et al. 2003, Lu et al. 2011, Ranjbaran et al. 2011]. Salicylic acid is known to inhibit ethylene biosynthesis and action by blocking the conversion of aminocyclopropane-1-carboxylic acid (ACC) to ethylene by decreasing ACC oxidase production and activity [Leslie and Romani 1988]. It is also reported to retard the ripening process through the induction of antioxidant enzymes such as peroxidase, catalase and superoxide dismutase and reduction of lipoxygenase activity [Zhang et al. 2003].

Furthermore, pathogens infection is known to play an important role in decreasing marketability of any fresh consumed fruits. Obtained data of the present work showed positive influence of gibberellic, salicylic, ascorbic and citric acids, as well as, chitosan and yeast applications in decreasing the percentage of unmarketable clusters after harvest and during cold storage. Accordingly, this influence would not only refer to their effect on decreasing fruit weight loss and maintaining its firmness, but also on their ability to protect the berries from pathogens attack. Chitosan

elicitation of fruits biochemical defense responses, as well as its antimicrobial properties by impeding the movement of microbial cells is evident [Liu et al. 2007]. Moreover, salicylic acid role in controlling fungal decay is referred either to its activation of antioxidant defense responses [Xu and Tian 2008] or its direct antifungal effects on fungus development [Amorabe et al. 2002]. They isolated a new ascosporic yeast (*Metschnikowia pulcherrima*) from grapes and they considered it as a biocontrol agent against post-harvest diseases as it rapidly colonizes and survive on fruit surface for long periods under different conditions, and thus limits nutrients availability to the pathogen and generally inhibits their growth. They stated this yeast activity in reducing incidence of rot by 100% apricot fruits stored at 4°C. Similarly, Sabry et al. [2013] found that yeast preharvest treatment enhanced storability, since it reduced weight loss, decay, shattering and total spoilage percentages, as well as, firmness loss of grape berries during cold storage.

In addition, SA treatments improved berry color due to increased concentration of total and individual anthocyanins in 'Crimson Seedless' grapes [García-Pastor et al. 2020]. Preharvest spraying with putrescine, ascorbic acid, salicylic acid and citric acid led to markedly increase quality and extended the storage periods at (2°C with 85–95% RH) of 'Anna' apple fruits [Aly et al. 2019].

Moreover, SA treatments of sweet cherry trees applied at key points of fruit development increased fruit quality attributes at harvest, such as weight, firmness, and content of bioactive compounds namely phenolics, including anthocyanins, which were maintained during storage [Giménez et al. 2014]. In addition, SA is natural compounds in plants which are recognized as GRAS (generally recognized as safe) for the United States Food and Drug Administration (FDA) and previous reports showed that fruit treatments with salicylates do not impart taste or off-flavor to fruit, although some sensory attributes, such as sweetness and firmness, increased [Giménez et al. 2014].

CONCLUSION

According to the obtained results it might be concluded the efficient and safe use of the examined agrochemicals in enhancing quality and extending storage

life of the 'Crimson Seedless' grape clusters, especially gibberellic acid.

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