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EFFECT OF GIBBERELLIC ACID AND ETHEPHON APPLICATIONS ON BERRY COLOR AND QUALITY OF "FLAME SEEDLESS" GRAPES

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ABSTRACT

This investigation aimed to evaluate the effect of spraying "Flame Seedless" grapes with gibberellic acid (GA_3) and ethephon at 250 ppm when berry size was about 10, 12 and 14 mm on berry color and quality. The present results revealed that spraying "Flame Seedless" grapes with GA_3 alone significantly increased cluster weight, berry weight and yield/vine, whereas decreased anthocyanin content and SSC/A ratio in comparison with the control. Moreover, adding ethephon at 250 ppm to the vine which sprayed with GA_3 when berry size was about 14 mm significantly increased cluster weight, berry weight and yield/ vine than those treated with ethephon when berry size was about 10 or 12 mm or with GA_3 alone. Also, this treatment accelerated harvest date through their its effect on increasing SSC/A ratio in berry juice. Furthermore, vines sprayed with ethephon at 250 ppm when berry size was about 14 mm produced superior color and increased anthocyanin content in berry skin than those sprayed with GA_3 alone or the control. It could be concluded that spraying "Flame Seedless" grapes with ethephon at 250 ppm when berry size reached about 14 mm was the suitable date for ethephon application as compared to those sprayed when berry size was about 10 or 12 mm or with GA_3 alone.

INTRODUCTION

Grapevine (*Vitis vinifera* L) is considered one of the most popular fruit crop in the world. In Egypt, it ranks the second fruit crop after citrus since, the total cultivated area reached about 160632 fed. with an annual total production of about 1389133 metric tons according to FAO (2013). "Flame Seedless" grapes is one of the most important grapes varieties due to their high production which give a high net income to the grape growers.

Plant growth regulators are usually applied in order to improve berry size and quality of grapevine (Rusjan, 2010). Gibberellic acid (GA_3) is used widely for improving the yield and fruit quality. GA_3 is applied to grape cultivars during the fruit set to increase berry size (Peacock and Beede, 2004) Application of gibberellic acid (GA_3) is necessary to increase yield, cluster and berry size and to improve berry color. Generally, growers using ethephon as ethylene releasing compound (Szyjewicz *et al*., 1984 and Celia *et al*., 2007).

Anthocyanin accumulation begins at veraison stage in grapes since, it is the onset of maturation (Celia *et al*., 2007). Moreover, most table grapes grown in warm climate regions may develop less red color than those from colder regions, because high temperature inhibits anthocyanin accumulation in grape skin (Spayed *et al*., 2002). Poor coloration substantially reduces the economic value of grapes. However, cultural practices such as leaf removal, shoot and cluster thinning can enhance the quality of "Flame Seedless" grapes (Dokoozlian and Hirschfelt, 1995). Ethephon applications enhance color development in pigment cultivars, increase soluble solids, reduce acidity and berry firmness (Szyjewicz *et al*., 1984).

Moreover, most growers used ethephon on "Flame Seedless" grapes to accelerate berry color and enhance time of harvest (Jensen *et al*., 1982) Also, ABA is associated with the physiology of fruit maturation in grape including anthocyanin accumulation in berry skin. Since, ABA treatment can increase skin anthocyanin concentration but the high cost of ABA produced the development of practical applications for viticulture and compound was never tested in Flame Seedless (Kataoka *et al*., 1982 and Peppi *et al.,* 2006).

In this respect, many investigators reported that spraying "Flame Seedless" grapes with ethephon at veraison stage in order to enhance berry color and maturation. Therefore, this study was undertaken to evaluate the effect of spraying ethephon application at 250 ppm when berry size reached about 10, 12 and 14 mm with GA_3 on berry coloring and quality of "Flame Seedless" grapes

MATERIALS AND METHODS

Plant materials and experimental procedure:

This experiment was carried out during the two seasons of 2014 and 2015 on "Flame Seedless" grapes grown in a sandy soil, under drip irrigation system, cultivated at 2 m within plants and 3 m between rows, spur pruned under baron trellis system and received normal cultural practices at a commercial vineyard, located at El-khatatba city, Monofia Governorate. Vines were pruned on the 15^{th} of December as spur pruning leaving about 80 eyes per vine. Dormex was used at 0.5 % at the first week of January in order to enhance bud burst and increase its percentage. Vines were sprayed with GA_3 at 5 ppm when cluster length reached about 7-8 cm and also with the same concentration at full bloom stage. After fruit set vines also were sprayed with GA_3 at 30 ppm when the average berries size was about 6-8 mm and after four days with GA_3 at 40 ppm .

Ethephon applications were carried out three times at 250 ppm when berry size (diameter) was about 10, 12, 14 mm, respectively in order to present the suitable date of spraying on berry color and its quality. Forty vines were selected, eight vines for each treatment (with four replicates) and presented on the following treatments:

No.	Treatments
1	Control (vines sprayed with tap water)
2	Vines sprayed with GA_3
3	Vines sprayed with GA_3 and Ethephon at 250 ppm when berries size were about 10 mm
4	Vines sprayed with GA_3 and Ethephon at 250 ppm when berries size were about 12 mm
5	Vines sprayed with GA_3 and Ethephon at 250 ppm when berries size were about 14 mm

When soluble solids content of berry juice was about 18% from the untreated vine, clusters from each treatment were counted and classified to different groups according to the color of each cluster in order to determine cluster color score using the following scoring system: (1) Cluster color ranged from 0-25%; (2) Cluster color ranged from 25-50%.; (3) Cluster color ranged from 50-75% and (4) Cluster color ranged from 75-100 %. The percentage of cluster color score was estimated using the following equations:

$$\text{Cluster color score} = \frac{\text{Av. No. of each grade color X No. of clusters}}{\text{Total number of clusters / vine}}$$

At harvest, when SSC of berry juice was about 18% for the untreated vine, clusters were counted and weighted to estimate yield/ vine (kg). Samples of 12 clusters from each treatment were taken and transported to the laboratory of Pomology Dept., Agric. Fac., Mansoura Univ. to determine:

1- Average cluster weight (g).
2- Average berry weight (g) and berry size (berry diameter, mm).
3- Soluble solids content (SSC%) It was measured by using a hand refractometer according to (Chen and Mellenthin, 1981).
4- Titratable acidity (TA%) 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).
5- SSC/acid ratio was calculated from the results recorded from juice SSC and titratable acidity.

6- Total anthocyanin content:

It was measured according to (Mazumdar and Majumder, 2003) by extracting half gram of fresh berry skin in 10 mL of ethanolic hydrocholric acid 1.5 N. Samples put overnight at room temperature 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 covering of the container. The optical density (OD) value of the solution was measured through 535 nm by a spectrophotometer against blank. The amount of total anthocyanin in berry skin (pericarp) was calculated using the following equation:

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.
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.

Statistical analysis of data:

The obtained data were statistically analyzed, and the differences between treatments were tested by analysis of variance (ANOVA) according to Snedecor and Cochran (1980). Means of treatments were compared using L.S.D value at 5 % level of probability.

RESULTS AND DISCUSSION

1-Effect of gibberellic acid and ethephon applications on yield/ vine :

Data from Table (1) showed that vines sprayed with GA_3 alone or with ethephon at different dates significantly increased yield/ vine as compared to the control. Moreover, vines sprayed with GA_3 and ethephon at 250 ppm when berry size reached about 14 mm gave higher significant values as compared to other treatments, since this treatment increased yield/vine by about 41.2% higher than the control as mean of two seasons .Whereas, yield/ vine showed no clear response when vines spraying with GA_3 alone or with ethephon at 250 ppm when berry size reached 10 and 12 mm. Yet, vine sprayed with ethephon at 10 mm berry size with GA_3 had a somewhat reduction in yield/ vine as compared to other treatments.

Similarly, Peacock and Beede (2004) reported that raisin yield and fresh weight of "Thompson Seedless" grapes were the greatest when GA_3 was applied 14 days after full bloom. The role of GA_3 in improving the fruit weight may be due to its effect in increasing cell enlargement (Pharis and King, 1985).

Table (1): Effect of gibberellic acid and ethephon applications on yield and clusters weight of "Flame Seedless" grapes:

Treatments	Yield/ vine (kg)		Mean	Cluster weight(g)		Mean
	2014	2015		2014	2015	
Control	13.3 c	12.9 c	13.1	630.5 c	561.3 c	595.9
GA_3	16.3 b	17.6 b	17.0	772.3 ab	739.0 c	755.7
GA_3 and ethephon at 10 mm	15.8 b	17.3 b	16.6	749.8 b	750.5 bc	750.2
GA_3 and ethephon at 12 mm	15.5 b	18.2 b	16.9	734.3 b	787.5 b	760.9
GA_3 and ethephon at 14 mm	17.5 a	19.4 a	18.5	828.8 a	843.8 a	836.3
L.S.D at 5%	1.18	1.12	------	56.93	45.76	------

2-Effect of gibberellic acid and ethephon applications on cluster weight :

It is clear from Table (1) that vines sprayed with GA_3 alone or with ethephon at different berry size significantly increased average cluster weight as compared to the control. Yet, vines sprayed with GA_3 and ethephon at 250 ppm when berry size was about 14 mm (veraison stage) led to a significant increment in cluster weight in comparison with the other treatments and the control. So, the increment in cluster weight due to this treatment was about 40.3% over the control as mean of two seasons. Furthermore, vines sprayed with GA_3 only or with ethephon at 250 ppm when berry size was about 10 or 12 mm had no clear response for the average of cluster weight.

In this respect, Zahedi, *et al*. (2013) recorded that weight of clusters is one of the quality factors that influenced the sale price. Clusters treated with GA_3 and ethephon had the highest weight as compared to all other treatments in both cultivars. The lowest fruit weight was belonged to untreated vines. Also, Dimovska *et al*. (2014) mentioned that GA_3 applications were effective in increasing the cluster weight as well as the number of berries of Flame Seedless grapes. The increase of berry size is a result of the enhanced cell division and cell expansion. Furthermore, Dimovska *et al* .(2014) showed that treated “Flame Seedless” grapes with GA_3 10 or 20 mg/l were effective in increasing cluster weight as compared to untreated treatments.

3-Effect of gibberellic acid and ethephon applications on berry weight and size:

Results presented in Table (2) revealed that sprayed Flame Seedless grapes with GA_3 alone or with ethephon at 250 ppm when berry size reached about 10, 12 and 14 mm produced significantly higher berry weight and size as compared to the control. Moreover, vines sprayed with GA_3 alone or followed by ethephon at 250 ppm when berry size was about 14 mm produced a higher berry weight and size in comparison with the other treatments or the control. These treatments increased berry weight and size by about 32.0, 20.4 %, respectively as mean of two seasons. So, clusters of “Flame Seedless” grapes sprayed with ethephon at 250 ppm when berry size was about 14 mm had a superior effect in this respect. Szyjewicz *et al* (1984) mentioned that the effect of ethephon on fruit composition varied with cultivars, timing, concentration and method of application. The obtained results are in agreement with the findings of Lombard *et al.* (2004) who reported that average berry size was influenced by dosage of ethephon and tended to reach a maximum at 200 ppm. Dimovska *et al*. (2014) recorded that applications of GA_3 at 10 or 20 mg/l on Flame Seedless grapes were effective in increasing berry weight and size since, the increase in berry size is a result of the enhanced cell division and expansion . Reynolds *et al.* (1992) presented that ethephon application increased berry size

Table (2): Effect of gibberellic acid and ethephon applications on berry weight and size of “Flame Seedless” grapes:

Treatments	Berry weight (g)		Mean	Berry size (mm)		Mean
	2014	2015		2014	2015	
Control	2.4 c	2.5 c	2.5	14.0 d	16.3 c	15.2
GA_3	3.1 ab	3.0 b	3.1	17.8 ab	17.5 b	17.7
GA_3 and ethephon at 10 mm	2.9 b	3.0 b	3.0	16.5 c	17.4 b	17.0
GA_3 and ethephon at 12 mm	3.0 b	3.1 ab	3.1	17.0 bc	17.6 b	17.3
GA_3 and ethephon at 14 mm	3.3 a	3.2 a	3.3	18.3 a	18.2 a	18.3
L.S.D at 5%	0.200	0.201	------	0.96	0.52	-------

4-Effect of gibberellic acid and ethephon applications on SSC, total acidity and SSC/ acid ratio:

It is cleared from Table (3) that vine sprayed with GA_3 and ethephon at 250 ppm when berry size was about 14 mm gave significantly higher values of soluble solid content in berry juice in comparison with the other treatments and the control. Moreover, the untreated vines produced significantly higher SSC than those sprayed with GA_3 alone or with ethephon when berry size was about 10 or 12 mm. Whereas, vines sprayed with GA_3 alone gave significantly lower values of SSC as compared to the other treatments and the control.

Concerning the effect of GA_3 alone or with ethephon at different times on total acidity, data from the same table revealed that vines sprayed with GA_3 and ethephon when berry size was about 12 or 14 mm gave a lower significant values of total acidity compared with the other used treatments. Furthermore, vines sprayed with GA_3 alone gave a higher significant values of total acidity as compared to other treatments. Also, the obtained results showed that vines sprayed with GA_3 and ethephon at 250 ppm when berry size reached about 12 or 14 mm gave a significantly higher values of SSC/ acid ratio of berry juice than those sprayed with ethephon when berry size was about 10 mm. Whereas, vines sprayed with GA_3 alone produced a significant lower values of SSC/ acid ratio of berry juice than the other treatments. The current results showed that vines sprayed with GA_3 and ethephon at 250 ppm when berry size was about 14 mm gave a significant higher values of SSC/ Acid ratio as compared to other treatments.

Table (3): Effect of gibberellic acid and ethephon applications on SSC, Acidity and SSC/ A ratio of "Flame Seedless" grapes:

Treatments	SSC %		Acidity %		SSC/A ratio	
	2014	**2015**	**2014**	**2015**	**2014**	**2015**
Control	18.5 b	18.9 b	0.725 ab	0.816 ab	25.5 c	23.2 c
GA_3	16.8 d	17.0 d	0.755 a	0.849 a	22.3 d	20.0 d
GA_3 and ethephon at 10 mm	17.1 cd	17.3 cd	0.688 b	0.774 b	24.9 c	22.4 c
GA_3 and ethephon at 12 mm	17.5 c	17.7 c	0.588 c	0.679 c	29.8 b	26.1 b
GA_3 and ethephon at 14 mm	19.5 a	19.7 a	0.605 c	0.681 c	32.2 a	28.9 a
L.S.D at 5%	0.60	0.56	0.039	0.061	1.89	2.30

The increment in these values may be due to the effect on increasing the values of SSC with reducing the content of total acidity in berry juice. However, vines sprayed with GA_3 alone gave a significant lower values of SSC/ acid ratio than the other treatments or the control. In this respect, Szyjewicz *et al.* (1984) mentioned that the effect of ethephon on fruit composition varied with cultivars, timing, concentration and method of application, so contradictory results have been noted about its effects on SSC, TA. Also, Lombard *et al.* (2004) reported that SSC and TA levels of "Flame Seedless" grape decreased by increasing ethephon dosages above100 mg·L-1. The higher dosages had significantly lower SSC and TA levels than the control. Moreover, Elsabagh (2010) revealed that ethephon increased SSC and SSC/acid ratio and decreased acidity especially at 500 ppm. The obtained results are in agreement with Hashim *et al*. (2010) who presented that spraying Earlimart and Sweet Scarlet grapes with GA3 at 10, 20, 30 and 40 mg/ l at fruit set stage significantly reduced soluble solids content and titratable acidity. Vines treated with GA3 at 10-20 mg/l showed a somewhat reduction in total acidity (Rusjan, 2010).

5-Effect of gibberellic acid and ethephon applications on cluster color score and total anthocyanin content:

Data presented in Table (4) showed that vines sprayed with GA_3 and ethephon at 250 ppm when berry size was about 14 mm produced significantly higher values of cluster coloration as compared to the other treatments. Since, this treatment produced a grade of 3.9 of clusters color and this percent showed that most of clusters on the vine reached full colored (grade No.4) . Also, vines sprayed with GA_3 and ethephon when berry size was about 12 mm and the control gave a higher cluster color score than those treated with GA_3 alone or with ethephon when berry size was about 10 mm, since the value of this treatment was showed grade 3.6 . However, vines sprayed with GA_3 alone produced a lower value of cluster color since the grade color was 3.0. This grade showed that clusters color was about 75%.

Table (4): Effect of gibberellic acid and ethephon applications on total anthocyanin and cluster color scores of "Flame Seedless" grapes:

Treatments	Cluster color score		Mean	Total anthocyanin mg/ 100 gm (F. W)		Mean
	2014	2014		2014	2015	
Control	3.4 b	3.7 b	3.6	55.7 d	61.7 d	58.7
GA_3	3.3 b	2.7 b	3.0	56.3 d	47.7 e	52.0
GA_3 and ethephon at 10 mm	3.5 ab	3.3 c	3.4	71.3 c	67.2 c	69.3
GA_3 and ethephon at 12 mm	3.4 b	3.7 b	3.6	77.5 b	82.2 b	79.9
GA_3 and ethephon at 14 mm	3.8 a	4.0 a	3.9	81.8 a	88.0 a	84.9
L.S.D at 5%	0.304	0.118	--------	3.44	4.22	-------

Concerning the effect of anthocyanin content, data showed that the content of anthocyanin reflected those obtained from average color score. Vines sprayed with GA_3 and ethephon at 250 ppm when berry size was about 14 mm significantly produced higher values of anthocyanin content in berry skin in comparison with the other treatments and the control. Furthermore, vines treated with ethephon when berries size was about 12 mm gave significantly higher values of anthocyanin content in berry skin than those sprayed when berry size was about 10 mm or those treated with GA_3 alone. Vine sprayed with GA_3 alone gave a lower value of anthocyanin content in berry skin than the other used treatments. Moreover, the present results showed that sprayed Flame Seedless grapes with GA3 and ethephon at 250 ppm when berry size was about 12 or 14 mm produced a higher anthocyanin content and cluster coloration as compared to other treatments.

Also, the present data showed that sprayed vines with GA_3 and ethephon when berry size was about 14 mm gave superior color as compared to those sprayed when berry size was about 10 or 12 mm. In this respect, Dokoozlian and Hirschfelt (1995) observed that poor coloration substantially reduced the economic value of table grapes. Cultural practices such as leaf removal, shoot and cluster thinning can enhance the quality of

"Flame Seedless" grapes. Ethephon can stimulate anthocyanin production and color improvement in the skin of grape berries (Lombard *et al*., 2004)

Peppi *et al*. (2006) observed that anthocyanin accumulation begins at veraison, the onset of maturation. ABA increased the anthocyanin content of grape skin. Celia *et al*. (2007) found that ABA was more effective than ethephon in improving color of Crimson Seedless grapes. Yet, Rusjan (2010) found that GA_3 over 50 ppm led to a reduction in the value of anthocyanin content in berries. Mohammad and Parseh (2011) found that spraying ethephon at 100, 200 and 300 ppm at 20-30 % at veraison stage enhanced berry maturation of Beidaneh Ghermez grape. Ethephon application significantly increased fruit maturation, color and berry size .

CONCLUSION

It might be concluded that spraying "Flame Seedless" grapes with GA_3 and ethephon at 250 ppm when berry size was about 14 mm significantly increased yield/ vine, cluster weight, berry size , SSC/A ratio, improved berry color and anthocyanin content as compared to those treated with ethephon when berry size was about 10, 12 mm or the control. Therefore, spraying Flame Seedless grapes with GA_3 and ethephon at 250 ppm when berry size was about 14 mm could be suggested for improving berry color and quality of "Flame Seedless" grapes

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تأثير الرش بحمض الجبريليك و الإثيفون على التلوين و صفات الجودة في صنف العنب الفليم سيدلس

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[2] قسم النبات – كلية الزراعة – جامعة المنصورة

أجريت هذه الدراسة خلال موسمى ٢٠١٤-٢٠١٥ لدراسة تأثير رش العنب صنف الفليم سيدلس بحمض الجبريليك و الإثيفون بتركيز ٢٥٠ جزء فى المليون عند وصول حجم حبات العنب ١٠، ١٢، ١٤ مللم على التلوين و صفات الجودة فى الحبات.

و لقد أوضحت النتائج أن رش العنب صنف الفليم سيدلس بحمض الجبريليك بمفردة أدى لزيادة كل من وزن العناقيد،وزن الحبات و كذا محصول الكرمة معنويا فى حين أدى إلى نقص محتوى الحبات من صبغة الأنثوسيانين و كذلك نسبة المواد الصلبة الذائبة إلى الحموضة الكلية فى عصير الحبات مقارنة بالكنترول. علاوة على ذلك فإن الرش بالاثيفون بتركيز ٢٥٠ جزء فى المليون على الكرمات عندما وصل قطر الحبات ١٤ مللم أدى إلى زيادة وزن الحبات و المحصول معنويا بالمقارنة بتلك التى تم رشها بالاثيفون بنفس التركيز عند قطر الحبات ١٠، ١٢ مللم أو تلك التى تم رشها بحمض الجبريليك. كما أدت تلك المعاملة إلى إسراع الجمع و ذلك من خلال أثرها على زيادة نسبة المواد الصلبة الذائبة إلى الحموضة فى عصير الحبات، علاوة على ذلك فإن رش كرمات العنب بالاثيفون بتركيز ٢٥٠ جزء فى المليون عندما وصلت قطر الحبات ١٤ مللم أظهرت تلوين ممتاز بالإضافة الى زيادة محتوى الحبات من صبغة الأنثوسيانين بالمقارنة بتلك التى تم رشها بحمض الجبريليك أو تلك غير المعاملة. لذلك يمكن التوصية برش العنب صنف الفليم سيدلس بالاثيفون بتركيز ٢٥٠ جزء فى المليون عندما وصل حجم الحبات ١٤ مللم تعتبر مناسبة عن تلك التى تم رشها عندما وصلت حجم الحبات ١٠،١٢ مللم أو تلك المرشوشة بحمض الجبريليك فقط.