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RESEARCH ARTICLE

Effect of methyl jasmonate, abscisic acid spraying on improving coloration and quality of flame seedless under Upper Egypt climate

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Abstract

Fruit color development in table grapes growth and fruiting are affected by hot weather mainly in warm regions. Here, 2-year field study, during 2017/2018 and 2018/2019 seasons in a factorial experiment in randomized complete block design were conducted to study the effects of some of tested materials and time/stage of spraying on Flame Seedless grapevines grown in Upper Egypt and harvested during 15th May to 10th June. In this time, global and domestic markets of table grape are affected by quality of fruits focusing color in colored varieties. Thus, prices decrease considerably in the market. On other contrary, harvesting early (before mid of May) leads to at least a 50% increase in the total income. In this experiment methyl jasmonate and ABA are used at verasion stage followed by ethephon one time compared to spraying ethephon twice. all tested treatments are sprayed at different time/stages at 25 and 50 % colored bunched and the findings did not affect significantly on the yield components while using ABA or methyl jasmonate followed by ethephon one time improved berry quality in terms of berry color and firmness compared by spraying ethephon twice , on the other hand using methyl jasmonate and ABA affect significantly on reducing the uncolored and the best results were when spraying at 95 % berry softening approximately and 50% bunch coloring especially the methyl jasmonate. It could be recommended to use methyl jasmonate followed ethephon at 50% bunch coloring to improve berry and more coloring and keeping the berry quality after harvesting.

Keywords: Ethephon; Grapes; Methyl jasmonate; Verasion; Yield.

1. Introduction

Grape is one of the most important fruit crops for local consumption and export. The total world area of grapes reached 10.5 million ha with a total production of 89 million ton fruits (FAO, 2015). In Egypt grapes are an economically important crop and cultivated area was 197293 feddan that produced about 1734424 ton of fruits. It's one of the most important export horticultural crops and its export value is about 10% while the quantity is about 3% of total horticultural export (MALR, 2019). Flame Seedless cultivar is recognized as an important commercial and early cultivar in the Egyptian market, hence it has a great importance either for the local or international markets which exported to European and Arabian countries. The

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commercial value of table grapes is affected by their external appearance, including berry color. Therefore, poor coloring of red table grapes is a major problem facing the producers. Grown in worm climate zones, like Flame Seedless, is an issue and reduces the efficiency of grape production (Peppi *et al.,* 2006). Poor coloring of red grapes grown in warm regions substantially reduces the economic value of table grapes. Cultural practices such as leaf removal, shoot thinning, and cluster thinning can enhance the quality of Flame Seedless grapes, but often these practices are insufficient to repair color problems (Dokoozlian and Hirschfelt, 1995).. To enhance berry color, growers generally spray ethephon, an ethylene-releasing compound, but its effects on color are discordant, and the concentrations commonly needed for color improvement often reduce fruit firmness (Jensen *et al.,* 1975; Jensen *et al.,* 1982; Szyjewicz *et al.,* 1984). The recommended practices for achieving accepted berry color such as, Ethephon spraying and foliage management improve berry color of Flame Seedless berries (Schrader *et al.,* 1994), but in many times, it may remain unacceptable colored. This can be seen in subtropical or worm areas (Kliewer, 1970; Schrader *et al.,* 1994; Spayd *et al.,* 2002). Therefore, it was urgent require to find other co-effective solutions able to improve berry color without resulting excessive berry softening Few studies suggest using the external using of Methyl Jasmonate to improve the accumulation of anthocyanin content. During the last few years Colorsave is a commercial product used as a source of Methyl Jasmonate treatments, which contained 15.5% of Methyl Jasmonate (Khan and Sing, 2007and Portu *et al.,* 2015). Ethephon (2-chloroethyl phosphonic acid) speed up the ripening process of many fruits, including red table grapes produce ethylene when it starts to degraded. Ethephon (Ethrel 480 g Ethephon /l) as a trade name commonly are used on poor colored red table grape cultivars to enhance Jasmonate are considered as endogenous regulators that contributed in many important functions, including defense against insects and pathogens by prompt phytoalexin production, protection from abiotic stresses, impunity, and plant growth, suggesting that they have critical roles in plant physiology (Avanci *et al*., 2010 and Aubert *et al*., 2015).

Jasmonic acid biologically like to abscisic acid (ABA) has been shown to exhibit a senescence-promoting activity in the leaves of many plant families (Yilmaz *et al.*, 2007). Induced senescence by Jasmonic acid is described by a drastic loss of chlorophyll, the damage inhibition of its biosynthesis and increases in the respiratory rate and in protease and peroxidase activities. (Koda,1992). In addition, Jasmonic acid enhanced the resistance of tissues against decay by improving their antioxidant system and their free radical scavenging capability and there is a positive interconnection between antioxidant activity and anthocyanin content in berry skin. (Wang and lin, 2000; Lalel *et al.,* 2003; Khan and Singh, 2007; Kondo *et al.,* 2007). Many previous studies suggested that the exogenous treatment of Abscisic acid (ABA) increases the anthocyanin content in red table grape berries (Peppi *et al.,* 2006; 2007a; Cantin *et al.,* 2007; Peppi *et al.,* 2008). ABA was found more effective in enhancing grape berry color development than Ethephon (Peppi *et al.,* 2006; Cantin *et al.,* 2007; Roberto *et al.,* 2012). ProTone is the commercial product used as a source of ABA treatments, it contained 10% of effective material ABA, but it still expensive in comparison with Ethephon product. So, the aim of the current study was to investigate the effect of the vital role of timing and concentration of methyl Jasmonate and Abscisic acid mixed with Ethephon applications on enhancing the poor coloring and fruit quality of Flame seedless grapevines cultivated in warm regions.

2. Materials and Methods

The current Experiments were carried out during two consecutive seasons (2017/2018 and 2018/2019) on own-rooted ‘Flame Seedless’ grapevines cultivated in 2008 at private orchard located at Sohag governorate, Tema district. The soil is sandy under drip irrigation system (0.5 m between dripper). The Spanish Barron system was used as a trellising system, vines were short cane-pruned. Vines were spaced 2 m within rows and 3m between rows. Each row was oriented north to south. Vines were cane pruned (84 buds/vine were left, 12 canes x 6 eyes/cane plus of 6 spurs with 2 eyes) during the last week of December in each season. Crop load at all vines was adapted to 25 clusters/vine after berry set. All vines received the Standard cultural practices that are used in vineyard. The experiment was set up as a complete randomized block design in spilt plots with three replicates each one consists of three vines, and then the total experimental vines were seventy two vines.

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2.1. The first factor was as follows

A1: Verasion stage (95 % berry softening approximately and 25% bunch coloring).

A2: Verasion stage (95 % berry softening approximately and 50% bunch coloring).

2.2. The second factor was as follows

B1. Control treatment (sprayed with water)

B2. Ethephon twice 250 ppm and 250 ppm three days later.

B3. Methyl Jasmonate 3 ml/L and Ethephon at 250 ppm.

B4. ABA 1.5 ml/L and ethephon at 250 ppm.

Ethrel 720 SLTM is the commercial product of Teda and contains 720 g ethephon /liter. ProTone SLTM is the commercial product of abscisic acid (ABA) and contains 10% ABA. While, ColorsaveTM is the commercial product of Methyl jasmonate and contains 15.5% methyl jasmonate.

Bunches were harvested when achieved the minimum requirement, berries of each bunch became red (more than 90% of berries/bunch). The following parameters were measured on selected vines during the two studied seasons.

2.3. Cluster and berry characteristics

Sample of 9 clusters per treatment, each replicate consists of 3 clusters were harvested and transported to estimate physical and chemical characteristics of berries and clusters.

Cluster weight (g) and berry weight (g) were determined by an electrical sensitive balance.

Total soluble solids (TSS %): This was estimated as a percent in juice of fresh berries, a Carl Zeiss hand refractometer was used in that respect. Reducing sugars was determined as outlined in A.O.A.C. (1985).

Total acidity content (%) was estimated using titration of 5 ml clear juice against (0.1 N) NaOH after the addition of a little drops of phenolepthalene indicator.

Total anthocyanin: The anthocyanin pigments were extracted by ethanolic HCl, a mixture of 95% ethanol and 1.5 M HCl acid (85:15 v/v). A sample of 0.5 g from berry skin was ground and kept overnight with about 20 ml of the solvent. The samples were then washed by aliquots of ethanolic HCl several times until the berry skin samples were colorless. The mixture was filtrated through a centered glass funnel 6.3 and extract was transferred to 25 ml volumetric flask and completed to volume with the acidified alcohol then measured on spectrophotometer at wave length 530. The anthocyanin content was determined from the standard calibration curve of cyaniding-3-glucosid as pointed out by Markham (1982). Uncolored bunches (loss %): number of uncolored bunches from each replicate/ total bunch number *100

Yield (Kg/vine): After harvest the yield in weight (kg) was determined by multiplying number of clusters/vine by the cluster average weight.

Bunch weight: Weight of 25 berries

Berry Texture Analyzer: The berries of each sample were weighed and subjected to tests with a fruit texture analyzer instrument (Fruit Hardness Tester, No. 510-1) as a small cylinder used a flat plate traveling at a speed of 5mms to compress each whole berry by 3 mm. Peak 1 force expressed in (g /cm) was recorded and considered 2 to be an indicator of fruit firmness.

All the obtained data were tabulated and analyzed according to Gomez and Gomez (1984) and Snedecor and Cochran (1990) using L.S.D. test for distinguishing the significance differences between various treatments means according to Steel and Torrie (1980).

3. Results

The findings in Table 1 indicate the unassuming effects of the evaluated treatments in both seasons on yield/vine, bunch weight and weight of 25 berries. The obtained results are logical because all treatments applied after cell division stage. In addition, the interaction between the spraying materials and stage of spraying showed the insignificant differences in both seasons. This result is in a harmony with those of (Peppi *et al.*, 2007b; Roberto *et al.,* 2012) they reported that no significant differences among Ethephon and ABA treatments were obtained in terms of berry mass. During the 1st season of study it could be found that potassium at 1.5%, amino acids at 2%, micronutrients at 5% and calcium at 2.5% produced the highest values. During the 2nd season data presented in such table suggested that the obtained results in Table 2 indicated that the tested treatments significantly improved the berry quality in terms of increasing the TSS and reducing sugar, while the tested treatments reduced titratable acidity percentage substantially in both seasons. The time/stage of spraying either at 50 or 25 colored bunches percentage induces a significant increase of TSS and reducing sugar and during the two studied seasons, respectively. In addition Treatment by Methyl Jasmonate or ABA one time followed by Ethephon one time followed by Ethephon had recorded the highest TSS percentage (14.10 & 14.68) & (14.40 & 14.84) compared to spraying Ethephon twice and control (13.9 & 14.26) and without significant differences between them in both seasons, respectively. The interaction between the spraying materials and time/stage of spraying showed the significant differences in both seasons these results are in a harmony with (Peppi *et al.,* 2006; Osman and Mohsen, 2015). The obtained data in Table 3 , showed that the time/stage of spraying either at 50 or 25 colored bunches percentage induces a significant increase concerning the percentage of uncolored bunches percentage of uncolored bunches when spraying at stage (95 % berry softening approximately and 25% bunch coloring (15.73 & 14.64) in both seasons respectively compared to (9.56 & 10.24) when spraying at 95 % berry softening approximately and 50% bunch coloring with loss percentages (38.71 & 30.01) in the both seasons respectively. While spraying methyl jasmonate followed by one time ethephon at Verasion stage (95 % berry softening approximately and 25% bunch coloring) increased the uncolored bunched percentages (15.73 & 14.64) compared to spraying the same treatment at Verasion stage (95 % berry softening approximately and 50% bunch coloring (9.56 & 10.24) in addition to spraying methyl jasmonate followed by one time ethephon clarified strong significant effects (5.45 & 4.36) in both seasons compared with spraying ethephon twice or spraying ABA followed by one time ethephon and control (11.43 & 9.05) (3.27 & 3.38)in both seasons respectively.) compared to spraying Ethephon twice and control (13.9 & 14.26) without significant differences between them in both seasons, respectively. The interaction between the spraying materials and time/stage of spraying showed the significant differences in both seasons.

Table 1. Influence of Methyl Jasmonate and Abscisic acid on bunch weigh, yield / vine and weight of 25 berries of Flame Seedless grapes during 2017/2018 and 2018/2019 seasons.

	Bunch weight (gm)						yield / vine (kg)						weight of 25 berries (gm)					
A	2017/2018			2018/2019			2017/2018			2018/2019			2017/2018			2018/2019		
B	A1	A2	Mean	A1	A2	Mean	A1	A2	Mean	A1	A2	Mean	A1	A2	Mean	A1	A2	Mean
B1	303.3	209.3	301.4	296.6	305.2	300.9	7.5	5.2	6.35	7.4	7.6	7.5	63.18	43.6	53.39	61.79	63.58	62.68
B2	304.4	292	298.2	305.3	302.7	304	7.6	7.3	7.4	7.6	7.5	7.55	63.41	60.83	62.12	62.60	63.60	63.23
B3	308.5	318.6	313.6	314.7	310.8	312.7	7.7	7.9	7.8	7.8	7.7	7.75	64.27	66.37	65.32	65.56	64.75	65.15
B4	309.3	321.3	315.3	313.4	304.5	313	7.7	8.0	7.8	7.8	7.6	7.7	64.43	66.93	65.68	65.29	63.43	64.36
Mean	306.4	307.8		307.5	308.3		7.6	7.6		7.65	7.6		63.82	63.82		63.82	63.82	
LSD$_{0.05}$ A		NS			NS			NS			NS			NS			NS	
LSD$_{0.05}$ B		NS			NS			NS			NS			NS			NS	
LSD$_{0.05}$ A * B		NS			NS			NS			NS			NS			NS	

Table 2. Influence of Methyl Jasmonate and Abscisic acid on TSS %, acidity and reducing sugar of Flame Seedless grapes during 2017/2018 and 2018/2019 seasons.

	Reducing sugar%						titratable acidity %						TSS %					
A	2017/2018			2018/2019			2017/2018			2018/2019			2017/2018			2018/2019		
B	A1	A2	Mean	A1	A2	Mean	A1	A2	Mean	A1	A2	Mean	A1	A2	Mean	A1	A2	Mean
B1	10.60	10.89	10.75	10.79	10.73	10.76	0.65	0.58	0.62	0.56	0.61	0.59	12.93	13.10	13.02	13.00	12.86	12.93
B2	10.85	12.05	11.45	11.08	12.52	11.80	0.56	0.45	0.51	0.53	0.44	0.49	13.30	14.50	13.90	13.41	15.10	14.26
B3	11.40	11.75	11.58	11.90	12.35	12.12	0.45	0.44	0.45	0.43	0.41	0.42	13.86	14.35	14.10	14.45	14.90	14.68
B4	11.63	12.10	11.87	12.03	12.35	12.19	0.46	0.42	0.44	0.44	0.42	0.43	14.10	14.70	14.40	14.57	15.10	14.84
Mean	11.13	11.67		11.45	11.99		0.53	0.47		0.49	0.47		13.55	14.16		13.86	14.50	
LSD$_{0.05}$ A		*			*			*			*			*			*	
LSD$_{0.05}$ B		0.34			0.38			0.041			0.039			0.43			0.48	
LSD$_{0.05}$ A * B		0.48			0.55			0.058			0.055			0.61			0.67	

Table 3. Influence of Methyl Jasmonate, Abscisic acid and Ethephon on uncolored bunches percentage of Flame Seedless grapes during 2017/2018 and 2018//2019 seasons.

A / B	2017/2018			2018/2019		
	A1	A2	Mean	A1	A2	Mean
B1	32.23	28.67	30.45	34.50	31.0	32.75
B2	19.35	3.50	11.43	14.83	3.33	9.05
B3	7.12	3.73	5.45	5.33	3.80	4.36
B4	4.17	2.37	3.27	3.90	2.85	3.38
Mean	15.73	9.56		14.64	10.24	
$LSD_{0.05}$ A		*			*	
$LSD_{0.05}$ B		1.08			1.19	
$LSD_{0.05}$ A * B		1.56			1.69	

Methyl jasmonate at verasion stage (95 % berry softening approximately and 50% bunch coloring).

Methyl jasmonate at verasion stage (95 % berry softening approximately and 25% bunch coloring).

The findings in Table 4, in both seasons, noticed that no significant differences concerning the time/stage of sparing either at Verasion stage 95 % berry softening approximately and 25% or 50 % bunch coloring. On the other hand, the tested materials significantly decreased berry firmness. The treatment of spraying Ethephon twice significantly reduced berry firmness gaining the lowermost berry firmness (111 & 126 gm/cm2) in both seasons, respectively. Compared to control treatment, while Compared to Methyl Jasmonate followed by ethephon recorded the highest berry firmness force (189 & 201 gm/cm2), without significant differences between them in the two seasons respectively. The interaction between the spraying materials and time/stage of spraying showed the significant differences in both seasons. This result was agreed with reported by (Cantin *et al.*, 2007).

Table 4. Influence of Methyl Jasmonate and Abscisic acid on berry firmness (gm/cm2) of Flame Seedless grapes during 2017/2018 and 2018/20 19seasons.

A / B	2017/2018			2018/2019		
	A1	A2	Mean	A1	A2	Mean
B1	134	121	128	140	135	138
B2	119	103	111	125	126	126
B3	192	186	189	203	198	201
B4	185	173	179	201	200	201
Mean	158	146		167	165	
$LSD_{0.05}$ A		NS			NS	
$LSD_{0.05}$ B		10.11			11.61	
$LSD_{0.05}$ A * B		14.56			NS	

With regard to anthocyanin content they obtained data in table (5), significant differences were noticed among the tested treatments in both seasons. A superior significant difference concerning the time/stage of sparing either at Verasion stage 95 % berry softening approximately and 25% or 50 % bunch coloring on the other hand It could be noticed that spraying with Ethephon combined with Methyl Jasmonate (1.05 & 1.01 mg/kg) and Ethephon combined with (1.01 & 1.01 mg/kg) gained the highest anthocyanin content in both seasons, followed by the treatment of spraying Ethephon twice (0.779 & 0.81 mg/kg) in the two seasons, respectively.

Table 5. Influence of Methyl Jasmonate and Abscisic acid on anthocyanin (mg/kg) of Flame Seedless grapes during 2017/2018 and 2018/2019 seasons.

A / B	2017/2018			2018/2019		
	A1	A2	Mean	A1	A2	Mean
B1	0.55	0.63	0.59	0.59	0.52	0.56
B2	0.60	0.98	0.79	0.63	0.99	0.81
B3	1.02	1.07	1.05	0.97	1.04	1.01
B4	0.98	1.04	1.01	0.99	1.03	1.01
Mean	0.79	0.93		0.80	0.90	
$LSD_{0.05}$ A		**			*	
$LSD_{0.05}$ B		0.061			0.056	
$LSD_{0.05}$ A * B		0.085			0.081	

4. Discussion

Endogenous concentrations of ABA were closely related to the increase of soluble solids and the decrease of titratable acidity of grapes that occur during maturation (Coombe, 1976; Du¨ring *et al.*, 1978). Application of ethephon is known to sometimes decrease the acidity of grape juice (Jensen *et al.*, 1975; Szyjewicz *et al.*, 1984) and Lee *et al.* (1997) observed that at harvest, ABA-treated and nontreated fruits had similar soluble solids.

Jasmonic acid was found to be contributed in plant response to injury and biotic stress, such as resistance against insects and pathogens aggression (Shan *et al.,* 2009; Wasternack and Hanse, 2013). It also has a strong relevance with other hormones such as ABA, ethylene, and auxins (Memelink *et al.,* 2001; *Sasaki et al.,* 2001), in addition, Haifeng Jia1 *et al.* (2015) confirmed that JA plays a positive and vital role in the grape fruit ripening. Fruit-ripening is associated with the coloring, cell wall softening.

Perez *et al.* (1997) mentioned that treatment of jasmonic acid to immature green strawberries has enhanced respiration, ethylene production, and transitory induction of anthocyanin biosynthesis and chlorophyll deflection, which proves the vital role in ripening of this fruit. Also, Wang and lin (2000) reported that JA enhanced the resistance of tissues against decay by increased the antioxidant system and their free radical scavenging potency and there is a positive liaison between antioxidant activity and total phenolic or anthocyanin content. Moreover, Aubert *et al.* (2015) found that a positive effect of Jasmonic acid could be attributed to imrove the biosynthesis of such pigments. Jasmonate are contributed in many important functions, including safeguard against pathogens and insects by prompt phytoalexin in production, impunity, and plant growth, suggesting that they have vital roles in plant physiology Avanci *et al.* (2010). Also, Sabry Gehan *et al.* (2011) reported that treament with Jasmine oil concentrations especially at 0.2%oil + 3% dormex increased total anthocyanin of berry skin.

5. Conclusion

According to the findings it could be debrief that the treatment with Methyl Jasmonate could be a promising material and important application in the field for enhancing berry quality, enhancing berry anthocyanin content, decreasing the uncolored bunches compared to ABA treatments from the cost wise point of view. so. It could be recommended, using Methyl Jasmonate combined with Ethephon to improve berry anthocyanin content and keeping the quality after harvesting. Provided that spraying time at Verasion stage (95 % berry softening approximately and 50% bunch coloring). To avoid losses (uncolored bunches).

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