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FLAME SEEDLESS AND RED GLOBE CVS. BERRIES QUALITY IN RELATION TO SPRAYING ETHYLENE AND ABA

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ABSTRACT

Flame Seedless and Red globe considered as most popular grape cultivars in Egypt, successfully grown under different soil and climate conditions. Under Al-Minia conditions, these two *cvs.*, faces some problems such as poor chemical characteristics of berries especially poor coloration. In order to improving the physical and chemical characteristics of these two cultivars, field experiment was achieved, included nine treatments: Control, spraying abscisic acid or ethylene four concentrations (100 ppm, 200 ppm, 300 ppm and 400 ppm). Each treatment was replicated three times, one vine per each. All treatments were frequently applied three times.

The positive effect of ABA treatments were more clear on berry physical properties “represented by berry weight and berry dimensions” than those of ethylene treatments for the two *cvs*. Furthermore, the response of “Red globe” cluster and berry physical properties to spraying ABA was clearer than those of “Flame seedless”. The berry chemical properties of the two cultivars significantly enhanced as a result of spraying the two examined materials (Ethylene or ABA). However, this positive effect was clearer for ethylene treatments than those of ABA. The obtained data confirmed that, non-significant differences were observed between the two higher concentrations neither for physical properties nor for chemical properties.

Key words: Grapevines, Flame Seedless, Red globe, abscisic acid, ethylene, fruit quality.

INTRODUCTION

Grapevines (*Vitis vinifera L.*) considered as one of the most important and oldest fruit croup all over the world. It is widely cultivated in most countries of the world (**Winkler *et al.*, 1974; Delas 2000; Doring *et al.*, 2015 and Metawie 2020**). Grapes have a high nutritional value due to their contents of sugars, minerals (especially K), vitamins, amino acids and organic acids (**Metawie, 2020**

Flame Seedless considered as one of the most popular grape cultivar in Egypt successfully grown under different soil and climate conditions. This cultivar is one of the early ripens cultivars **(Saad, 2014 and Ibrahim *et al.*, 2020)**. It has a medium cluster with red or red-purplish berries with small to medium berry size and crunchy pulp **(OIV 1988** and **Galet 2015)**. Red globe cultivar is one of the Californian table grapes cultivars with highly fertile bud. It characterized by large and loose cluster and round or prostrate large or very large berries with not very juicy pulp, the berry colour vary from light pink to dark red (**OIV 1988** and **Galet, 2015**). This seeded cultivar is the most popular grapes in Asia, Middle East and Eastern Europe (**OIV, 1988** and **Galet, 2015**). Under Al-Minia conditions, these two important cultivars faces some problems such as poor yield and poor coloration of berries as well as shot berries, which in turn negatively affect marketing. More detailed studies are required for better understanding of poor berries colouring of Flame seedless and Red globe cultivars under Al-Minia Governorate climatic conditions. Based on this, ethylene and abscisic acid were sprayed at different concentrations on these two cultivars, as an attempt to overcome these problems under Al-Minia governorate conditions.

MATERIAL AND METHODS

This study was carried out during two consecutive seasons "2020 and 2021" on 27 vines from each cultivar (Flame seedless and Red globe) uniformed in vigor and12- years old for "Flame seedless" and 10 years old for "Red globe" grown in a private vineyard located at Al-Sharawyah village, Samalout Distract, Minia Governorate – Egypt, where the soil texture is clay and well drained water. Cane pruning system was followed at the first week of January, leaving 84 eyes per vine (on the basis of eight fruiting canes x 9 eyes plus six renewal spurs x two eyes) with the assistance of gabel shape supporting system. The vines were irrigated through surface irrigation system using Nile water.

Soil analysis: Physical and chemical analysis of orchard soil were carried out at the start of the experiment according to **Walsh & Beaton** (**1986**).

Experimental work: This experiment included the following nine treatments: Control (sprayed with tap water), Spraying Ethylene at 100 ppm, Spraying Ethylene at 200 ppm, Spraying Ethylene at 300 ppm, Spraying Ethylene at 400 ppm, Spraying ABA at 100 ppm, Spraying ABA at 200 ppm, Spraying ABA at 300 ppm and Spraying ABA at 400 ppm. Each treatment was replicated three times, one vine per each. All treatments were frequently applied three times during the growth cycle of vines. The ethylene were done at starting of the veraison stage (coloring 5% of berries), at coloring 50% of berries and at complete berries coloring. While, the ABA treatments were carried out three times; just after fruit setting, starting of veraison stage (coloring 5% of berries) and at complete berries coloring.

Experimental design: Randomized complete block design was followed where the present experiment consisted of 9 treatments, each treatment was replicated three times, one vine per each. Then, the present study contained 27 vines from each cultivar (Flame Seedless and Red globe). Then, this experiment included 54 vines used as a plant material to achieving this study.

Different determinations: The following determinations were achieved during the two experimental seasons:

Berry physical and chemical properties: 100 berries were randomly taken from the four clusters, which previously harvested, in order to determination the following physical and chemical characteristics.

1- Average berry weight (g). Average berry dimensions (longitudinal and equatorial, in cm).
2- Total soluble solids (TSS%) in the berries was achieved by using handy refractometer.
3- Percentage of reducing sugars in berry juice using volumetric method as described by A.O.A.C. (2000).
4- Percentage of titratable acidity (as a grams of tartaric acid/ 100 g of juice) by using titration against 0.1N NaOH in the presence of phenolphthalein as an indicator Rangana 1985 and AOAC. (2000).
5- Total anthocyanins: Total anthocyanins in berries juice were extracted and determined according to Singleton & Rossi (1965) and Fulcki & Frabcis (1968).
6- Separation and quantification of the main compounds of anthocyanins pigment was carried using 1200 Series HPLC–DAD (Algilent Technologies – Santa Clara, CA, USA) system, using a reversed phase chromatography Colum, according to the program developed by Paun et al., (2022).

Statistical analysis : The obtained data were tabulated and subjected to the proper statistical analysis using the analysis of variance (ANOVA). Comparisons between means were made by the least significant differences (New L.S.D) at p = 0.05 (Snedecor and Cochran, 1990).

RESULTS AND DISCUSSION:

1- Effect of ethylene and ABA on berry physical properties:

The results pertaining to the effect of spraying ethylene and ABA on berry physical properties (i.e. berry weight, berry length and berry diameter) of Flame seedless and Red globes cultivars are presented in Table (2).

1-1: Average berry weight (g): The perusal of data reveals that, during the first seasons, all ethylene treatments hasn't exerted any significant effect on the berry weight of Flame seedless *cv*. regardless the concentration used. Contrary of the first season, the higher concentrations of ethylene (300 and 400 ppm) exerted a significant effect on Flame seedless and Red globe *cvs.* berry weight rattier than untreated vines.

It can be visually observed from the representative data that all ABA concentrations were capable to increase the weight of "Flame seedless" and "Red globle" berry weight during the two experimental seasons. This increment in berry weight was parallel to increasing ABA concentration.

The vines received the higher concentration of ABA present the higher berry weight, during the two experimental seasons respectively. On the opposite side, untreated vines present the lowest berry weight, during the two seasons respectively.

1-2: Berry dimensions: data showed in the same Table present the effect of spraying ethylene and ABA at 100 to 400 ppm on berry length and diameter, during 2020 and 2021 seasons. It is clear from this Table that, only the highest concentration of ethylene was capable to increase the berry length of the two examined cultivars during the two experimental seasons, except the case of Red globe in the second season whereas non-significant differences were observed. Furthermore, ethylene treatment hasn't exerted any significant effect on the berry diameter of Flame seedless *cv.*, whereas the berry diameter of Red globe *cv* present a significant response to the higher concentration of ethylene.

On the other hand, spraying ABA was very impressive effect on berry length and diameter of the two examined cultivars; these findings were true during the two experimental seasons (2020 and 2021). However, all ABA concentrations were capable to improve significantly the berry length (cm) and berry diameter (cm). This increment was parallel to increasing the concentration of ABA. The vine received the highest concentration of ABA present the highest berry length and berry diameter, on the contrary, untreated vines present the lowest berry length and berry diameter, these data were true during the two experimental season. Furthermore, non-significant differences were observed between the two higher concentrations, neither in the first season nor in the second season.

The obtained data during the present study are in harmony with those obtained by **Wheeler (2006)** and **Giribaldi *et al.*, (2010)** on "Cabernet-Sauvignon" grapevines which studied the effect of ABA on berry growth and maturation.

2-Effect of ethylene and ABA on berry chemical properties:

Data presented in Table (3) shows the effect of spraying ethylene and ABA at different concentrations (100 to 400 ppm) on different berry chemical parameters (i.e. TSS%, total acidity%, reducing sugars % and total anthocyanins) of Flame seedless *cv.* and Red globe *cv.* berries during 2020 and 2021 seasons

2-1: TSS% and total acidity: It is clear from this Table that treating Flame seedless and Red globe cultivars three times with ethylene or ABA at 100 to 400 ppm significantly was responsible for stimulating TSS% and relative to the control treatment. There was a gradual promotion in TSS% parallel to increasing the concentrations of ethylene or ABA gradually. Significant differences were observed during the two experimental seasons, except the case of low concentration of both materials (ethylene and ABA). These data were true during the two experimental seasons.

Regarding the effect of spraying ethylene or ABA on total acidity (Table 3), during the two experimental season, it is clear that the Flame seedless response was clearer than those of Red globe cultivar to ethylene and ABA application. Whereas, all ethylene concentrations were able to induce significant decrease the total acidity in Flame seedless *cv*. berries. Concerning the total acidity of Red globe *cv.*, only the highest concentration (400 ppm) in first season and the two highest concentrations (300 and 400 ppm) during the second season were capable to reducing significantly the total acidity of berries in both examined cultivars (Table 4).

2-2: Reducing sugars %: data illustrated in Table (4) showed the effect of spraying ethylene and ABA on berry reducing sugars % of Flame seedless and Red globe cultivars, during 2020 and 2021 seasons. It is clear from this Table that spraying ethylene or ABA at 100 to 400 ppm on Flame seedless and Red globe grapevines *cvs.* had a positive significant effect on the berry reducing sugars contents, during the two seasons. So, a remarkable and gradual significant increase in reducing sugars % were observed. This increment was parallel to increasing the concentration used from each material (from 100 to 400 ppm). However, all ethylene and ABA concentrations were capable to significant increase the reducing sugars in berry, except the case of low ethylene concentration on “Red globe” and the low concentration of ABA on the two cultivars during the first season. Furthermore, the vines received the highest concentration of ethylene present the highest reducing sugars%. Contrary, the lowest total reducing sugars were obtained from untreated vines, during the two experimental seasons respectively.

2-3: Total anthocyanins (mg/100g F.W.): It is obvious from Table (3) that, subjected Flame seedless and Red globe cultivars to three sprays of ethylene and ABA (at 100, 200, 300 and 400 ppm) was significantly accompanied with increasing berry total anthocyanins, during the two experimental seasons. Except the case of low concentration of ABA which hadn’t any significant effect on total anthocyanins of the two examined cultivars. This increment was parallel to increasing the concentration used from 100 to 400 ppm. Furthermore, the vines received the highest concentration of ethylene present the highest total anthocyanins in there berries. Contrary, the lowest total anthocyanins were obtained from untreated vines, during the two experimental seasons respectively. It’s worth to mentioned that, non-significant differences were observed between the two higher concentrations (300 and 400 ppm), neither for ethylene nor for ABA, during the two experimental seasons.

Ban *et al.*, (2003) mentioned that spraying ‘Kyoho’ grape berries with ABA significantly enhanced the biosynthetic of main compounds of anthocyanins in the skin of berry during ripening stage. Regarding the effect of ABA at different concentrations (from 100 to 400 ppm) on Flame seedless and Red globe berry chemical properties (i.e. TSS%, TSS/Acidity, Reducing sugars % and total anthocyanins) are in harmony with those obtained by other authors on different fruit trees, such as: **Koussat *et al.*, (1993)** and **Giribaldi *et al.*, (2010)** on "Cabernet-Sauvignon" grapevines; **Han *et al.*, (1996)** on coloration and fruit quality in "Kyoho" grapevine during the ripening stage; **Cantın *et al.*, (2007)** on "Crimson Seedless" grapes; **Zhang & Dami (2012)** on Grapevines cultivar Chambourcin; **Tihero *et al.*, (2016)** on Sweet cherry (*Prunus avium* L.); **Villalobos-Gonzalez (2016)** on "Carmenere" grapevine.

On the contrary of our findings, **Zhang and Dami (2012)** while studding on the effect of ABA on quality and freezing tolerance of 'Chambourcin' Grapevines grown under Ohayo state (USA). The authors don't observe any significant effect of ABA treatments at 400 to 600 ppm on TSS%, reducing sugars% and anthocyanins contents.

2-4: Anthocyanins main compounds identified by HPLC MS: The data presented in Table (4) and Figures (1, 2, 3 and 4) shows the effect of spraying ethylene and ABA at 100 to 400 ppm on the berry skin main anthocyanins compounds of Flame seedless and Red globe cultivars during 2020 and 2021.

HPLC profile for treated flame seedless (Figures 1 & 2 and Table 6) with ethylene and abscisic acid showed higher amount in Epigallo catechin gallate and Trimethoxyflavone. While, treated red globe (Figures 3 & 4 and Table 6) proved to have (+) Catechin, hydroxy flavone and flavanone derivatives were obvious as shown comparing to the untreated samples.

In previous study, Setha (2012) found in berry skin delphinidin, cyanidin, petunidin, peonidin and malvidin 3,5-diglucoside using HPLC mass spectrum. As stated in the table 6, certain compounds didn't appear in untreated samples giving indicator for the importance of treatment such as disappearance of (+) catechin in untreated red globe. As well, trimethoxy flavone only appeared in flame seedless samples and didn't show in red globe treated or untreated samples. The flavonoid catechin has five hydroxy groups and commonly known as potent antioxidants. Hydroxy and methoxy groups in these derivatives showed importance as antioxidant and anticancer agents. Certain compounds didn't appear in some samples giving indicator for the importance of treatment as disappeared (+)catechin in untreated red globe. As well, trimethoxy flavone only appeared in flame seedless and didn't show in red globe treated or untreated samples. The flavonoid catechin has five hydroxy groups and commonly known as potent antioxidants. Hydroxy and methoxy groups in these derivatives showed importance as antioxidant and anticancer agents.

CONCLUSION:

Under Minia Governorate conditions in clay soil and similar conditions, it could be recommended to treat Flame seedless and Red globe grapevine cultivars with ethylene 300 ppm or ABA at 300 ppm three times yearly as a foliar application, in order to improve berry physical and chemical properties in order to produce good yield with high-quality.

Table (1): physical and chemical analysis of orchard soil

Sand %	**7.0**
Silt %	**21.5**
Clay %	**71.5**
Texture	**Clay**
pH (1: 2.5 extract)	**7.75**
E.C. (1: 2.5 extract)	**1.20**
O.M. %	**1.78**
$CaCO_3$ %	**2.85**
Total N %	**0.19**
Available P (Olsen method, ppm)	**2.44**
Available K (ammonium acetate, ppm)	**398**

Table 2: Effect of ethylene and abscisic acid at different concentration on berry weight (g), berry length (cm) and berry diameter (cm) of "Flame seedless" and "Red globe" grapevines cvs., during 2020 and 2021 seasons.

Berry physical properties						
First season (2020)						
Treatments	**Flame seedless**			**Red globe**		
	Berry weight (cm)	**Berry length (cm)**	**Berry diameter (cm)**	**Berry weight (cm)**	**Berry length (cm)**	**Berry diameter (cm)**
Control	1.91	1.59	1.50	7.29	3.24	3.11
Eth. At 100 ppm	1.94	1.69	1.54	7.25	3.45	3.19
Eth. At 200 ppm	1.99	1.72	1.55	7.34	3.44	3.18
Eth. At 300 ppm	1.94	1.77	1.58	7.85	3.49	3.22
Eth. At 400 ppm	2.09	1.95	1.61	7.90	3.55	3.29
ABA at 100 ppm	2.12	2.21	1.79	7.79	3.67	3.31
ABA at 200 ppm	2.29	2.39	2.01	7.99	3.73	3.49
ABA at 300 ppm	2.36	2.44	2.20	8.18	3.82	3.61
ABA at 400 ppm	2.38	2.59	2.22	8.24	3.88	3.66
New LSD 5%	**0.21**	**0.19**	**0.19**	**0.29**	**0.27**	**0.17**
Second season (2021)						
Treatments	**Flame seedless**			**Red globe**		
	Berry weight (cm)	**Berry length (cm)**	**Berry width (cm)**	**Berry weight (cm)**	**Berry length (cm)**	**Berry width (cm)**
Control	1.92	1.55	1.53	7.11	3.21	3.18
Eth. At 100 ppm	1.97	1.66	1.59	7.14	3.44	3.19
Eth. At 200 ppm	2.05	1.67	1.60	7.21	3.42	3.19
Eth. At 300 ppm	2.20	1.68	1.66	7.32	3.40	3.32
Eth. At 400 ppm	2.23	1.93	1.69	7.40	3.49	3.38
ABA at 100 ppm	2.22	2.29	1.66	7.89	3.55	3.39
ABA at 200 ppm	2.30	2.44	1.72	8.11	3.64	3.48
ABA at 300 ppm	2.40	2.69	2.07	8.29	3.88	3.59
ABA at 400 ppm	2.49	2.72	2.19	8.31	3.91	3.64
New LSD 5%	**0.22**	**0.17**	**0.18**	**0.21**	**0.24**	**0.19**

Table 3: Effect of ethylene and abscisic acid at different concentration on TSS%, Total acidity % and reducing sugars% and total anthocyanins of Flame seedless and Red globe grapevines cvs., during 2020 and 2021 seasons.

Chemical properties								
First season (2020)								
Treatments	**Flame seedless**				**Red globe**			
	TSS%	**Total acidity %**	**Reducing Sugars %**	**Total anthocy.**	**TSS%**	**Total acidity %**	**Reducing Sugars %**	**Total anthocy.**
Control	16.0	0.645	13.5	79	15.4	0.626	12.8	80
Eth. At 100 ppm	16.5	0.601	14.2	119	15.9	0.601	13.6	105
Eth. At 200 ppm	17.9	0.589	15.3	137	16.2	0.583	13.9	119
Eth. At 300 ppm	18.6	0.533	15.9	140	17.2	0.558	14.6	130
Eth. At 400 ppm	18.9	0.521	16.2	155	17.4	0.541	14.9	132
ABA at 100 ppm	16.1	0.632	14.0	108	15.4	0.601	13.3	84
ABA at 200 ppm	16.8	0.621	14.8	122	16.7	0.599	14.1	101
ABA at 300 ppm	17.5	0.598	15.0	137	16.8	0.587	14.5	119
ABA at 400 ppm	17.7	0.587	15.2	144	16.9	0.572	14.5	122
New LSD 5%	**0.8**	**0.043**	**0.7**	**21**	**0.7**	**0.044**	**0.8**	**19**
Second season (2021)								
Treatments	**Flame seedless**				**Red globe**			
	TSS%	**Total acidity %**	**Reducing Sugars %**	**Total anthocy.**	**TSS%**	**Total acidity %**	**Reducing Sugars %**	**Total anthocy.**
Control	15.9	0.622	13.0	82	15.7	0.622	12.6	75
Eth. At 100 ppm	16.7	0.533	14.4	121	16.9	0.604	14.2	112
Eth. At 200 ppm	17.9	0.521	15.8	139	17.7	0.587	14.9	125
Eth. At 300 ppm	18.4	0.514	16.3	156	17.9	0.543	15.1	139
Eth. At 400 ppm	18.8	0.501	16.8	169	18.2	0.522	15.3	143
ABA at 100 ppm	16.0	0.622	14.2	111	16.2	0.601	13.9	99
ABA at 200 ppm	16.9	0.593	14.8	123	16.9	0.585	14.7	109
ABA at 300 ppm	17.5	0.564	15.7	139	17.3	0.573	15.0	117
ABA at 400 ppm	17.9	0.556	15.9	143	17.5	0.561	15.2	120
New LSD 5%	**0.7**	**0.039**	**0.8**	**20**	**0.6**	**0.041**	**0.6**	**17**

Table 4: Peak assignment, retention time, mass special data, and percentage of anthocyanins of flame seedless and red globe (treated and untreated) using ESI/MS positive ion mode after: HPLC analysis

Mass Assignment	Red globe Anthocyanins %		Flame seedless Anthocyanins %		*m/z*	R_t min
	Nontreated	treated	Nontreated	treated		
2R,3S (+)Catechin $C_{15}H_{14}O_6$	-	3.35	3.37 Neg. mode	1.66	290	0.69
Unknown	5.82	3.84	3.45	5.57	381	0.82
Abscisic acid residue $C_{15}H_{20}O_4$	3.47	2.75	-	-	265	12.07
Dihydro Stilbene 4-O,2-prenyl	4.21	2.58	-	-	385	17.92
Unknown	4.81	6.38	5.94	5.47	401	18.34
Epigallo catechin gallate $C_{22}H_{18}O_{11}$	9.04	5.9	4.3	6.37	457	19.14
3’,4’-Dimethoxy-7-hydroxy flavanone $C_{17}H_{14}O_5$	12.01	13.27	13.69	11.12	298	19.52
5,4’-Dihydroxy-7-methoxy-flavone $C_{16}H_{12}O_5$	9.8	10.03	9.75	7.7	284	22.4
2’,3’,6-Trimethoxy flavone $C_{18}H_{16}O_5$	-	-	3.74	8.63	312	24.68

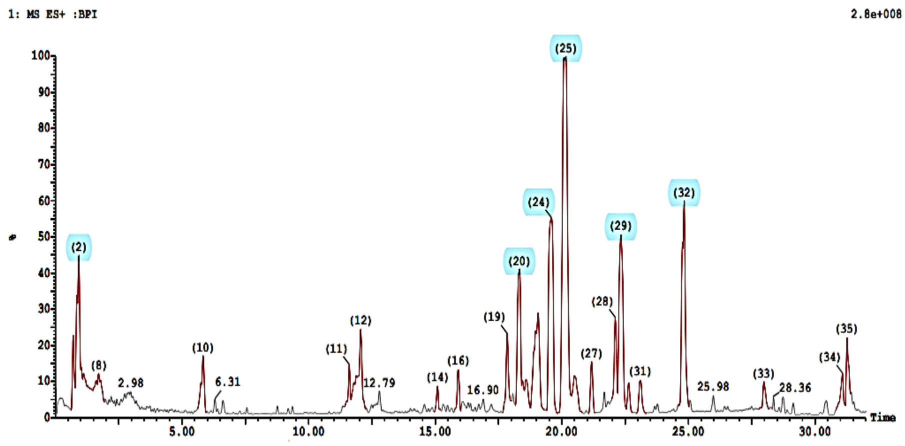

Figure 1: HPLC profile of anthocyanins of Flame seedless grapes treated with ethylene and abscisic acid at 400 ppm.

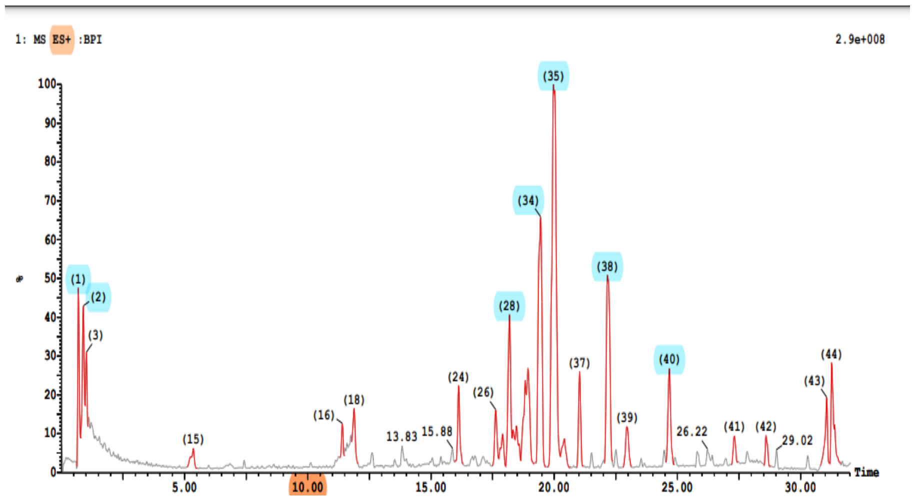

Figure 2: HPLC profile of anthocyanins of Flame seedless grapes extract

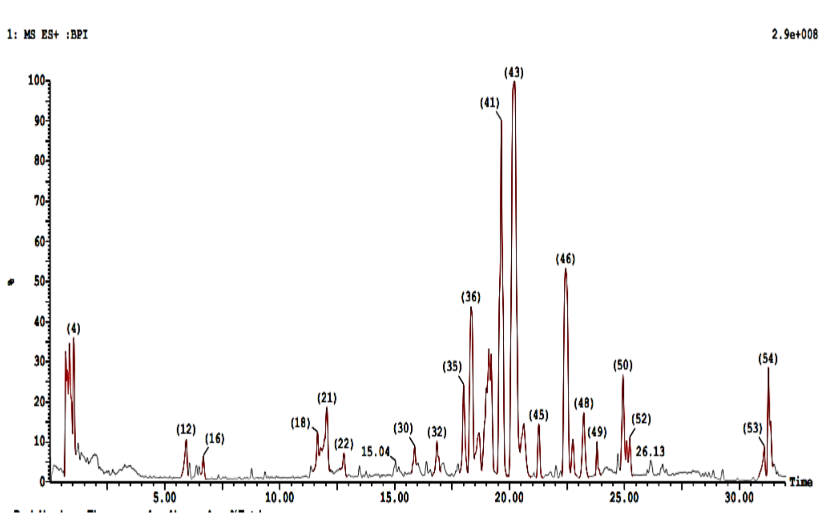

Figure 3: HPLC profile of anthocyanins of Red globe grapes treated with ethylene and abscisic acid at 400 ppm.

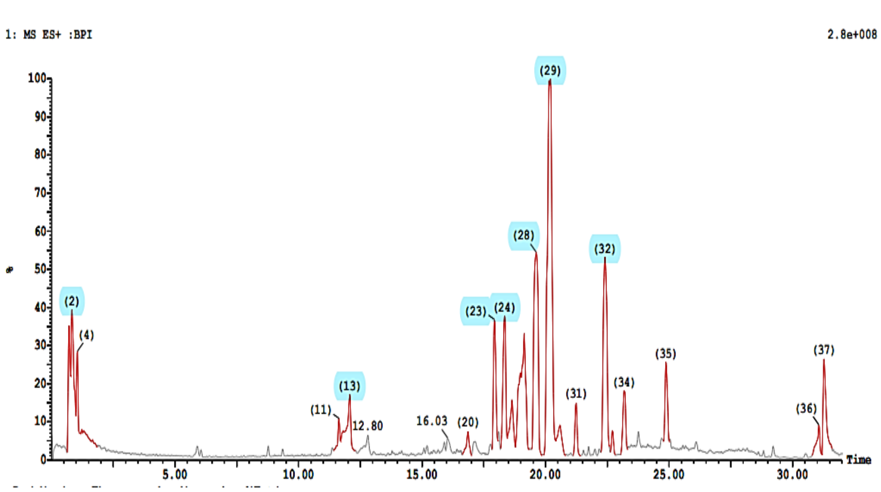

Figure 4: HPLC profile of anthocyanins of Red globe grapes extract

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الملخص العربي

جودة ثمار العنب صنفى الفليم سيدلس والريد جلوب وعلاقتها برش الأيثلين وحامض الأبسيسيك

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يعتبر صنفى الفليم سيدلي والريد جلوب من الأصناف المحببة لدى المستهلكين المصريين, وهذان الصنفان تنجح زراعتهم بصورة جيدة تحت أنواع متعددة من الأراضى والمناخ وهذا يجعلهم من الأصناف الواسعة الأنتشار. وتحت ظروف محافظة المنيا يواجه هذين الصنفين بعض المشاكل المتعلقة بجودة الثمار، مثل التلوين الغير جيد للحبات. من أجل تحسين المواصفات الفيزيائية والكيميائية لهذين الصنفين أجريت التجربة الحقلية الحالية. وقد أشملت التجربةعلى تسعة معاملات بيانها كالتالى: الكنترول (تم رش الكروم بالماء فقط)، رش الأيثلين فى صورة مركب الأيثيفون بأربعة تركيزات هى 100، 200، 300 و 400 جزء فى المليون، وتم رش حامض الأبسيسيك بأربعة تركيزات هى 100، 200، 300 و 400 جزء فى المليون. وقد أوضحت النتائج المتحصل عليها أن التأثير الأيجابى لحامض الأبسيسيك على المواصفات الفيزيائية الحبات (متمثلة فى وزن الوزن وأبعاد الحبة) لكلا الصنفين محل الدراسة كان أكثر وضوحاً عن تأثير الأيثلين فى هذا الشأن، خلال موسمى الدراسة. وعلاوة على ذلك فأن أستجابة الصنف ريد جلوب للمعاملة بحامض الأبسيسيك كانت أكثر وضوحا من أستجابة صنف الفليم سيدلس. وقد أوضحت النتائج المتحصل عليها أن المواصفات الكيميائية للحبات قد تحسنت بصورة معنوية نتيجة رش الأيثلين وحامض الأبسيسيك. وقد أوضحت النتائج أن تأثير الأيثلين كان أكثر وضوحا من تأثير حامض الأبسيسيك خلال موسمى الدراسة لكلا الصنفين. ومما هو جدير بالذكر أن زيادة تركيز كلا المركبين محل الدراسة من 300 جزء فى المليون إلى 400 جزء فى المليون لم يكن له تأثير معنوى على الصفات الكيميائية والفيزيائية لكلا الصنفين. وبناءً علية يمكن التوصية بأهمية الرش بأى من المركبين (حماض الأبسيسك او الايثلين بتركيز 300 جزء فى المليون لتحسين جودة حبات العنب صنفى الفليم سيدلس والريد جلوب.