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Anthocyanin Accumulation and Its Corresponding Gene Expression, Total Phenol, Antioxidant Capacity, and Fruit Quality of 'Crimson Seedless' Grapevine (*Vitis vinifera* L.) in Response to Grafting and Pre-Harvest Applications

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Abstract: The color of grapevine berries is one of the most important quality parameters that define the appearance, attracts the consumer, and determines the price, especially in red grape cultivars. Some grape varieties show less color development due to many factors such as temperature, light intensity, and agricultural management. The present study aimed to evaluate diverse phytohormones coupled with grafting on quality and fruit coloration of 'Crimson Seedless' grapes. Pre-harvest foliar treatments of abscisic acid (ABA) at 400 mg L^{-1}, methyl jasmonate (MeJ) at 1 mM L^{-1}, ethephon (Eth) at 480 mg L^{-1}, and melatonin (Mel) at 100 µmol were applied after 7 days and repeated after 21 days of 'Veraison' (beginning of the coloring phase). The results exposed that the ABA application provided the best anthocyanin accumulation with grafted grapevines whereas Eth displayed the maximum anthocyanin accumulation with ungrafted grapevines. Moreover, the expression of anthocyanin biosynthesis genes (chalcone synthase (CHS), flavanone 3-hydroxylase (F3H), and flavanol 3-O-glucosyl transferase (UFGT)) was consistent with the anthocyanin concentration in berry peel. Moreover, Eth treatment revealed the highest total phenols and antioxidant capacity for both grafted and ungrafted grapevines. MeJ phytohormone exhibited the largest total yield, whereas the treatment of Eth increased the weight of 50 berries and the bunch. Furthermore, ABA induced the uppermost firmness and removal force. Regarding the chemical attributes, the Mel treatment revealed the minimum value of titratable acidity (TA) and the highest polyphenol oxidase (PPO) activity, while the MeJ treatment recorded the superior soluble solids content (SSC), in addition to phenylalanine ammonia-lyase (PAL) and catalase (CAT) enzymes activities. Eth treatment produced the highest activity of the peroxidase (POD) enzyme. Finally, the grafting treatment improved most of the studied fruit attributes.

Keywords: grapevine; antioxidant activity; pre-harvest; gene expression; flavonoid; phytohormones; sustainability

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1. Introduction

Grapevines are the second largest fruit crop worldwide in terms of cultivated area and fourth in terms of production with more than 73 million tons. In Egypt, grapes rank third in terms of the cultivated area as well as production, with one and a half million tons [1]. Recently, great changes have happened globally in consumers' habits regarding

the quality, nutritional value, and impact of food on human health. In this context, table grapes are considered one of the richest sources of phenolic compounds that have high antioxidant and anti-inflammatory impacts, which are helpful in preventing several human diseases [2].

The berry color is one of the most important quality parameters that define the appearance, attracts the consumer, and determines the price, especially in red grape cultivars. It is affected by many factors, such as the growth environment [3], cultivation management [4,5], and plant growth regulators [6–9]. The red color of grapes is mainly determined by the quantity and composition of anthocyanins in the fruit peel [10], which starts at the key point of berry development 'Veraison', due to the accumulation of anthocyanins in the fruit skin of red cultivars in parallel with the increase in sugars and aroma compounds, coupled with the decrease in acid content and firmness [11]. Many enzymes are involved in anthocyanin synthesis, including phenylalanine ammonia-lyase (PAL) as a key enzyme in the general phenylpropanoid pathway [12], and chalcone synthase (CHS), which is considered the first enzyme of the anthocyanin pathway that catalyzes naringenin chalcone production [13]. Then the isomerization of naringenin chalcone to the flavanone naringenin is catalyzed by flavanone 3-hydroxylase (F3H) producing dihydrokaempferol. Meanwhile, flavonols synthase (FLS) catalyzes flavonol synthesis, while dihydroflavonol 4-reductase (DFR) reduces dihydroflavonol to leucoanthocyanidins. The colorless leucoanthocyanidins are subsequently oxidized to colored anthocyanidins by anthocyanidin synthetase (ANS). UDP-glucose-flavonoid 3-O-glucosyltransferase (UFGT) catalyzes the production of chemically stable and water-soluble anthocyanins by adding one glucose molecule in the 3-OH position of the colored anthocyanidins [14,15].

Many factors are involved in the intensity or failure of some grape cultivars to achieve the required level of red color [10]. Among the environmental factors that affect the red color are temperature and light intensity [16]. Generally, in the cooler regions or seasons, more anthocyanin pigments are produced. In addition, high light intensities are also conducive to the synthesis of anthocyanins [17]. The development of berry color could occur in some berries, while a few remain green and unripe, or even an entire bunch may remain in various stages of color break such as in the 'Crimson Seedless' cultivar. This is one of the most important late red grape cultivars in Egypt, and it has a great taste, attractive shape and crunch, and withstands long shipping [18].

Abscisic acid (ABA) is considered one of the plant hormones that was first identified in the early 1960s to regulate many aspects of plant growth and development including embryo maturation, seed dormancy, germination, cell division and elongation, and responses to environmental stresses such as drought and salinity [19]. It plays an important role in physiological processes during the grapevine cycle [20], as well as being the signal that triggers fruit maturation and regulates the ripening and senescence of non-climacteric fruits [21]. ABA treatments at the veraison stage induced skin anthocyanin accumulation [11,22] and phenolic compounds [21,23,24]. Furthermore, ABA and sucrose treatments improved table grape coloration, allowing earlier harvest of 'Crimson Seedless' [25,26].

Jasmonic acid (JA) and its volatile derivative methyl jasmonate (MeJ), known as jasmonate, are hormones that act in the regulation of a wide range of physiological processes in plants [27]. In recent years, several reports presented the effects of JA when applied as pre- or postharvest treatments on fruit quality parameters and ripening at harvest and during storage [28]. Moreover, the application of MeJ to a vineyard led to an increase in the phenolic content, mainly anthocyanins, flavonols, and stilbenes in the grapevine [29].

Ethylene is a natural ripening hormone that is considered a common color enhancer for many fruits. The application of ethephon (Eth) increased the accumulation of anthocyanins by advancing the ripening in grapes [30], apples [31], and plums [32]. During the ripening process of these fruits, coloration is accompanied by ethylene release. Previous studies revealed that ethylene could induce anthocyanin biosynthesis in some climacteric fruits [31], and its components in the signaling pathway are involved in ethylene-induced anthocyanin accumulation [33]. Furthermore, its effect on anthocyanin biosynthesis can vary depending

on the type of fruit [34]. Ethylene may decrease the anthocyanin content in some fruits such as the 'Red Zaosu' pear (climacteric fruit) [35] and strawberries (non-climacteric fruits) [36].

Melatonin (Mel, N-acetyl-5-methoxytryptamine) is an indoleamine that has many functions in plant physiology as a growth regulator [37]. Much credible evidence has shown its effects in preventing oxidative damage in plants by either the direct detoxification of reactive species of oxygen and nitrogen or indirectly by stimulating antioxidant enzymes [38]. Mel showed many effects during the different physiological stages that occur in grapes, and among these effects is the accumulation of anthocyanins in the skin of grapes. Its concentration depends on the grape variety and the physiological stage [39]. Additionally, Mel is indicated to function as a growth regulator [37], with the potential to increase the berry size and improve the homogeneity of grape berries in the bunch [40].

Grafting has many benefits for fruit production, such as alleviating diseases transmitted through the soil, increasing tolerance to abiotic stress, improving nutrient uptake, and enhancing yield and quality [41,42]. Recently, some studies have shown the impact of grafting on fruit quality [43], particularly total soluble solids, titratable acidity, vitamin C, and total anthocyanins. The content of bioactive compounds has been improved by grafting in many fruits such as apples [44], peaches and nectarines [45], sour cherries [46], and 'Red Alexandria' grapes [47]. In grapevines, the rootstock can affect the duration of phenological stages, canopy structure, growth, yield, and fruit quality [48,49]. Information on the biochemical interaction among scions and rootstocks is limited. Under Egyptian conditions and climate change, the problem of fruit discoloration has increased. More studies have to be conducted on the effect of grafting and growth regulators on fruit coloring and anthocyanin accumulation.

The aim of the present study is to demonstrate the effects of grafting and preharvest applications of abscisic acid (ABA), methyl jasmonate (MeJ), ethephon (Eth), and melatonin (Mel) on anthocyanin accumulation and the corresponding gene expression, total phenol, antioxidant capacity, and fruit quality of 'Crimson Seedless' grapevines (*Vitis vinifera* L.).

2. Materials and Methods

2.1. Plant Materials and Evaluated Treatments

The experiment was carried out on the 'Crimson Seedless' cultivar (*Vitis vinifera* L.), grown in a private vineyard located in Al-Khatatbeh district, Beheira Governorate (30°22′31.4″ N, 30°38′55.3″ E), Egypt. A total of 30 vines were divided into two groups (15 vines each). The first group involved fifteen ungrafted vines (cuttings), which were selected and divided into five treatments (three vines each) as follows: (1) Control, (2) abscisic acid (ABA) at a concentration of 400 mg L^{-1}, (3) methyl jasmonate (MeJ) at a concentration of 1 mM L^{-1}, (4) ethephon (Eth) at a concentration of 480 mg L^{-1}, and (5) melatonin (Mel) at a concentration of 100 µmol. The second group contained fifteen grafted vines on 110 Richter (*V. rupestris* × *V. berlandieri*) rootstocks and was divided into five treatments as mentioned previously in the first experiment. Vines were approximately 5 years old, planted at 2 × 3 m spacing in sandy soil under a drip irrigation system. All treatments were sprayed twice in the season, after 7 and 21 days of beginning the coloring phase 'Veraison'. At the harvest stage, samples of grape fruits were collected and directly brought to the Physiology and Breeding of Horticultural Crops Lab, Faculty of Agriculture, Kafrelsheikh University, Kafr El-Sheikh, Egypt for further study.

2.2. Measurements and Analysis

2.2.1. Fruit Yield and Physical Attributes

The total yield (kg per vine, with an average number of 30–35 bunch/vine), average yield/treatment, bunch weight (g), and weight of 50 berries (g) were estimated. Berry size (cm^3, by water displacement) for 50 berries and berry diameter (mm, by Vernier caliper) as the average of 30 berries per treatment were measured. Firmness (Newton (N)/cm^2) was evaluated by a penetrometer pressure tester (Push-full dynamometer) equipped with a probe (1 mm diameter) for 30 berries per treatment at the equatorial area of both sides

for each fruit without removing the peel. The removal force (Newton) was calculated by a penetration pressure gauge of three berries [50].

2.2.2. Berry Color

Berry color was calorimetrically estimated on two opposite sides at the equatorial area of 30 berries per treatment using a Minolta colorimeter (Minolta Co. Ltd., Osaka, Japan) according to the following variables: L*, a*, and b*. The L* (lightness) values correspond to the brightness of the fruit surface and range from 0 (black) to 100 (white color), while a* with positive values represents the red color and negative values indicate the green color and b* represents the yellow color with positive values and blue color with negative values [51].

2.2.3. Soluble Solids Content (SSC) and Titratable Acidity (TA)

A total of 30 berries from each vine were cut and ground to obtain a homogeneous juice sample and then soluble solid content and titratable acidity% were evaluated. SSC was determined with a digital refractometer Atago PR-101 (Atago Co., Ltd., Tokyo, Japan) at 20 °C whereas the total acidity was detected in the same juice by automatic titration (785 DMP Titrino, Metrohm, Filderstadt, Germany) with 0.1 N NaOH up to pH 8.2, and the obtained results were expressed as % of tartaric acid equivalent [52].

2.2.4. Total Phenols and Antioxidative Capacity

Total phenols were estimated by Folin–Ciocalteu reagent and external calibration with gallic acid according to Martínez-Esplá et al. [53], while antioxidative capacity was measured by DPPH assay as described by Binsan et al. [54]. From the berries' skin, 100 mg was collected in a 10 mL solution of water: methanol (2:8) for 24 h and extracted in dark conditions. To estimate the total phenols, 0.5 mL from the previous extract was taken and filled to 5 mL with 2.5 mL of the Folin-Ciocalteu reagent and 2 mL of the sodium carbonate solution, then incubated for 30 min in dark conditions at room temperature. The total phenols were quantified spectrophotometrically (Double-beam UV/Visible Spectrophotometer Libra S80 PC, biochrome Ltd., Cambridge, UK) by absorbance at 765 nm. For the antioxidant capacity detection, 2 mL of the previous extract was taken and filled to 4 mL by DPPH and subsequently incubated for 30 min in the dark and measured spectrophotometrically at 517 nm.

2.2.5. Anthocyanins Accumulation and Content

Total anthocyanins were estimated before harvest (every 6 days from the beginning of the color break) to determine the development of anthocyanins, as well as at harvest, as follows: Total anthocyanins extracted from 3 g of berry skin in volumetric flask filled with 25 mL of acidified methanol (HCl 1.5 mol + ethyl alcohol 99%) and left in a dark environment for 24 h, then estimated spectrophotometrically at 535 nm. The results were expressed in milligrams of total anthocyanins as malvidin-3-glucoside per gram of berry skin [55].

2.2.6. Determination of Enzymes Activities

The enzyme activity of phenylalanine ammonia-lyase (PAL), polyphenol oxidase (PPO), peroxidase (POD), and catalase (CAT) was estimated in the treated samples. Samples (0.2 mg) of berry skin were homogenized in ice-cold 0.1 M sodium phosphate buffer pH 7.0, at 4 °C, centrifuged at 11,200× *g* at 4 °C for 10 min, and finally, the clear supernatant was subjected to an enzyme assay. Phenylalanine ammonia-lyase (PAL) activity was evaluated according to the method used by Assis et al. [56] by recording the absorbance at 290 nm. One PAL unit was defined as the formation of 1 µg of cinnamic acid equivalents per hour, and the specific activity was expressed as unit mg/FW. Polyphenol oxidase (PPO) activity was recorded according to Kar and Mishra [57], with minor modifications, as the 3 mL reaction mix contained 25 mM phosphate buffer (pH 7), 0.1 mM pyrogallol, 0.1 mL of the enzyme extract, and blank without pyrogallol. The absorbance of the purpurogallin formed was registered at 420 nm. Peroxidase (POD) and catalase (CAT) activities were estimated

in a 3 mL reaction mix containing 50 mM potassium phosphate buffer (pH 7.0), 20 mM guaiacol, 40 mM H_2O_2, and 0.1 mL of the enzyme extract as an increase in absorbance for 3 min at 470 nm for peroxidase and 50 mM phosphate buffer (pH 7.0), 5.9 mM H_2O_2, and 0.1 mL of the enzyme extract as a decrease in absorbance for 3 min at 240 nm for catalase [58].

2.2.7. RNA Extraction, cDNA Synthesis, and Quantitative Real-Time PCR

Samples of vines treated with abscisic acid (ABA), methyl jasmonate (MeJ), ethephon (Eth), and melatonin (Mel) against the control (untreated vines) from grafted and ungrafted plants were collected 12 days after the second treatment. The total RNA was extracted from 100 mg of frozen vine skin as described in the manufacturer's instructions of the total RNA purification kit (K0731, Thermo Scientific, Vilnius, Lithuania). RNA samples were treated with the ezDNase enzyme (11766051, ezDNase™ Enzyme, Invitrogen, Thermo Scientific, Vilnius, Lithuania) to eliminate the contaminated DNA according to the manufacturer's instructions. The RNA concentration and purity were assessed by the nanodrop spectrophotometer (NanoDrop® ND-1000, Thermo Scientific).

First-strand cDNA was obtained from the RNA by the reverse transcription process using the revert Aid H minus First Strand cDNA Synthesis Kit (K1632, Thermo Scientific) according to the manufacturer's protocol. The quantitative real-time PCR assay (qRT-PCR) was performed for three genes including flavanone3-hydroxylase (F3H), chalcone synthase (CHS), and flavonoid3-o-glucosyltransferase (UFGT), whereas β-actin housekeeping gene expression was used to normalize the results (Table 1) [59]. To measure the expression of target genes, the 2X Maxima SYBR Green/ROX qPCR Master Mix (K0221, Thermo scientific) was selected to evaluate the expression as mentioned by the manufacturer's guidelines. Next, 50 ng of cDNA was used as a template in 20 µL total reactions, and three replicates were employed to quantify the gene expression as a fold change through the comparative threshold cycle (Ct) method by calculating the $2^{-\Delta\Delta CT}$ [60].

Table 1. Forward and reverse primers sequence for F3H, CHS, and UFGT genes.

Gene Name	Abbreviation	Primer Sequence
Flavanone3-hydroxylase	F3H	F/5 CAGTGCAAGACTGGCGCGAGATCGTA/3 R/5 TAGCCTCAGACAACACCTCCAGCAACT/3
Chalcone synthase	CHS	F/5 CACTCTTCGAACTCGTCTCT/3 R/5 CCACCAAGCTCTTCTCTATG/3
flavonoid3-o-glucosyltransferase	UFGT	F/5 TGCAGGGCCTAACTCACTCT/3 R/5 GCAGTCGCCTTAGGTAGCAC/3
Beta-Actin	β-actin	F/5 GTGCCTGCCATGTATGTTGCC R/5GCAAGGTCAAGACGAAGGATA

2.3. Statistical Analysis

All the data were expressed as means ± S.E. Statistical significance was evaluated by one-way analysis of variance (ANOVA) using costate whereas the individual comparisons were obtained by the L.S.D multiple range test (DMRT). Values were considered statistically significant when $p < 0.05$ [61].

3. Results

3.1. Yield (kg/Vine)

The performance of vines (grafted and ungrafted) under the implementation of four phytohormones (Abscisic acid (ABA), Methyl jasmonate (MeJ), Ethephon (Eth), and Melatonin (Mel)) and their interaction was evaluated. The effects of grafting, treatments, and their interaction on the grape yield are presented in Figure 1. Significant and positive differences were noted, as the grafted grapevines had a higher effect on the yield compared to the ungrafted vines. In addition, the treatment of MeJ showed the highest effect on

yield, followed by Eth, ABA, and Mel, respectively, when compared to the control. The interaction between grafting and treatments exhibited a significant and positive impact on yield since the grafted grapevines treated with MeJ gave the best yield and reached 17 kg/vine, followed by grafted grapevines treated with Eth and ABA.

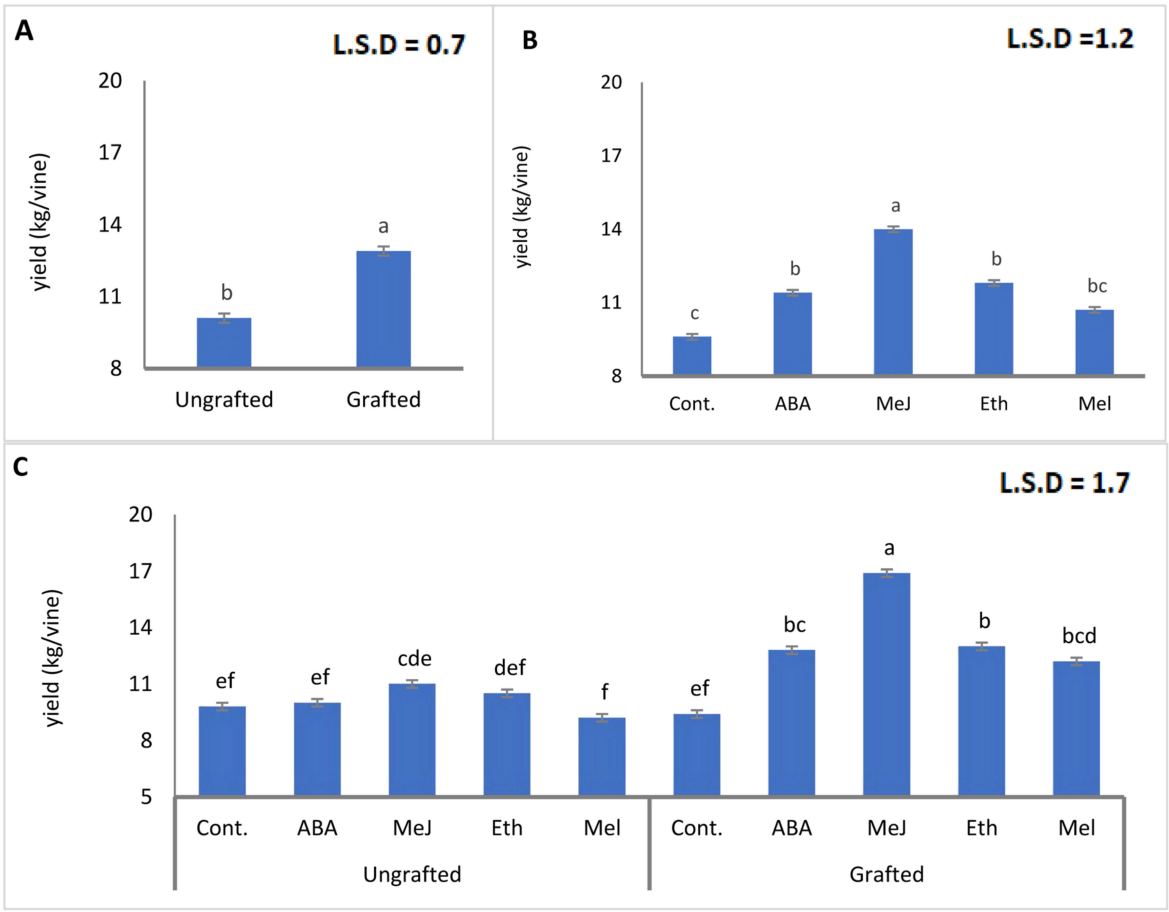

Figure 1. Effect of grafting (**A**), treatments (**B**), and their interaction (**C**) on the yield of 'Crimson Seedless' grapes. The data represent the average of three replicates. Cont: Control, ABA: Abscisic acid, MeJ: Methyl jasmonate, Eth: Ethephon, and Mel: Melatonin. a–f, different letters indicate significant differences among treatments at $p < 0.05$ according to Duncan's multiple rang test.

3.2. Bunch Weight, Weight, and Size of 50 Berries

The average bunch weight, weight, and size of 50 berries were assessed under grafting and treatments as shown in Table 2. The grafted grapevines produced higher bunch weight, while ungrafted plants displayed a considerable increase in berry weight and size of 50 berries. Furthermore, the Eth and MeJ treatments gave the highest average bunch weight and weight of 50 berries without significant differences, whereas the ABA treatment exhibited the best size of 50 berries. The interaction between grafting and treatment application manifested wide and significant variations for the evaluated traits. The grafted grapevines with treated MeJ and Eth caused a significant and positive increment in the average bunch weight, which reached 46% and 39%, respectively, when compared to the grafted control, while the lowest values were assigned to ungrafted grapevines treated with MeJ. Moreover, the heaviest weight of 50 berries was recorded for ungrafted grapevines treated with Eth (17.4% increment) compared with the ungrafted control, whereas the lowest values were assigned for ungrafted grapevines with Mel. Concerning the size of 50 berries, ungrafted grapevines treated with ABA and Eth in addition to grafted grapevines treated with Me1 showed the highest values without any significant differences. Likewise, the lowest 50-berry size was seen in the grafted control.

Table 2. Preharvest application of growth regulators on average bunch weight, weight, and size of 50 berries at harvest.

Treatment		Average Bunch Weight (g)	Weight 50 Berries (g)	Size 50 Berries (cm^3)
Ungrafted		325.2 ± 4.6 b	193.9 ± 2.6 a	162 ± 1.6 a
Grafted		398.1 ± 5.2 a	185.9 ± 3.3 b	160 ± 2.3 a
Cont.		343.3 ± 5.1 b	176.8 ± 3.1 b	150.8 ± 3 c
ABA		336.8 ± 16.9 b	191.8 ± 1.7 a	174.1 ± 4.4 a
MeJ		388 ± 28.9 a	193.3 ± 3.7 a	155 ± 0 bc
Eth		400 ± 6.7 a	199.3 ± 6.2 a	170.8 ± 8.2 ab
Mel		340.2 ± 7.9 b	188.3 ± 1.8 ab	157.5 ± 7.2 bc
Ungrafted	Cont.	350 ± 10.4 b	189.6 ± 2.8 cde	166.6 ± 4.4 ab
	ABA	339.1 ± 8.7 b	202.3 ± 5.2 bc	178.3 ± 8.8 a
	MeJ	284.7 ± 23.3 c	192.3 ± 7.8 cd	150 ± 7.6 bcd
	Eth	331.7 ± 21.3 b	222.6 ± 6.3 a	176.6 ± 7.2 a
	Mel	320.5 ± 14.9 bc	162.6 ± 3.7 f	141.6 ± 1.4 cd
Grafted	Cont.	336.6 ± 1.6 b	164 ± 4 f	135 ± 2.8 d
	ABA	334.4 ± 25.8 b	181.3 ± 5.3 de	170 ± 0 ab
	MeJ	491.4 ± 46.9 a	194.3 ± 0.3 cd	160 ± 7.6 abc
	Eth	468.3 ± 12.5 a	176 ± 9.1 ef	165 ± 10 ab
	Mel	360 ± 16.4 b	214 ± 2.6 ab	173.3 ± 13 a
$L.S.D_{0.05\%}$ Grafting		20.3	7.2	10.3
$L.S.D_{0.05\%}$ Treatment		32.1	11.5	16.3
$L.S.D_{0.05\%}$ Interaction		45.4	16.3	23

Values in the same column (for main effect and for their interaction) that are followed by the same letters do not differ significantly according to Duncan's multiple range test at the 5% level. Cont.: Control, ABA: Abscisic acid, MeJ: Methyl jasmonate, Eth: Ethephon, Mel: Melatonin.

3.3. Berry Firmness, Removal Force, and Berry Diameter

The berry firmness, removal force, and berry diameter displayed no significant effect of grafting. The application of ABA, Eth, MeJ, and Mel had the highest and most significant values for berry firmness, respectively, compared with untreated plants. In the same manner, applying the selected phytohormones significantly affected the removal force, and the Mel treatment exhibited the best value, followed by the MeJ treatment. The interaction between grafting and phytohormone treatments led to a significant effect on berry firmness, diameter, and removal force. Ungrafted grapevines treated with ABA significantly increased the berry firmness compared to the control of ungrafted grapevines. Moreover, the interaction between ungrafted and Mel produced the highest value of removal force, while the largest berry diameter was recorded in grafted grapevines with ABA and ungrafted grapevines with MeJ without significant differences (Table 3).

3.4. Titratable Acidity and Soluble Solids Content

Titratable acidity (TA%) was significantly affected by grafting, where the lowest acidity was detected in grafted grapevines. In addition, the Mel treatment, followed by Eth, recorded lower acidity compared to the control. Moreover, the interaction between grafted grapevines and Mel treatment presented the lowest acidity, whereas the highest acidity value was found in ungrafted control grapevines (Figure 2). The soluble solids content (SSC%) was not significantly affected by grafting, but a positive and significant influence was observed regarding phytohormone treatment and its interaction with grafting. The uppermost SSC values were measured under MeJ followed by Eth treatment. The interaction between grafting and applied phytohormones exhibited that Mel treatment under grafted conditions followed by MeJ under both conditions (grafted and ungrafted grapevines) induced the highest content of SSC (Figure 3).

3.5. Total Phenols and Antioxidant Capacity

Both grafting and phytohormone treatments positively enhanced the total phenols in grapevines. Regarding treatment with phytohormones, Eth treatment improved the total phenols with a percentage of 52.4%. When going forward to detect the impact of the interaction, the treated grapevines with Eth in grafted and ungrafted plants showed the best total phenol values when compared to untreated grapevines (control) (Figure 4). In the same manner, the antioxidant capacity was significantly affected by grafting and treatments. The grafted grapevines produced superior values of antioxidant capacity (31%) when compared to ungrafted plants. Similarly, the Eth treatment produced the maximum significant and positive antioxidants (37.5%), followed by Mel treatment (25%) when compared with untreated plants. Furthermore, the interaction between the grafted grapevines and Eth treatment exhibited the highest positive and significant antioxidant capacity, followed by the grafted grapevines treated with MeJ (Figure 5).

3.6. Color

The data presented in Table 4 demonstrate that grafted grapevines expressed the largest significant and positive values of L* and a* in comparison to the ungrafted plants. On the opposite side, the highest b* values were noticed in the ungrafted grapevines. Among the treatments, high positive and significant L* and b* values were revealed by untreated grapevines (control), followed by Mel treatment. Meanwhile, Eth treatment displayed the most significant values of a*, followed by MeJ treatment. Exploring the interaction between grafting and treatments led to significant differences in the values of L*, a*, and b*, where the highest values of L* and b* were assigned for ungrafted control treatment. Eventually, a* caused a positive and significant increment in grafted grapevines treated with ABA, followed by ungrafted grapevines treated with Eth without any significant differences.

Table 3. Preharvest application of growth regulators on berry firmness, removal force, and berry diameter at harvest.

Treatment		Berries Firmness (N/cm^2)	Removal Force (N)	Berries Diameter (mm)
Ungrafted		667.3 ± 8.8 a	694.2 ± 15.6 a	16.5 ± 0.1 a
Grafted		652.8 ± 31.4 a	697.1 ± 38.4 a	16.4 ± 0.1 a
Cont.		605.5 ± 31.3 b	645.5 ± 27.4 b	16.4 ± 0.08 a
ABA		694.4 ± 14.4 a	676.6 ± 23.1 b	16.8 ± 0.3 a
MeJ		668.8 ± 15.1 a	697.7 ± 23.1 ab	16.3 ± 0.5 a
Eth		672.2 ± 38.8 a	675 ± 30.7 b	16.6 ± 0.2 a
Mel		659.4 ± 26.6 a	738.3 ± 51.1 a	16.2 ± 0.2 a
Ungrafted	Cont.	567.7 ± 24.3 c	628.8 ± 24.5 b	16.7 ± 0.3 ab
	ABA	728.8 ± 7.7 a	667.7 ± 20.4 b	16.7 ± 0.3 ab
	MeJ	681.1 ± 43.7 ab	686.6 ± 33.3 b	16.9 ± 0.4 a
	Eth	690 ± 30.2 ab	691.1 ± 56.1 b	16.7 ± 0.4 ab
	Mel	668.8 ± 65.4 ab	796.6 ± 29.1 a	15.9 ± 0.1 b
Grafted	Cont.	643.3 ± 48.5 b	662.2 ± 77.1 b	16.3 ± 0.2 ab
	ABA	660 ± 27.2 ab	685.5 ± 25.9 b	17.0 ± 0.6 a
	MeJ	656.6 ± 24.1 b	708.8 ± 43.3 b	15.9 ± 0.6 b
	Eth	654.4 ± 47.5 b	658.8 ± 28 b	16.5 ± 0.3 ab
	Mel	650 ± 27.1 b	680 ± 81.9 b	16.5 ± 0.3 ab
$L.S.D_{0.05\%}$ Grafting		32.2	37.6	0.3
$L.S.D_{0.05\%}$ Treatment		50.9	59.4	0.5
$L.S.D_{0.05\%}$ Interaction		72.1	84.1	0.8

Values in the same column (for main effect and for their interaction) that are followed by the same letters do not differ significantly according to Duncan's multiple range test at the 5% level. Cont: Control, ABA: Abscisic acid, MeJ: Methyl jasmonate, Eth: Ethephon, Mel: Melatonin.

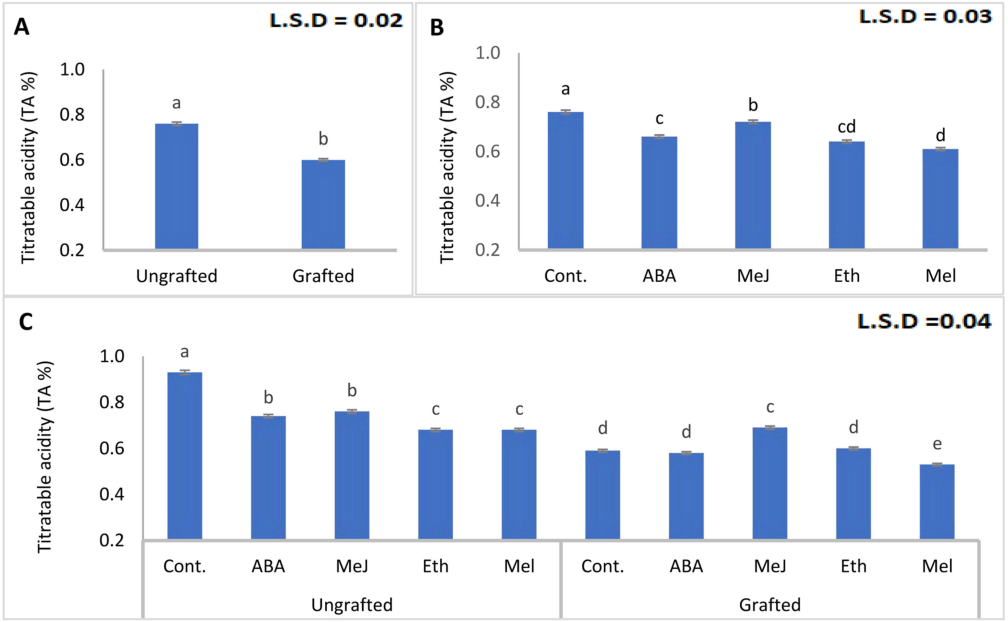

Figure 2. Effect of grafting (**A**), treatments (**B**), and their interaction (**C**) on titratable acidity (TA%) of 'Crimson Seedless' grape. The data represent the average of triplicates. Cont: Control, ABA: Abscisic acid, MeJ: Methyl jasmonate, Eth: Ethephon, Mel: Melatonin. a–e, different letters indicate significant differences among treatments at $p < 0.05$ according to Duncan's multiple rang test.

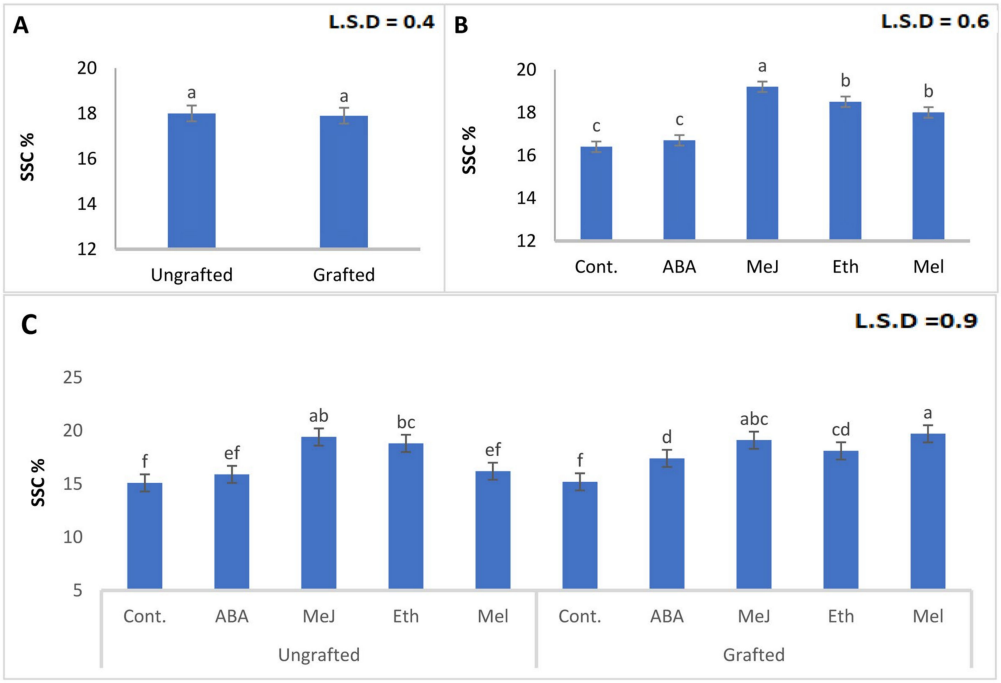

Figure 3. Effect of grafting (**A**), treatments (**B**), and their interaction (**C**) on soluble solids content (SSC%) of 'Crimson Seedless' grape. The data represent the average of triplicates. Cont: Control, ABA: Abscisic acid, MeJ: Methyl jasmonate, Eth: Ethephon, Mel: Melatonin. a–f, different letters indicate significant differences among treatments at $p < 0.05$ according to Duncan's multiple rang test.

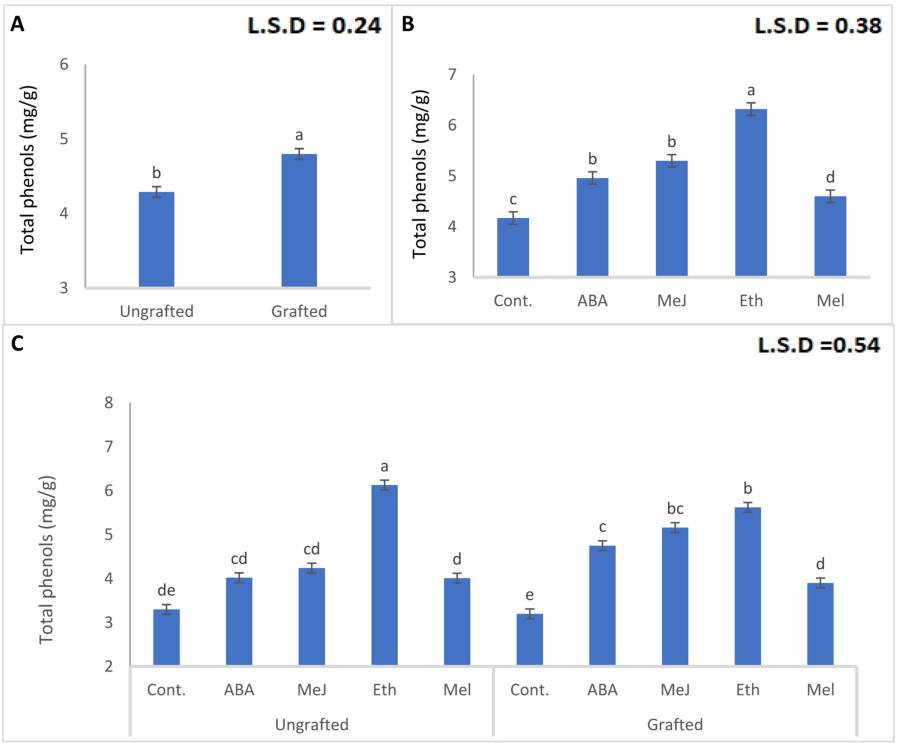

Figure 4. Effect of grafting (**A**), treatments (**B**), and their interaction (**C**) on total phenols of 'Crimson Seedless' grape. The data represent the average of triplicates. Cont: Control, ABA: Abscisic acid, MeJ: Methyl jasmonate, Eth: Ethephon, Mel: Melatonin. a–e, different letters indicate significant differences among treatments at $p < 0.05$ according to Duncan's multiple rang test.

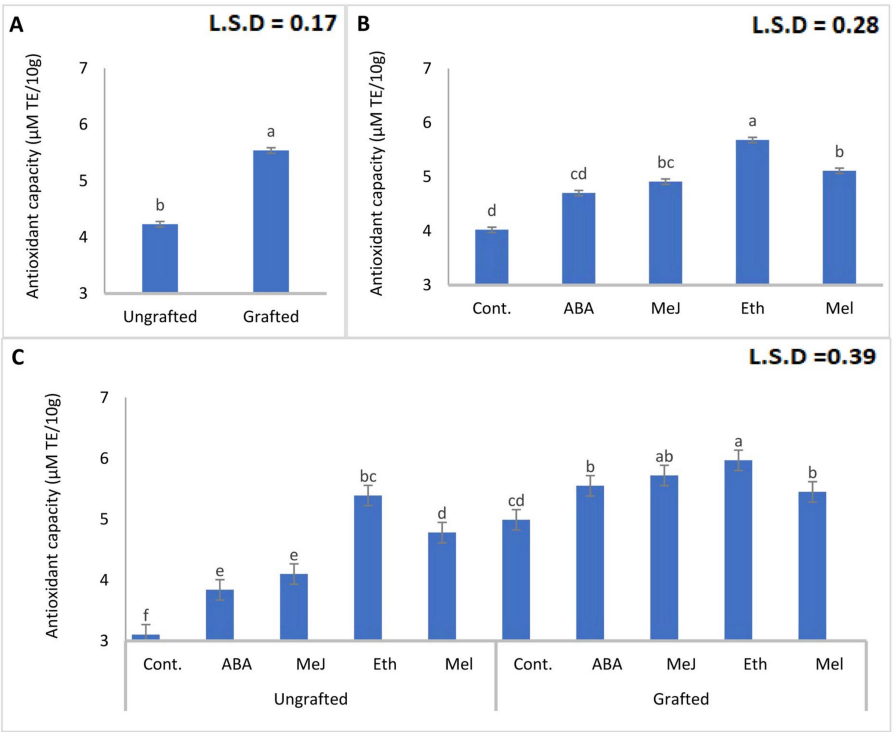

Figure 5. Effect of grafting (**A**), treatments (**B**), and their interaction (**C**) on antioxidant capacity of 'Crimson Seedless' grape. The data represent the average of triplicates. Cont: Control, ABA: Abscisic acid, MeJ: Methyl jasmonate, Eth: Ethephon, Mel: Melatonin. a–f, different letters indicate significant differences among treatments at $p < 0.05$ according to Duncan's multiple rang test.

Table 4. Preharvest application of growth regulators on berry color values.

Treatment		Color		
		L*	a*	b*
Ungrafted		27.4 ± 0.5 b	9.2 ± 0.4 b	23.0 ± 0.2 a
Grafted		30.3 ± 0.6 a	13.1 ± 0.8 a	21.1 ± 0.6 b
Cont.		40 ± 2.2 a	3.5 ± 0.9 d	30.4 ± 0.1 a
ABA		20.2 ± 0.2 d	12.5 ± 0.7 b	16.7 ± 0.6 d
MeJ		23.9 ± 0.7 cd	14.8 ± 0.7 ab	21.7 ± 0.9 c
Eth		27.1 ± 0.7 c	16 ± 0.6 a	15.8 ± 1.2 d
Mel		32.9 ± 1.9 b	8.9 ± 0.7 c	25.6 ± 1 b
Ungrafted	Cont.	42.3 ± 0.4 a	3.0 ± 0.6 c	34.3 ± 2.1 a
	ABA	25.6 ± 1.3 cd	8.0 ± 1.5 b	20.8 ± 1.2 cd
	MeJ	21.3 ± 1.6 de	13.6 ± 1.1 a	18.8 ± 0.1 de
	Eth	18.6 ± 0.6 ef	16.7 ± 1.5 a	16.0 ± 1.7 ef
	Mel	29.2 ± 2.1 c	4.8 ± 1.1 bc	25.2 ± 0.8 b
Grafted	Cont.	37.7 ± 2.8 ab	4.1 ± 0.7 bc	26.5 ± 0.6 b
	ABA	14.8 ± 0.8 f	17 ± 2.1 a	12.6 ± 1.3 f
	MeJ	26.6 ± 0.9 cd	16.1 ± 2.2 a	24.6 ± 2.4 bc
	Eth	35.6 ± 3.1 b	15.3 ± 1.1 a	15.6 ± 1.2 ef
	Mel	36.7 ± 0.8 b	13.1 ± 1.1 a	26.0 ± 1.3 b
$L.S.D_{0.05\%}$ Grafting		2.4	1.9	1.9
$L.S.D_{0.05\%}$ Treatment		3.8	3	3
$L.S.D_{0.05\%}$ Interaction		5.3	4.3	4.3

Values in the same column (for main effect and their interaction) that are followed by the same letters do not differ significantly according to Duncan's multiple range test at the 5% level. L*: (luminosity) & b*: (blue–yellow) & a*: (green–red). Cont.: Control, ABA: Abscisic acid, MeJ: Methyl jasmonate, Eth: Ethephon, Mel: Melatonin.

3.7. Total Anthocyanin Accumulation and Gene Expression

The grapevines treated with growth regulators manifested varied anthocyanin accumulation under grafted and ungrafted conditions. Grafted and ungrafted grapevines disclosed no significant difference in anthocyanin accumulation, whereas the phytohormones enhanced the anthocyanins significantly in comparison with untreated plants (Figure 6A,B). The anthocyanin content before the application of the growth regulators (first date) was fairly equal, but six days after the first application (second date), the grapevines began to show an increase in anthocyanin content. In grafted grapevines, all treatments appeared to show a significant and positive increase in the total anthocyanin content throughout the ripening period of the berries until harvest in comparison to the control treatment. ABA was considered the most superior treatment followed by MeJ, Mel, and Eth (Figure 6C). This remarkable superiority continued during the ripening period until harvest time (fifth date). In ungrafted grapevines, the anthocyanin accumulation was also significantly affected by the growth regulators applied, providing a positive and significant increase in the total anthocyanin content. However, Eth and MeJ treatments had the highest effect on anthocyanin accumulation during the ripening period of berries until harvest (fifth date) compared to the control group (Figure 6D). Moreover, regardless of the different treatments and grafting, the importance of the second application (i.e., 21 days after the first one) to maintain the accumulation of anthocyanins over time was clear.

Additionally, the expression of CHS, F3H, and UFGT biosynthesis genes was estimated by detecting the changes in mRNA levels using qRT-PCR. Applying grafting enhanced the gene expression of flavanone 3-hydroxylase (F3H) whereas ungrafted plants manifested better expression of chalcone synthase (CHS). The sprayed Eth and MeJ significantly and positively enhanced the CHS gene expression up to 14- and 13-fold, respectively, whereas the F3H upregulated by approximately 7-fold under the application of ABA and MeJ. Eventually, ABA, MeJ, and Eth overexpressed the UFGT mRNA level without any significant differences between the three treatments (Figure 7d–f). Under grafted conditions, the expression of CHS, F3H, and UFGT increased up to 12-, 11-, and 22-fold, respectively, under

ABA treatment (Figure 7g–i). With the same pattern, a significant increase in the gene expression level of target genes was also detected under other phytohormone treatments (MeJ, Eth, and Mel, respectively) when compared to untreated grapevines. Similarly, the ungrafted plants treated with Eth exhibited the uppermost expression and showed 22-, 8- and 20-fold changes in the relative mRNA expression of CHS, F3H, and UFGT, respectively (Figure 7j–l). MeJ phytohormone also induced the relative mRNA expression of target genes, followed by ABA and Mel.

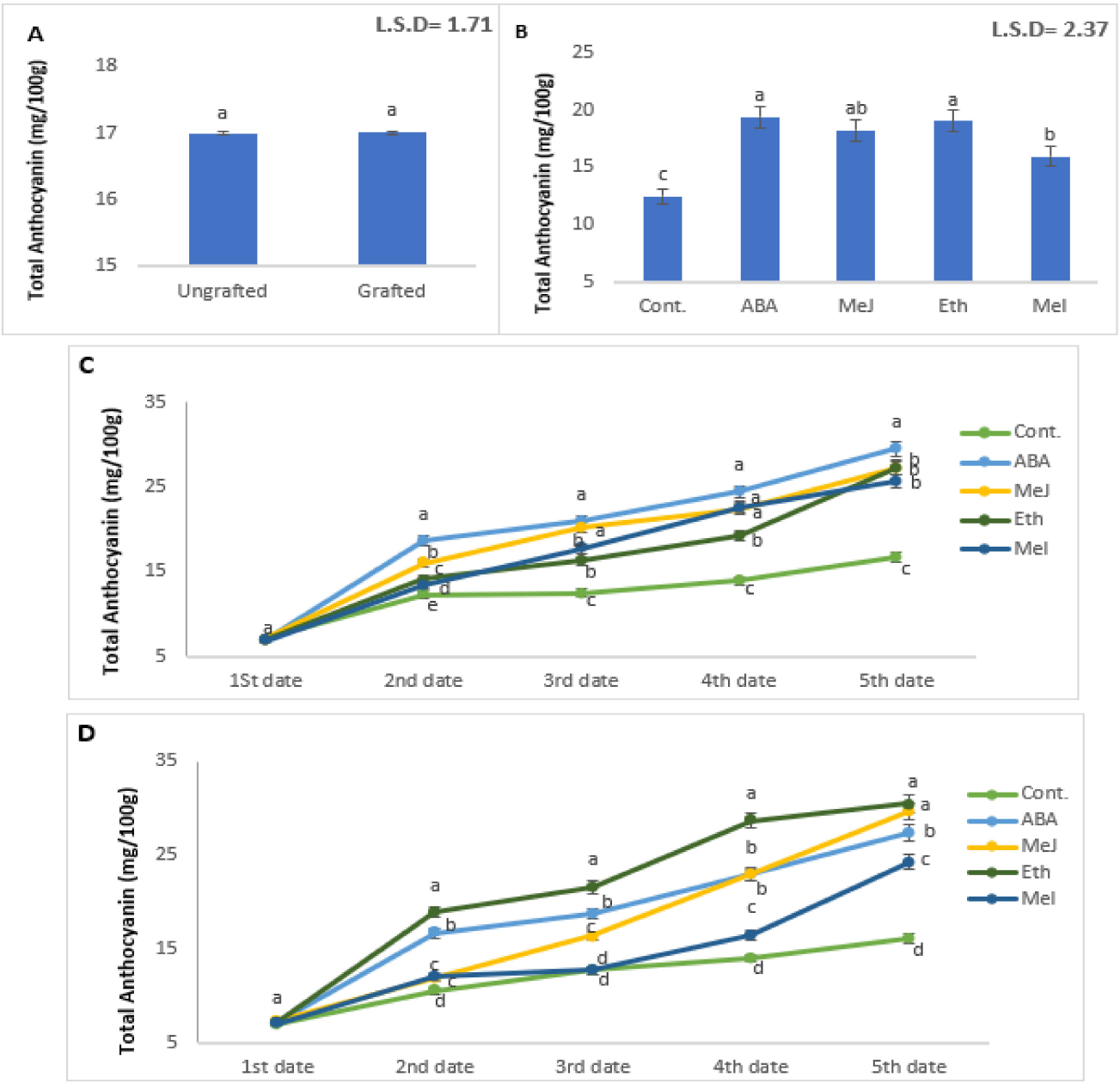

Figure 6. Effect of grafting (**A**), treatments (**B**), treatments with grafting (**C**), and treatments combined with ungrafted grapevines (**D**) on total anthocyanin development. 1st date: 6 days before spraying, 2nd date: 6 days after the first application, 3rd date: 12 days after the first application, 4th date: 6 days after the second application, 5th date: 12 days after the second application (At harvest). Cont.: Control, ABA: Abscisic acid, MeJ: Methyl jasmonate, Eth: Ethephon, Mel: Melatonin. a–d, different letters indicate significant differences among treatments at $p < 0.05$ according to Duncan's multiple rang test.

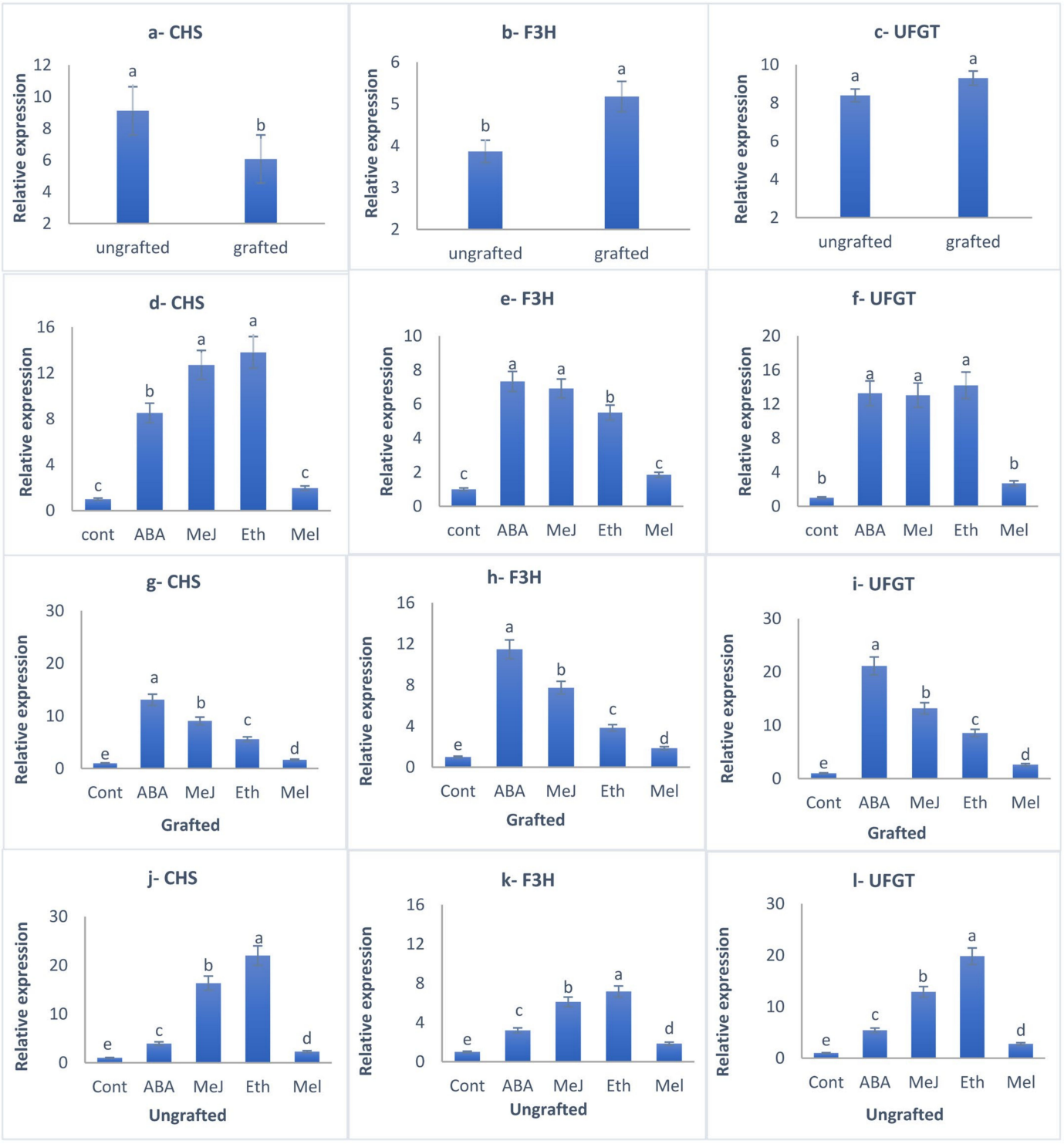

Figure 7. Graphical presentation of quantitative real-time PCR analysis for the expression of chalcone synthase (CHS), flavanone 3-hydroxylase (F3H), and flavanol 3-O-glucosyl transferase (UFGT) under effect of grafting (**a**–**c**), treatments (**d**–**f**), and their interaction (**g**–**l**). Cont (Control), Abscisic acid (ABA), Methyl jasmonate (MeJ), Ethephon (Eth), and Melatonin (Mel). a-d, different letters indicate significant differences among treatments at $p < 0.05$ according to Duncan's multiple rang test.

3.8. Enzymes (PAL, PPO, POD, and CAT)

Enzyme activities of phenylalanine ammonia-lyase (PAL), polyphenol oxidase (PPO), peroxidase (POD), and catalase (CAT) were evaluated under grafted and ungrafted conditions, phytohormone implementation, and their interaction. Grafted grapevines estimated

the best activity of PAL, POD, and CAT enzymes, while the ungrafted grapevines manifested significant and positive activity of the PPO enzyme. Among the treatments, the MeJ treatment displayed the highest positive activity of the PAL enzyme, followed by the Eth treatment without significant differences (Figure 8B). Mel treatment recorded the highest activity of the PPO enzyme, followed by the control treatment (Figure 9B). Eth and MeJ treatments also induced the highest activity of the POD enzyme (Figure 10B). In the case of the CAT enzyme, MeJ and Eth treatments demonstrated the highest activity as presented in Figure 11B. Enzyme activity was significantly affected by the interaction in relation to PAL enzyme activity. Grafted grapevines treated with MeJ had the highest value related to enzyme activity, whereas the enzyme activity declined under control treatments in grafted and ungrafted vines (Figure 8C). Mel treatments in grafted and ungrafted vines followed by ABA treatment in ungrafted plants expressed PPO enzyme activity with no significant differences, whereas the smallest effect was observed in the control treatment of grafted grapevines (Figure 9C). The largest POD enzyme activity was obtained under grafted grapevines treated with Eth when compared with the control treatment of ungrafted grapevines (the lowest value, Figure 10C). Regarding CAT enzyme activity, grapevines grafted and treated with MeJ showed the maximum activity, followed by grapevines grafted and treated with Eth (Figure 11C).

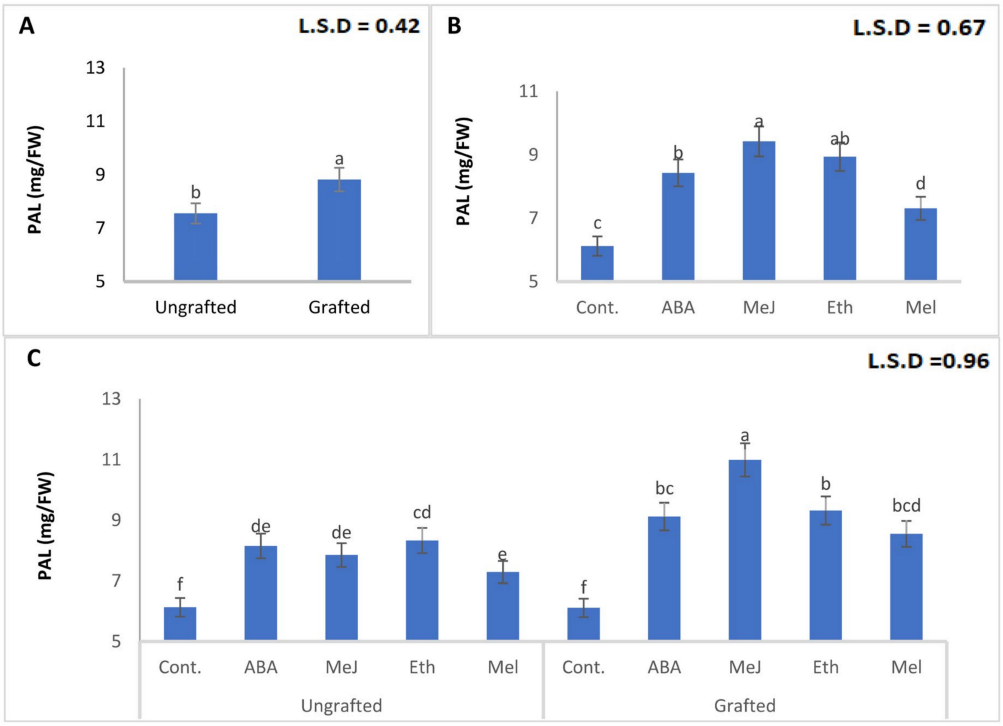

Figure 8. Effect of grafting (**A**), treatments (**B**), and their interaction (**C**) on Phenylalanine ammonia-lyase (PAL) activity of 'Crimson Seedless' grape. The data represent the average of triplicates. Cont.: Control, ABA: Abscisic acid, MeJ: Methyl jasmonate, Eth: Ethephon, Mel: Melatonin. a–f, different letters indicate significant differences among treatments at $p < 0.05$ according to Duncan's multiple rang test.

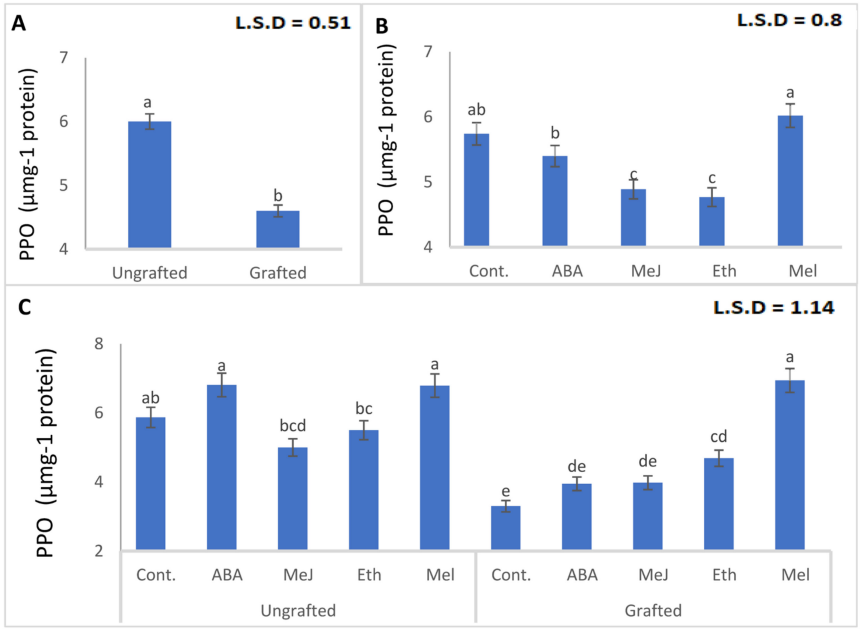

Figure 9. Effect of grafting (**A**), treatments (**B**), and their interaction (**C**) on Polyphenol oxidase (PPO) activity of 'Crimson Seedless' grape. The data represent the average of three replicates. Cont: Control, ABA: Abscisic acid, MeJ: Methyl jasmonate, Eth: Ethephon, Mel: Melatonin. a–e, different letters indicate significant differences among treatments at $p < 0.05$ according to Duncan's multiple rang test.

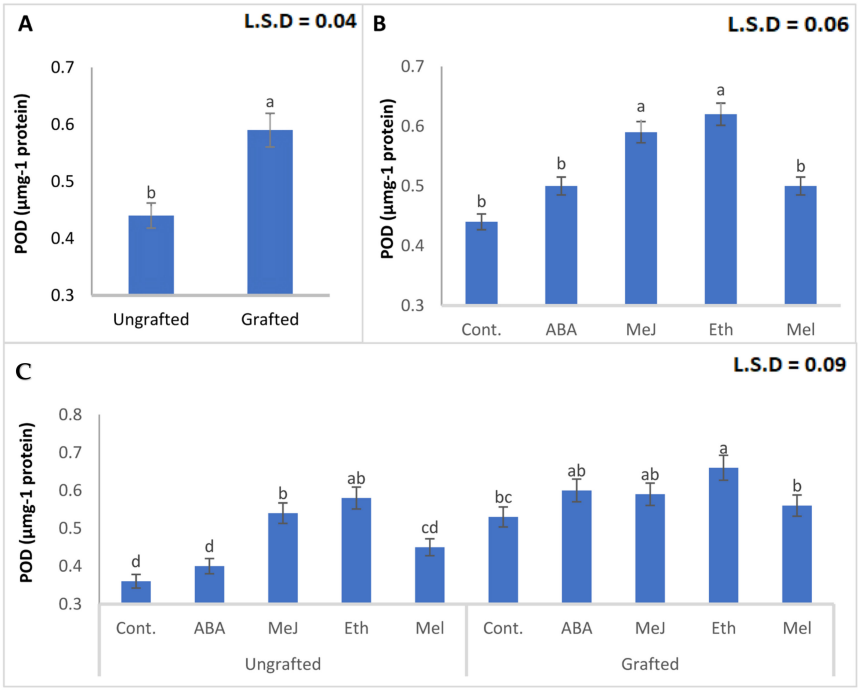

Figure 10. Effect of grafting (**A**), treatments (**B**), and their interaction (**C**) on Peroxidase (POD) activity of 'Crimson Seedless' grape. The data represent the average of triplicates. Cont: Control, ABA: Abscisic acid, MeJ: Methyl jasmonate, Eth: Ethephon, Mel: Melatonin. a–d, different letters indicate significant differences among treatments at $p < 0.05$ according to Duncan's multiple rang test.

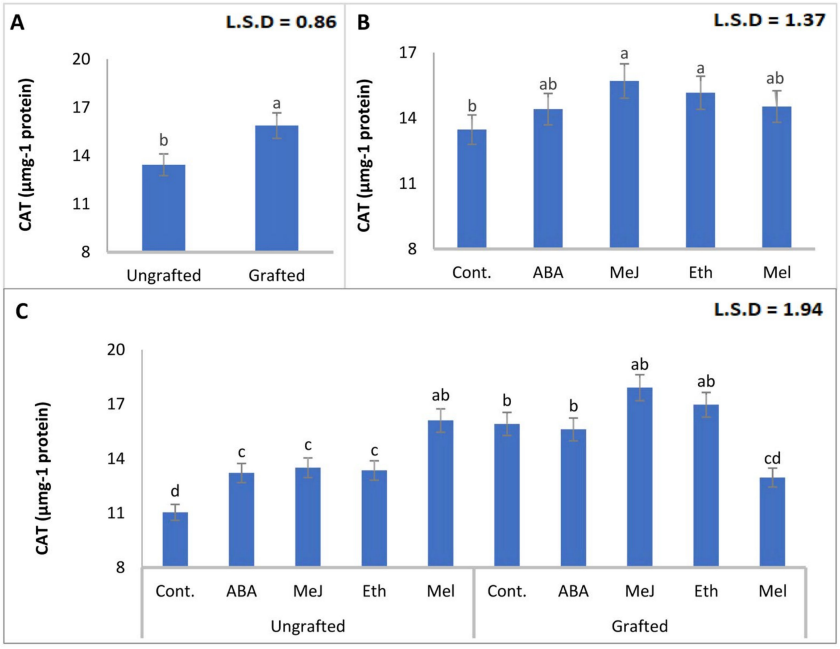

Figure 11. Effect of grafting (**A**), treatments (**B**), and their interaction (**C**) on Catalase (CAT) activity of 'Crimson Seedless' grape. The data represent the average of triplicates. Cont: Control, ABA: Abscisic acid, MeJ: Methyl jasmonate, Eth: Ethephon, Mel: Melatonin. a–d, different letters indicate significant differences among treatments at $p < 0.05$ according to Duncan's multiple rang test.

4. Discussion

Fruit color is one of the most important parameters that affect fruit quality. Some grapevine cultivars such as 'Crimson Seedless' fail to reach the proper fruit color due to abiotic stress, cultivation management, and growth regulators. Grafting is widely used (used in approximately 80% of all commercial vines) in viticulture for improving biotic and abiotic stress tolerance (i.e., controlling soil-borne diseases), optimizing fruit quality, and regulating vine vigor [62]. Moreover, rootstocks modulate the elemental composition and affect the fruit quality such as SSC, TA, anthocyanin content, and metabolomic profiles [63]. Rootstock genotypes (SO4, 5BB, and 101-14MG) enhanced the accumulation of phenolic compounds during development in grape berry skin.

In the present study, the fruit weight, size, and yield are highly affected by phytohormone treatment compared to the control. The application of Eth yielded the highest weight of 50 berries followed by MeJ, as well as for the bunch weight, while the ABA application produced the largest size of 50 berries followed by Eth without any significant effects. Moreover, the application of MeJ induced the highest fruit yield followed by Eth and ABA treatments. García-Pastor et al. [64] reported that the application of MeJ was correlated with water accumulation in grape berries that increased fruit weight. Furthermore, Mohamed et al. [65] mentioned that the increase in berry size after the application of ABA is due to the ability of ABA to close the stomata on the berry skin, which subsequently decreases the transpiration rate and increases the water accumulation in berries, which is reflected in fruit size and weight. It is also clear that grafting plays a role in weight gain, and these results are similar to those observed by Jia et al. [66].

Applying the plant phytohormones revealed a significant and positive enhancement in berry firmness especially under ABA spraying. In contrast, Shahab et al. [67] found that the firmness was decreased by the application of ABA on the 'Benitaka' grapevine. Moreover, Cantín et al. [68] noticed that the firmness of 'Crimson Seedless' was not affected by lower concentrations of ABA (75–300 mL), while higher concentrations (500–1000 mL) caused the softening of grape berries. Thus, the effect of ABA on fruit firmness could depend on the concentration and the cultivar [19,57]. Moreover, the berry firmness decreased in grafted vines without any significant differences, and this is in harmony with results obtained

by Ulas et al. [69] on tomatoes. Moreover, rootstocks influence the water relationship and nutritional status of scions, which affect the fruit cells' turgor and the chemical and mechanical properties of fruits [70].

SSC and TA are critical parameters related to fruit quality. Sugars play an essential role in fruit edibility, which significantly determines the consumer's satisfaction. According to the present results, the application of MeJ induced the highest soluble solid content in "Crimson Seedless". García-Pastor et al. [64] mentioned that MeJ treatment enhanced the net photosynthetic rate of the vine, which could improve fruit SSC and sugar content. On the other hand, the results also showed that the application of Mel leads to a decrease in titratable acidity. Similar results have been observed on sweet cherry fruits [71] and apples [72]. The authors suggested that Mel could activate fruit respiration and increase the degradation of soluble acid. The degradation of organic acids in fruits is due to the utilization of organic acids as a substrate in respiration. Consequently, the effectiveness of Mel depends on its concentration, the time of application, and the cultivar. Notably, the TA was lower in fruits on grafted vines, while no significant difference was detected in SSC. Similar results were reported by Gong et al. [73].

Grapes are rich sources of phenolic compounds and either flavonoids or non-flavonoids such as anthocyanins, flavonols, flavonoids, phenolic acids, and stilbenes [30]. The highest phenolic compounds in the skin of grape berries were induced by grafting. In addition, Eth application on ungrafted grapes produced the highest phenolic compounds followed by Mel and Eth on grafted grapes. Zhang et al. [74] disclosed an increase in phenolic compound content after grafting in grape berry skin. They suggested that the rootstocks could increase the expression of genes related to the synthesis of phenolic compounds or transcriptional factors that regulate the transcription or post-transcription of genes in the phenolic compound synthesis pathway. In the current study, the maximum antioxidant capacity and phenolic compounds in berries were expressed when plants were sprayed with Eth, reflecting the high correlation between them. Previous studies showed that ethephon has a strong effect on the synthesis of proanthocyanidins and flavonols in the grapes through increasing the gene expression of enzymes related to phenolic compounds synthesis such as VvLAR1, VvLAR2, VvANR, and VvMYBPA1 [35,64,65]. This response was also related to the high activity of PAL (phenylalanine ammonia lyase), the first enzyme in the phenolic compounds biosynthesis pathway [75]. Antioxidant capacity is strongly associated with the phenolic compounds of foods, which is suggested to be beneficial to human health [67,68]. Similar results were observed by Zhang and Zhou [76] in lemon fruits after ethephon treatment.

The color of fruits is considered one of the most important attributes of fruit quality. The grapevines treated with Eth indicated higher a* values and minimum b* values, which showed the more intense red color of grape berries [77]. These results are consistent with the results of anthocyanins in the present study. However, the fruit lightness (L* value) recorded the lowest value under the ABA application. The lower L* values could be due to a greater amount of wax since the opposite situation was true in the control treatment. The results are similar to those observed by Wang et al. [78] in blueberries. Remarkably, grafting revealed a positive and significant improvement in fruit coloration with the highest a* values [62].

The accumulation of anthocyanins increased with the desirable application of growth regulators, as well as the expression of genes related to the biosynthesis of anthocyanins: CHS, F3H, and UFGH. The application of ABA was recognized as an excellent treatment for the accumulation of anthocyanins in berries from the ripening period to harvest. Similar results were observed by Shahab et al. [67], as they disclosed that the anthocyanin daily accumulation rate increased as a result of ABA treatment on 'Benitaka' grapes. Anthocyanins are a group of flavonoids that are usually produced by the secondary metabolites pathway. Marco et al. [79] depicted that the application of ABA enhanced the level of secondary metabolites in grapes. Furthermore, ABA plays a key role in regulating the number of genes at the onset of veraison, including those involved in anthocyanin biosynthesis [80].

Moreover, the expression of ABA receptor genes could be upregulated by grafting [81]. Grafting reinforces the accumulation of the transcription factor VvMYBA1, a regulator of UFGT gene expression, which interacts with the other genes involved in anthocyanin biosynthesis such as CHS and F3H [75,82,83]. Given the crucial role of ABA on pigmentation, it is widely used in promoting fruit coloration, especially for non-climacteric fruits as in 'Crimson Seedless' grapes at a concentration of 400 mg/L [84]. In ungrafted grapevines, the superiority of ethephon over anthocyanin accumulation was consistent with its effect on the biological expression of the studied genes. These findings are in accordance with the results of Dong et al. [85], who reported that ethephon promoted the accumulation of anthocyanins compared to other plant growth regulators. The improvement in anthocyanin accumulation was a result of enhancing the synthesis of UFGT, CHS, and F3H genes, due to their essential role in the anthocyanin synthesis pathway. Previous studies revealed that ethephon, which is a source of ethylene, can induce the biosynthesis of anthocyanins in some fruits, such as apples [86] and plums [32].

PAL is the key enzyme in the phenylpropanoid pathway that produces different phenolic compounds [87]. The application of MeJ and Eth induced PAL activities, which is in harmony with parallel results observed by Sarabandi et al. [88], who demonstrated that MeJ significantly enhanced the expression levels of the VvPAl gene, which led to the higher activity of the PAL enzyme. Additionally, the activity of the PPO enzyme in the fruit tissue causes the oxidation of phenolic compounds, which could significantly reduce anthocyanins as well as the fruit color [89]. In line with our findings, Molla et al. [90] noted that the application of melatonin increased PPO activity in the peel of pomegranate fruit. In the present study, the highest PPO activity was produced by Mel and control treatments, whereas opposite trends were detected for total phenols, antioxidant capacity, POD, and CAT activities. POD and CAT activities increased by Eth and MeJ application and the same trend was observed with PPO activity in ungrafted vines. Ethylene is known to accelerate fruit maturation, which is accompanied by an increase in POD activity [91,92]. Ethylene showed an increment in POD activity in sweet potato, cucumber, pea, tomato, and tobacco.

5. Conclusions

In view of the limitations related to the fruits of 'Crimson Seedless' grape (late ripening and coloration), the results showed that all pre-harvest applications and grafting were highly effective in improving the berry coloration and quality of 'Crimson Seedless' grapes. All pre-harvest applications with grafting showed an enhancement in fruit yield and some of the fruit's attributes such as weight, size, firmness, and removal force. Phytohormones coupled with grafting had a high ability to improve the internal quality of berries in terms of the soluble solid content (SSC), titratable acidity (TA), total phenols, and antioxidant capacity. Our findings showed an improvement in color due to the increase in the anthocyanin concentration in the berry skin. However, the application of ethephon at a concentration of 480 mg/L 7 and 21 days after the beginning of the coloring phase 'Veraison' was the most effective treatment at enhancing the coloration and berry quality of 'Crimson Seedless' grapes.

Author Contributions: Conceptualization, A.-M.S. and M.A.A.; methodology, M.A.A.; software, A.-M.S. and M.A.A.; validation, M.A.A.; formal analysis, A.-M.S. and M.A.A.; investigation, A.-M.S. and M.A.A.; resources, M.A.A.; data curation, M.A.A.; writing—original draft preparation, A.-M.S. and M.A.A.; writing—review and editing, A.-M.S., M.A.A., and M.R.; visualization, A.E.-S.; supervision, M.E.; project administration, A.-M.S.; funding acquisition, M.R. All authors have read and agreed to the published version of the manuscript.

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Data Availability Statement: The datasets generated during the current study are available from the corresponding author upon reasonable request.

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Conflicts of Interest: The authors declare no conflict of interest.

References

1. Food and Agriculture Organisation of the United Nations. Crops and Livestock Products. 2021. Available online: http://www.fao.org/faostat/en/#data/qc (accessed on 30 July 2023).
2. Domingues Neto, F.J.; Pimentel Junior, A.; Borges, C.V.; Cunha, S.R.; Callili, D.; Lima, G.P.P.; Roberto, S.R.; Leonel, S.; Tecchio, M.A. The Exogenous Application of Abscisic Acid Induce Accumulation of Anthocyanins and Phenolic Compounds of the 'Rubi' Grape. *Am. J. Plant Sci.* **2017**, *8*, 2422–2432. [CrossRef]
3. Arias, I.; Lacau, B.; Astrain, J.; Barón, C.; Fenandez-Zurbano, P.; Ferreira, V.; Escudero, A. Effects of Vineyard 'Potential' and Grape Maturation on the Aroma-Volatile Profile of Grenache Wines. *Oeno One* **2019**, *53*, 695–707. [CrossRef]
4. Bouzas-Cid, Y.; Falqué, E.; Orriols, I.; Mirás-Avalos, J.M. Effects of Irrigation over Three Years on the Amino Acid Composition of Treixadura (*Vitis Vinifera* L.) Musts and Wines, and on the Aromatic Composition and Sensory Profiles of Its Wines. *Food Chem.* **2018**, *240*, 707–716. [CrossRef] [PubMed]
5. Ji, X.H.; Wang, B.L.; Wang, X.D.; Shi, X.B.; Liu, P.P.; Liu, F.Z.; Wang, H.B. Effects of Different Color Paper Bags on Aroma Development of Kyoho Grape Berries. *J. Integr. Agric.* **2019**, *18*, 70–82. [CrossRef]
6. Suehiro, Y.; Mochida, K.; Tsuma, M.; Yasuda, Y.; Itamura, H.; Esumi, T. Effects of Gibberellic Acid/Cytokinin Treatments on Berry Development and Maturation in the Yellow-Green Skinned 'Shine Muscat' Grape. *Hortic. J.* **2019**, *88*, 202–213. [CrossRef]
7. Chong, Z.; Hao, C.; Lianjun, W. Effect of ABA Treatment on the Expression of ADH Gene Family and C6 Volatile Production in Table Grape (V. Vinifera Cv. Muscat Hamburg) during Postharvest Storage. *Acta Physiol. Plant.* **2020**, *42*, 55. [CrossRef]
8. Gao, X.T.; Wu, M.H.; Sun, D.; Li, H.Q.; Chen, W.K.; Yang, H.Y.; Liu, F.Q.; Wang, Q.C.; Wang, Y.Y.; Wang, J.; et al. Effects of Gibberellic Acid (GA3) Application before Anthesis on Rachis Elongation and Berry Quality and Aroma and Flavour Compounds in *Vitis vinifera* L. 'Cabernet Franc' and 'Cabernet Sauvignon' Grapes. *J. Sci. Food Agric.* **2020**, *100*, 3729–3740. [CrossRef]
9. Xi, X.; Zha, Q.; He, Y.; Tian, Y.; Jiang, A. Influence of Cluster Thinning and Girdling on Aroma Composition in 'Jumeigui' Table Grape. *Sci. Rep.* **2020**, *10*, 6877. [CrossRef]
10. Downey, M.O.; Dokoozlian, N.K.; Krstic, M.P. Cultural Practice and Environmental Impacts on the Flavonoid Composition of Grapes and Wine: A Review of Recent Research. *Am. J. Enol. Vitic.* **2006**, *57*, 257–268. [CrossRef]
11. Kuhn, N.; Guan, L.; Dai, Z.W.; Wu, B.H.; Lauvergeat, V.; Gomès, E.; Li, S.H.; Godoy, F.; Arce-Johnson, P.; Delrot, S. Berry Ripening: Recently Heard through the Grapevine. *J. Exp. Bot.* **2014**, *65*, 4543–4559. [CrossRef]
12. Nakazawa, A.; Nozue, M.; Yasuda, H.; Takeba, G.; Kubo, H. Expression Pattern and Gene Structure of Phenylalanine Ammonia-Lyase in Pharbitis Nil. *J. Plant Res.* **2001**, *114*, 323–328. [CrossRef]
13. Jaakola, L. New Insights into the Regulation of Anthocyanin Biosynthesis in Fruits. *Trends Plant Sci.* **2013**, *18*, 477–483. [CrossRef]
14. Sicilia, A.; Scialò, E.; Puglisi, I.; Lo Piero, A.R. Anthocyanin Biosynthesis and DNA Methylation Dynamics in Sweet Orange Fruit [*Citrus sinensis* L. (Osbeck)] under Cold Stress. *J. Agric. Food Chem.* **2020**, *68*, 7024–7031. [CrossRef]
15. Wen, W.; Alseekh, S.; Fernie, A.R. Conservation and Diversification of Flavonoid Metabolism in the Plant Kingdom. *Curr. Opin. Plant Biol.* **2020**, *55*, 100–108. [CrossRef] [PubMed]
16. Tian, M.; Ma, W.; Xia, N.; Peng, J.; Hu, R.; Duan, C.; He, F. Soil Variables and Reflected Light Revealed the Plasticity of Grape and Wine Composition: Regulation of the Flavoromics under Inner Row Gravel Covering. *Food Chem.* **2023**, *414*, 135659. [CrossRef]
17. Wang, Y.U.; Li, H.Q.; Gao, X.T.; Lu, H.C.; Peng, W.T.; Chen, W.U.; Li, S.P.; Duan, C.Q.; Wang, J. Influence of Attenuated Reflected Solar Radiation from the Vineyard Floor on Volatile Compounds in Cabernet Sauvignon Grapes and Wines of the North Foot of Mt. Tianshan. *Food Res. Int.* **2020**, *137*, 109688. [CrossRef]
18. Samaan, M.; Nasser, M.A. Effect of Spraying Paclobutrazol (PP333) on Yield and Fruit Quality of Crimson Seedless Grape. *J. Plant Prod.* **2020**, *11*, 1031–1034. [CrossRef]
19. Peppi, M.C.; Fidelibus, M.W. Effects of Forchlorfenuron and Abscisic Acid on the Quality of "Flame Seedless" Grapes. *HortScience* **2008**, *43*, 173–176. [CrossRef]
20. Setha, S. Roles of Abscisic Acid in Fruit Ripening. *Walailak J. Sci. Technol.* **2012**, *9*, 297–308.
21. Yang, F.W.; Feng, X.Q. Abscisic Acid Biosynthesis and Catabolism and Their Regulation Roles in Fruit Ripening. *Phyton-Int. J. Exp. Bot.* **2015**, *84*, 444–453. [CrossRef]
22. Marzouk, H.A.; Kassem, H.A. Improving Fruit Quality, Nutritional Value and Yield of Zaghloul Dates by the Application of Organic and/or Mineral Fertilizers. *Sci. Hortic.* **2011**, *127*, 249–254. [CrossRef]
23. Rattanakon, S.; Ghan, R.; Gambetta, G.A.; Deluc, L.G.; Schlauch, K.A.; Cramer, G.R. Abscisic Acid Transcriptomic Signaling Varies with Grapevine Organ. *BMC Plant Biol.* **2016**, *16*, 72. [CrossRef] [PubMed]
24. Zhang, Y.; Butelli, E.; Martin, C. Engineering Anthocyanin Biosynthesis in Plants. *Curr. Opin. Plant Biol.* **2014**, *19*, 81–90. [CrossRef] [PubMed]

25. Ferrara, G.; Mazzeo, A.; Matarrese, A.M.S.; Pacucci, C.; Punzi, R.; Faccia, M.; Trani, A.; Gambacorta, G. Application of Abscisic Acid (S-ABA) and Sucrose to Improve Colour, Anthocyanin Content and Antioxidant Activity of Cv. Crimson Seedless Grape Berries. *Aust. J. Grape Wine Res.* **2015**, *21*, 18–29. [CrossRef]
26. Olivares, D.; Contreras, C.; Muñoz, V.; Rivera, S.; González-Agüero, M.; Retamales, J.; Defilippi, B.G. Relationship among Color Development, Anthocyanin and Pigment-Related Gene Expression in 'Crimson Seedless' Grapes Treated with Abscisic Acid and Sucrose. *Plant Physiol. Biochem.* **2017**, *115*, 286–297. [CrossRef]
27. Dar, T.A.; Uddin, M.; Khan, M.M.A.; Hakeem, K.R.; Jaleel, H. Jasmonates Counter Plant Stress: A Review. *Environ. Exp. Bot.* **2015**, *115*, 49–57. [CrossRef]
28. Portu, J.; López, R.; Santamaría, P.; Garde-Cerdán, T. Methyl Jasmonate Treatment to Increase Grape and Wine Phenolic Content in Tempranillo and Graciano Varieties during Two Growing Seasons. *Sci. Hortic.* **2018**, *240*, 378–386. [CrossRef]
29. Gómez-Plaza, E.; Bautista-Ortín, A.B.; Ruiz-García, Y.; Fernández-Fernández, J.I.; Gil-Muñoz, R. Effect of Elicitors on the Evolution of Grape Phenolic Compounds during the Ripening Period. *J. Sci. Food Agric.* **2017**, *97*, 977–983. [CrossRef]
30. Bellincontro, A.; Fardelli, A.; De Santis, D.; Botondi, R.; Mencarelli, F. Postharvest Ethylene and 1-MCP Treatments Both Affect Phenols, Anthocyanins, and Aromatic Quality of Aleatico Grapes and Wine. *Aust. J. Grape Wine Res.* **2006**, *12*, 141–149. [CrossRef]
31. An, J.P.; Wang, X.F.; Li, Y.Y.; Song, L.Q.; Zhao, L.L.; You, C.X.; Hao, Y.J. EIN3-LIKE1, MYB1, and Ethylene Response Factor3 Act in a Regulatory Loop That Synergistically Modulates Ethylene Biosynthesis and Anthocyanin Accumulation. *Plant Physiol.* **2018**, *178*, 808–823. [CrossRef]
32. Cheng, Y.; Liu, L.; Yuan, C.; Guan, J. Molecular Characterization of Ethylene-Regulated Anthocyanin Biosynthesis in Plums During Fruit Ripening. *Plant Mol. Biol. Report.* **2016**, *34*, 777–785. [CrossRef]
33. An, J.P.; Yao, J.F.; Xu, R.R.; You, C.X.; Wang, X.F.; Hao, Y.J. Apple BZIP Transcription Factor MdbZIP44 Regulates Abscisic Acid-Promoted Anthocyanin Accumulation. *Plant Cell Environ.* **2018**, *41*, 2678–2692. [CrossRef] [PubMed]
34. Wang, L.; Yang, S.; Ni, J.; Teng, Y.; Bai, S. Advances of Anthocyanin Synthesis Regulated by Plant Growth Regulators in Fruit Trees. *Sci. Hortic.* **2023**, *307*, 111476. [CrossRef]
35. Ni, J.B.; Bai, S.L.; Zhao, Y.; Tao, R.Y.; Teng, Y.W. Ethylene Suppresses Anthocyanin Accumulation in "Red Zaosu" Pear. In *Proceedings of the Acta Horticulturae*; International Society for Horticultural Science (ISHS): Leuven, Belgium, 2020; pp. 513–520.
36. Figueroa, N.E.; Gatica-Meléndez, C.; Figueroa, C.R. Ethylene Application at the Immature Stage of Fragaria Chiloensis Fruit Represses the Anthocyanin Biosynthesis with a Concomitant Accumulation of Lignin. *Food Chem.* **2021**, *358*, 129913. [CrossRef] [PubMed]
37. Arnao, M.B.; Hernández-Ruiz, J. Functions of Melatonin in Plants: A Review. *J. Pineal Res.* **2015**, *59*, 133–150. [CrossRef]
38. Zhang, H.M.; Zhang, Y. Melatonin: A Well-Documented Antioxidant with Conditional pro-Oxidant Actions. *J. Pineal Res.* **2014**, *57*, 131–146. [CrossRef]
39. Murch, S.J.; Hall, B.A.; Le, C.H.; Saxena, P.K. Changes in the Levels of Indoleamine Phytochemicals during Véraison and Ripening of Wine Grapes. *J. Pineal Res.* **2010**, *49*, 95–100. [CrossRef]
40. Meng, J.F.; Yu, Y.; Shi, T.C.; Fu, Y.S.; Zhao, T.; Zhang, Z.W. Melatonin Treatment of Pre-Veraison Grape Berries Modifies Phenolic Components and Antioxidant Activity of Grapes and Wine. *Food Sci. Technol.* **2019**, *39*, 35–42. [CrossRef]
41. Rouphael, Y.; Kyriacou, M.C.; Colla, G. Vegetable Grafting: A Toolbox for Securing Yield Stability under Multiple Stress Conditions. *Front. Plant Sci.* **2018**, *8*, 10–13. [CrossRef]
42. Kumar, R.M.S.; Ji, G.; Guo, H.; Zhao, L.; Zheng, B. Over-Expression of a Grafting-Responsive Gene from Hickory Increases Abiotic Stress Tolerance in Arabidopsis. *Plant Cell Rep.* **2018**, *37*, 541–552. [CrossRef]
43. Lana, G.; Modica, G.; Las Casas, G.; Siracusa, L.; La Malfa, S.; Gentile, A.; Sicilia, A.; Distefano, G.; Continella, A. Molecular Insights into the Effects of Rootstocks on Maturation of Blood Oranges. *Horticulturae* **2021**, *7*, 468. [CrossRef]
44. Parvaneh, T.; Abedi, B.; Davarynejad, G.H.; Ganji Moghadam, E. Enzyme Activity, Phenolic and Flavonoid Compounds in Leaves of Iranian Red Flesh Apple Cultivars Grown on Different Rootstocks. *Sci. Hortic.* **2019**, *246*, 862–870. [CrossRef]
45. Font i Forcada, C.; Gogorcena, Y.; Moreno, M.A. Fruit Sugar Profile and Antioxidants of Peach and Nectarine Cultivars on Almond×peach Hybrid Rootstocks. *Sci. Hortic.* **2013**, *164*, 563–572. [CrossRef]
46. Milošević, T.; Milošević, N.; Mladenović, J. Combining Fruit Quality and Main Antioxidant Attributes in the Sour Cherry: The Role of New Clonal Rootstock. *Sci. Hortic.* **2020**, *265*, 109236. [CrossRef]
47. Cheng, J.; Wei, L.; Mei, J.; Wu, J. Effect of Rootstock on Phenolic Compounds and Antioxidant Properties in Berries of Grape (*Vitis vinifera* L.) Cv. 'Red Alexandria'. *Sci. Hortic.* **2017**, *217*, 137–144. [CrossRef]
48. Bascuñán-Godoy, L.; Franck, N.; Zamorano, D.; Sanhueza, C.; Carvajal, D.E.; Ibacache, A. Rootstock Effect on Irrigated Grapevine Yield under Arid Climate Conditions Are Explained by Changes in Traits Related to Light Absorption of the Scion. *Sci. Hortic.* **2017**, *218*, 284–292. [CrossRef]
49. Da Silva, M.J.R.; Vedoato, B.T.F.; Lima, G.P.P.; Moura, M.F.; Coser, G.M.D.A.G.; Watanabe, C.Y.; Tecchio, M.A. Phenolic Compounds and Antioxidant Activity of Red and White Grapes on Different Rootstocks. *Afr. J. Biotechnol.* **2017**, *16*, 664–671. [CrossRef]
50. Balic, I.; Ejsmentewicz, T.; Sanhueza, D.; Silva, C.; Peredo, T.; Olmedo, P.; Barros, M.; Verdonk, J.C.; Paredes, R.; Meneses, C.; et al. Biochemical and Physiological Study of the Firmness of Table Grape Berries. *Postharvest Biol. Technol.* **2014**, *93*, 15–23. [CrossRef]
51. Lancaster, J.E.; Lister, C.E.; Reay, P.F.; Triggs, C.M. Influence of Pigment Composition on Skin Color in a Wide Range of Fruit and Vegetables. *J. Am. Soc. Hortic. Sci.* **1997**, *122*, 594–598. [CrossRef]

52. Chironi, S.; Sortino, G.; Allegra, A.; Saletta, F.; Caviglia, V.; Ingrassia, M. Consumer Assessment on Sensory Attributes of Fresh Table Grapes Cv "Italia" and "Red Globe" after Long Cold Storage Treatment. *Chem. Eng. Trans.* **2017**, *58*, 421–426. [CrossRef]
53. Martínez-Esplá, A.; Zapata, P.J.; Castillo, S.; Guillén, F.; Martínez-Romero, D.; Valero, D.; Serrano, M. Preharvest Application of Methyl Jasmonate (MeJA) in Two Plum Cultivars. 1. Improvement of Fruit Growth and Quality Attributes at Harvest. *Postharvest Biol. Technol.* **2014**, *98*, 98–105. [CrossRef]
54. Binsan, W.; Benjakul, S.; Visessanguan, W.; Roytrakul, S.; Tanaka, M.; Kishimura, H. Antioxidative Activity of Mungoong, an Extract Paste, from the Cephalothorax of White Shrimp (*Litopenaeus vannamei*). *Food Chem.* **2008**, *106*, 185–193. [CrossRef]
55. Von Wettstein, D. Chlorophyll-Letale Und Der Submikroskopische Formwechsel Der Plastiden. *Exp. Cell Res.* **1957**, *12*, 427–506. [CrossRef] [PubMed]
56. Assis, J.S.; Maldonado, R.; Muñoz, T.; Escribano, M.I.; Merodio, C. Effect of High Carbon Dioxide Concentration on PAL Activity and Phenolic Contents in Ripening Cherimoya Fruit. *Postharvest Biol. Technol.* **2001**, *23*, 33–39. [CrossRef]
57. Kar, M.; Mishra, D. Catalase, Peroxidase, and Polyphenoloxidase Activities during Rice Leaf Senescence. *Plant Physiol.* **1976**, *57*, 315–319. [CrossRef]
58. Chance, B.; Meahly, A.C.I. Catalase Assay by Disappearance of Peroxide. *Methods Enzymol.* **1955**, *2*, 764–775.
59. Singh, R.K.; Martins, V.; Soares, B.; Castro, I.; Falco, V. Chitosan Application in Vineyards (*Vitis vinifera* L. Cv. Tinto Cão) Induces Accumulation of Anthocyanins and Other Phenolics in Berries, Mediated by Modifications in the Transcription of Secondary Metabolism Genes. *Int. J. Mol. Sci.* **2020**, *21*, 306. [CrossRef]
60. Livak, K.J.; Schmittgen, T.D. Analysis of Relative Gene Expression Data Using Real-Time Quantitative PCR and the 2-ΔΔCT Method. *Methods* **2001**, *25*, 402–408. [CrossRef]
61. Duncan, D.B. A Bayesian Approach to Multiple Comparisons. *Technometrics* **1965**, *7*, 171–222. [CrossRef]
62. Jin, Z.X.; Sun, T.Y.; Sun, H.; Yue, Q.Y.; Yao, Y.X. Modifications of 'Summer Black' Grape Berry Quality as Affected by the Different Rootstocks. *Sci. Hortic.* **2016**, *210*, 130–137. [CrossRef]
63. Shahda, M. Effect of Rootstocks on Growth, Yield and Fruit Quality of Red Globe Grape. *Ann. Agric. Sci. Moshtohor* **2016**, *54*, 339–344. [CrossRef]
64. García-Pastor, M.E.; Serrano, M.; Guillén, F.; Castillo, S.; Martínez-Romero, D.; Valero, D.; Zapata, P.J. Methyl Jasmonate Effects on Table Grape Ripening, Vine Yield, Berry Quality and Bioactive Compounds Depend on Applied Concentration. *Sci. Hortic.* **2019**, *247*, 380–389. [CrossRef]
65. Mohamed, A.; El-Salhy, A.; Mostafa, R.; El-Mahdy, M.; Hussein, A. Effect of Exogenous Abscisic Acid (ABA), Gibberellic Acid (GA3) and Cluster Thinning on Yield of Some Grape Cultivars. *J. Plant Prod.* **2019**, *10*, 101–105. [CrossRef]
66. Jia, H.; Zhang, C.; Pervaiz, T.; Zhao, P.; Liu, Z.; Wang, B.; Wang, C.; Zhang, L.; Fang, J.; Qian, J. Jasmonic Acid Involves in Grape Fruit Ripening and Resistant against Botrytis Cinerea. *Funct. Integr. Genom.* **2016**, *16*, 79–94. [CrossRef] [PubMed]
67. Shahab, M.; Roberto, S.R.; Ahmed, S.; Colombo, R.C.; Silvestre, J.P.; Koyama, R.; De Souza, R.T. Anthocyanin Accumulation and Color Development of 'Benitaka' Table Grape Subjected to Exogenous Abscisic Acid Application at Different Timings of Ripening. *Agronomy* **2019**, *9*, 164. [CrossRef]
68. Cantín, C.M.; Fidelibus, M.W.; Crisosto, C.H. Application of Abscisic Acid (ABA) at Veraison Advanced Red Color Development and Maintained Postharvest Quality of "Crimson Seedless" Grapes. *Postharvest Biol. Technol.* **2007**, *46*, 237–241. [CrossRef]
69. Ulas, F.; Fricke, A.; Stützel, H. Effect of Grafting on Yield and Quality of Tomato (*Solanum Lycopersicum* Mill.) Cultivars. *Tanzan. J. Agric. Sci.* **2018**, *17*, 39–45.
70. Rouphael, Y.; Schwarz, D.; Krumbein, A.; Colla, G. Impact of Grafting on Product Quality of Fruit Vegetables. *Sci. Hortic.* **2010**, *127*, 172–179. [CrossRef]
71. Xia, H.; Shen, Y.; Shen, T.; Wang, X.; Zhang, X.; Hu, P.; Liang, D.; Lin, L.; Deng, H.; Wang, J.; et al. Melatonin Accumulation in Sweet Cherry and Its Influence on Fruit Quality and Antioxidant Properties. *Molecules* **2020**, *25*, 753. [CrossRef]
72. El-Anany, A.M.; Hassan, G.F.A.; Ali, F.R. Effects of Edible Coatings on the Shelf-Life and Quality of Anna Apple during Cold Storage. *J. Food Technol.* **2009**, *7*, 5–11.
73. Gong, T.; Zhang, X.; Brecht, J.K.; Serrano, T.; Sims, C.A.; Colee, J.; Zhao, X. Instrumental and Sensory Analyses of Fruit Quality Attributes of Grafted Grape Tomatoes in High Tunnel Organic Production Systems. *J. Sci. Food Agric.* **2022**, *102*, 7268–7281. [CrossRef] [PubMed]
74. Zhang, F.; Zhong, H.; Zhou, X.; Pan, M.; Xu, J.; Liu, M.; Wang, M.; Liu, G.; Xu, T.; Wang, Y.; et al. Grafting with Rootstocks Promotes Phenolic Compound Accumulation in Grape Berry Skin during Development Based on Integrative Multi-Omics Analysis. *Hortic. Res.* **2022**, *9*, uhac055. [CrossRef] [PubMed]
75. Li, Z.H.; Gemma, H.; Iwahori, S. Stimulation of "Fuji" Apple Skin Color by Ethephon and Phosphorus-Calcium Mixed Compounds in Relation to Flavonoid Synthesis. *Sci. Hortic.* **2002**, *94*, 193–199. [CrossRef]
76. Zhang, P.; Zhou, Z. Postharvest Ethephon Degreening Improves Fruit Color, Flavor Quality and Increases Antioxidant Capacity in 'Eureka' Lemon (*Citrus Limon* (L.) Burm. F.). *Sci. Hortic.* **2019**, *248*, 70–80. [CrossRef]
77. Ribeiro, T.P.; de Lima, M.A.C.; Alves, R.E. Maturação e Qualidade de Uvas Para Suco Em Condições Tropicais, Nos Primeiros Ciclos de Produção. *Pesqui. Agropecu. Bras.* **2012**, *47*, 1057–1065. [CrossRef]
78. Wang, Y.W.; Malladi, A.; Doyle, J.W.; Scherm, H.; Nambeesan, S.U. The Effect of Ethephon, Abscisic Acid, and Methyl Jasmonate on Fruit Ripening in Rabbiteye Blueberry (*Vaccinium virgatum*). *Horticulturae* **2018**, *4*, 24. [CrossRef]

79. Tecchio, M.A.; Neto, F.J.D.; Junior, A.P.; da Silva, M.J.R.; Roberto, S.R.; Smarsi, R.C. Improvement of Color and Increase in Anthocyanin Content of Niagara Rosada Grapes with Application of Abscisic Acid. *Afr. J. Biotechnol.* **2017**, *16*, 1400–1403. [CrossRef]
80. Gambetta, G.A.; Matthews, M.A.; Shaghasi, T.H.; McElrone, A.J.; Castellarin, S.D. Sugar and Abscisic Acid Signaling Orthologs Are Activated at the Onset of Ripening in Grape. *Planta* **2010**, *232*, 219–234. [CrossRef]
81. Zhang, Z.; Cao, B.; Li, N.; Chen, Z.; Xu, K. Comparative Transcriptome Analysis of the Regulation of ABA Signaling Genes in Different Rootstock Grafted Tomato Seedlings under Drought Stress. *Environ. Exp. Bot.* **2019**, *166*, 103814. [CrossRef]
82. Liu, M.Y.; Song, C.Z.; Chi, M.; Wang, T.M.; Zuo, L.L.; Li, X.L.; Zhang, Z.W.; Xi, Z.M. The Effects of Light and Ethylene and Their Interaction on the Regulation of Proanthocyanidin and Anthocyanin Synthesis in the Skins of *Vitis vinifera* Berries. *Plant Growth Regul.* **2016**, *79*, 377–390. [CrossRef]
83. Goulas, V.; Manganaris, G.A. Exploring the Phytochemical Content and the Antioxidant Potential of Citrus Fruits Grown in Cyprus. *Food Chem.* **2012**, *131*, 39–47. [CrossRef]
84. Lurie, S.; Ovadia, R.; Nissim-Levi, A.; Oren-Shamir, M.; Kaplunov, T.; Zutahy, Y.; Weksler, H.; Lichter, A. Abscisic Acid Improves Colour Development in "Crimson Seedless" Grapes in the Vineyard and on Detached Berries. *J. Hortic. Sci. Biotechnol.* **2009**, *84*, 639–644. [CrossRef]
85. Dong, T.; Zheng, T.; Fu, W.; Guan, L.; Jia, H.; Fang, J. The Effect of Ethylene on the Color Change and Resistance to *Botrytis cinerea* Infection in "kyoho" Grape Fruits. *Foods* **2020**, *9*, 892. [CrossRef] [PubMed]
86. An, J.P.; Zhang, X.W.; Bi, S.Q.; You, C.X.; Wang, X.F.; Hao, Y.J. The ERF Transcription Factor MdERF38 Promotes Drought Stress-Induced Anthocyanin Biosynthesis in Apple. *Plant J.* **2020**, *101*, 573–589. [CrossRef]
87. Singh, R.; Rastogi, S.; Dwivedi, U.N. Phenylpropanoid Metabolism in Ripening Fruits. *Compr. Rev. Food Sci. Food Saf.* **2010**, *9*, 398–416. [CrossRef] [PubMed]
88. Sarabandi, M.; Farokhzad, A.; Mandoulakani, B.A.; Ghasemzadeh, R. Biochemical and Gene Expression Responses of Two Iranian Grape Cultivars to Foliar Application of Methyl Jasmonate under Boron Toxicity Conditions. *Sci. Hortic.* **2019**, *249*, 355–363. [CrossRef]
89. Dong, Y.; Liu, L.; Zhao, Z.; Zhi, H.; Guan, J. Effects of 1-MCP on Reactive Oxygen Species, Polyphenol Oxidase Activity, and Cellular Ultra-Structure of Core Tissue in 'Yali' Pear (Pyrus Bretschneideri Rehd.) during Storage. *Hortic. Environ. Biotechnol.* **2015**, *56*, 207–215. [CrossRef]
90. Molla, S.M.H.; Rastegar, S.; Omran, V.G.; Khademi, O. Ameliorative Effect of Melatonin against Storage Chilling Injury in Pomegranate Husk and Arils through Promoting the Antioxidant System. *Sci. Hortic.* **2022**, *295*, 110889. [CrossRef]
91. Jeong, S.T.; Goto-Yamamoto, N.; Kobayashi, S.; Esaka, M. Effects of Plant Hormones and Shading on the Accumulation of Anthocyanins and the Expression of Anthocyanin Biosynthetic Genes in Grape Berry Skins. *Plant Sci.* **2004**, *167*, 247–252. [CrossRef]
92. Azuma, A.; Kobayashi, S.; Yakushiji, H.; Yamada, M.; Mitani, N.; Sato, A. VvmybA1 Genotype Determines Grape Skin Color. *Vitis—J. Grapevine Res.* **2007**, *46*, 154–155.