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**Abstract:** Increasing fruit crop production sustainability under climate change, particularly increasing temperatures, is a major challenge in modern agriculture. High temperatures affect apple fruit quality and decrease its color. Herein, we constructed an experimental field under temperature simulation to evaluate climate change mitigation strategies for apples. 'Hongro' apples were subjected to three treatments: (1) cultivation inside a vinyl house for heat treatment (heat induction), (2) cultivation under water irrigation (heat reduction), and (3) cultivation under normal atmospheric temperature (control). At harvest, the fruits of the heat treatment group exhibited poor coloration, with a lower gene expression and pigment accumulation than those of the water irrigation and control groups. Furthermore, the fruit quality of the heat treatment group decreased, with a lower soluble solid content (SSC) and titratable acidity (TA), and smaller fruits. Additionally, a higher fruit disorder (cracking and spots) ratio was observed in the heat treatment group exhibited higher quality indexes (flesh firmness, SSC, and TA) and less cracking than those of the heat treatment and control groups. Heat reduction, including water irrigation, may be used for orchard management to prevent climate change-induced increasing temperatures.

Keywords: climate change; heat stress; water irrigation; fruit skin color; fruit quality; pigment contents

### 1. Introduction

Owing to their widespread consumption and nutritional and economic value, apples have emerged as a valuable fruit; this makes them an important commodity in the fruit crop industry. High-quality apples are an essential source of nutrients, vitamins, and bioactive compounds in the human diet. Apples with attractive color, taste, and nutritional content are the major factors that help decide their quality, increasing the prospects of the fruit in the international market and attracting consumer attention. According to the Korea Agro-Fisheries & Food Trade Corporation, apples are among the top fruits produced in the Korean fruit production industry [1]. However, global warming poses a threat to apple production. According to the Intergovernmental Panel on Climate Change, global warming is continuously increasing the annual mean temperature, with the occurrence of abnormally extreme temperatures [2]. Climate change affects crop productivity, cultivation, and postharvest handling and storage [3,4]. High-temperature climates affect the phenological stages of trees, including accelerating the blooming period [5,6].

Recently, the repercussions of global warming have exacerbated the issue of the inadequate coloration of several fruits, particularly those of which the coloration considerably affects their commercial appeal and value. Temperature plays a vital role in determining fruit coloration and is the most significant factor in this regard [7,8]. The vibrant hues observed in the fruit stem are owing to the synthesis and accumulation of



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**Copyright:** © 2024 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). pigments, particularly anthocyanins and carotenoids. Studies have been undertaken to investigate the pathways responsible for the biosynthesis of these pigments [9–11]. In general, low temperatures promote peel color development, whereas high temperatures impede this development; this emphasizes the phenomenon of poor coloration in response to high temperatures [8,12]. In addition, heat substantially decreases the transcript levels of anthocyanin biosynthesis-related genes and the accumulation of anthocyanins in apple peel [8].

High-temperature climates present significant challenges for agricultural production, particularly in terms of cultivation, which has been well reviewed in woody fruit crops, including apple [13]. Among the various effects of high temperatures on apple orchards, one notable consequence is the poor apple fruit color. High temperatures during fruit development disturb the natural pigmentation process in apple fruit [14]. Anthocyanins and carotenoids, the pigments responsible for the vibrant colors of apple varieties, are particularly sensitive to temperature fluctuations. Increased temperatures can inhibit the biosynthesis of these pigments, leading to poor coloration, as well as color loss in apples. In contrast, we hypothesized that applying a water mist spraying system to avoid hightemperature climates can decrease excess heat production; therefore, trees can produce high-quality and rich-color fruits. Considering these challenges, adaptation strategies must be developed to ensure apple production sustainability. For this, understanding the effect of increased temperatures on the potential damage to apple orchards is vital with rapid climate change. However, the molecular mechanisms underlying the effect of external heat stress during summer on apple fruit development remain unelucidated. Therefore, in this study, we investigated the effects of heat stress on apple fruit coloration and quality in vinyl houses.

#### 2. Materials and Methods

### 2.1. Orchard Management and Fruit Materials

The study was carried out in the experimental field of the Apple Research Center (Gunwi-gun, Daegu, South Korea) (36.28 N, 128.47 E) in 2022. Eight-year-old 'Hongro' apple trees grafted onto M9 rootstock were utilized. The trees were planted with a spacing distance of  $4.0 \times 1.5$  m and cultivated in the same experimental block under identical soil conditions. Trees were managed according to the standard field management with the stable fruit load management of flower/fruitlet thinning (eight fruits per cm<sup>2</sup> of trunk crosssectional area). The experimental field was managed using a pest control management system and irrigation with drip pipelines. The fertilization was applied three times in February, May, and October with the microelement fertilizer (N:P:K = 18:6:17) at 17.5 g per tree. Then, based on orchard management practices, they were divided into three groups, with five trees in each group: (1) trees cultivated inside a vinyl house for excessive heat treatment, (2) trees cultivated under a water mist spraying system, and (3) trees cultivated under normal conditions, which served as the control. The treatment period was 22 days (4–26 July) during summer with weather conditions as shown in Figure 1. Meteorological data including monthly temperatures, rainfall, and precipitation collected at the experimental field during 2022 are reported in Figure S1. During the 22-day treatment period, the water mist spraying system was automatically activated daily from 13:00 to 17:00 via a monitoring system with a timer. During the operating time, the water was pumped from a water stank via an air pump system to pipelines installed above trees and then mist-sprayed from the top of the trees. For the heat treatment group, 'Hongro' apple trees were completely covered with a vinyl house ( $10.9 \times 2.6 \times 5.5$  m in length  $\times$  width  $\times$  height) with a conditional roof/side opening only when the temperature reached 41 °C to avoid overheating. This allowed for the trees to be subjected to an environment of increased temperatures from hot climatic conditions caused by climate change (Figure 2).



**Figure 1.** Real-time data of the (**a**) temperature and (**b**) relative humidity in both indoor and outdoor settings collected from the experimental field in 2022.

The fruits from the three groups were simultaneously harvested in early September 2022. Their quality attributes, coloration, and fruit disorders or symptoms such as cracking, spotting, bitter rot, stink bug, and russeting were assessed. Furthermore, fruit skin tissues were peeled for RNA isolation and pigment content determination.



**Figure 2.** Orchard management practices: (a) trees cultivated under water irrigation, (b) trees cultivated inside a vinyl house for heat treatment, and (c) trees cultivated under normal conditions, which served as the control. For heat treatment, 'Hongro' apple trees were covered with a vinyl house ( $10.9 \times 2.6 \times 5.5$  m in length × width × height). An orchard-based heating system was used to increase the temperatures to imitate the hot climatic conditions caused by climate change (**d**).

# 2.2. Measurements of Fruit Color

Fruit color was measured using a chromameter (CR-400, Konica Minolta, Japan) to accurately quantify the color attributes. Fruits were collected from three random positions. The color indexes were expressed as Hunter values. Hunter values ( $L^*$ ,  $a^*$ , and  $b^*$ ) are described as  $L^*$  (lightness),  $a^*$  (red–green sensation), and  $b^*$  (yellow–blue sensation).

### 2.3. Assessments of Fruit Quality Attributes

Fruit quality attributes, including firmness, soluble solid content (SSC), and titratable acidity (TA), were assessed as described previously [15]. A fruit penetrometer (TR Turoni, Italy) equipped with a  $\emptyset$ 8 mm plunger was used to measure the firmness of peeled-skin fruit picked from three random positions. Firmness was expressed as a force to the plundered area and recorded in Newtons (N) per square centimeter (N/cm<sup>2</sup>). Fruit juice was extracted and used to measure the SSC and TA. A pocket refractometer (PAL-1; Atago, Kyoto, Japan) was used to measure the SSC. TA was determined by titration using 0.1 N NaOH and the results are expressed as malic acid equivalents.

### 2.4. Determination of Fruit Disorders

The harvested fruits were inspected to compare the ratios of fruit disorders such as cracking, spot, bitter rot, stink bug, and russet among the three groups. Fruit cracking is characterized by the rupture of the outer fruit (peel fruit) [16]. Cracking symptoms were assessed by comprehensively examining the fruit surface to identify and classify cracks. Various parameters, including crack location, size, and depth, were used to assess cracking. Spots on the fruit surface [17] were evaluated for size, shape, and coloration to differentiate between different lesion types. Bitter rot symptoms were identified as characteristic brown lesions with a foul odor. Stink bug damage [18,19], as evidenced by puncture marks and necrotic tissues, was recorded and assessed for its effect on fruit quality. Russeting [20], fruit surface discoloration, was quantified using a standardized scoring system based on browning extent and severity.

### 2.5. RNA Extraction and Quantification of Gene Expression

First, peeled-skin fruit tissues were homogenized. Then, the total RNA was extracted using the CTAB method. Before cDNA synthesis, the extracted RNA was treated with DNase to eliminate genomic DNA contamination. An ultraviolet spectrophotometer was used to measure the quality and concentration of the isolated RNA. Subsequently, first-strand cDNA was synthesized from 1.0  $\mu$ g of total RNA using the PrimeScript<sup>TM</sup> 1st strand cDNA Synthesis Kit with oligo dT primer (Takara, Kusatsu, Japan).

Gene expression was analyzed via a quantitative real-time polymerase chain reaction (qRT-PCR) using the LightCycler 480 II Real-Time PCR System (Roche Diagnostics, Mannheim, Germany). The qRT-PCR reaction mixture volume was 20  $\mu$ L and comprised 50 ng of cDNA and 1  $\mu$ L of each (forward and reverse) primer (0.5  $\mu$ M). It was mixed with the LightCycler 480 SYBR Green I Master Mix (Roche, Basel, Switzerland). qRT-PCR was performed according to the previously established conditions [15]. Target gene expression was quantified by normalizing it to that of the reference gene *MDP0000336547* [21]. Tables S1 and S2 list the primer sequences used for the qRT-PCR analysis of anthocyanin and carotenoid biosynthesis genes, respectively.

#### 2.6. Quantification of Anthocyanin Content

To quantify the pigment content, a blender was used to homogenize frozen apple skin samples to a fine powder. Anthocyanins were extracted and quantified as previously described [22]. Briefly, the ground powder of apple skin samples was pre-extracted with 80% acetone and filtered through a 3M filter paper. The extracted samples were diluted to 10 mL after evaporating at 45 °C using a rotary evaporator. The pH differential method was used to measure anthocyanin concentration [23]. The extracted samples were separately mixed with potassium chloride buffer (pH 1.0) and sodium acetate buffer (pH 4.5). Then, the absorbance was measured at 515 and 700 nm using a spectrophotometer (UV-1800, Shimadzu, Kyoto, Japan). Sample absorbance was determined using the following formula: absorbance = pH1.0 (A515 – A700) – pH4.5 (A515 – A700). Anthocyanin content was calculated as follows: anthocyanin = (absorbance × MW × 1000)/( $\varepsilon$  × C), where MW indicates the molecular weight (449.2),  $\varepsilon$  indicates the molar absorptivity of cyanidin-3-galactoside (26,900), and C indicates the concentration of the buffer solution. Anthocyanin content was expressed as micrograms equivalent to cyanidin-3-galactoside per gram of fresh weight.

## 2.7. Quantification of Carotenoid Content

A previously described high-performance liquid chromatography (HPLC) method was used to quantify carotenoid content [24]. Ground samples were suspended in a solution of 3% pyrogallol and 60% KOH for saponification, followed by incubation at 70 °C and cooling. After saponification, a solution containing 1% NaCl and a mixture of ethyl acetate and hexane was added, followed by homogenization by vortexing. After centrifugation, the supernatant was collected and subjected to repeated extractions until clarity, followed

by concentration with nitrogen gas and dissolution in ethanol. This ethanol solution was used as an analytical sample for assessing carotenoid content using HPLC. For HPLC analysis, 10  $\mu$ L of the sample was injected into the system comprising the Shiseido UG 120 column. The mobile phase was methanol, acetonitrile, and dichloromethane at a flow rate of 1 mL/min. Carotenoids were detected at a wavelength of 450 nm. Their concentrations were expressed as equivalents of  $\beta$ -carotene per gram of fresh tissue.

### 2.8. Statistical Analysis

IBM SPSS Statistics (IBM Corp., Armonk, NY, USA) and Microsoft Excel (Microsoft Corp., Redmond, WA, USA) were used to perform statistical analyses. The results are presented as the mean  $\pm$  standard deviation (SD) from triplicate experiments. Group comparisons for significant differences were conducted using Tukey's test (p < 0.05).

### 3. Results

## 3.1. Fruit Coloration under Different Orchard Management Practices

The color indexes of fruit skin were measured to accurately quantify the color attributes after harvesting. The coloration patterns of fruit skin in the three groups were evaluated and expressed as Hunter values ( $L^*$ ,  $a^*$ , and  $b^*$ ) (Figure 3a). The degree of fruit coloration positively correlated with the  $a^*$  value (denoted red); however, it negatively correlated with both  $L^*$  and  $b^*$  values. In general, these values did not significantly differ between the water spray and control groups. However, these values were significantly different from those of the vinyl group, which had higher  $L^*$  and  $b^*$  values but lower  $a^*$  values.

Regarding the Hunter values, the apparent differences in the coloration patterns among the three groups correlated with the color indexes (Figure 3b). The fruit skin of the vinyl house group was colorless, with lower  $a^*$  values and higher  $L^*$  and  $b^*$  values, whereas that of the water spray and control groups exhibited a redder coloration pattern. The fruit of the water spray and control groups exhibited a similar coloration pattern, with no significant differences in the Hunter values ( $L^*$ ,  $a^*$ , and  $b^*$ ).

### 3.2. Effects of Orchard Management on the Fruit Quality Attributes

Fruit quality is determined based on taste, texture, and nutritional content. Along with the overall appearance, including coloration pattern and size, various quality attributes, including firmness, SSC, and TA, were evaluated (Table 1). No significant differences were observed in the fruit weight between the water spray and control groups. However, the fruits of the vinyl house group were lighter than those of the other groups, although they had the same shape (L/D values of 0.88). Comparing the quality attributes of the three groups, the fruit of the water spray group exhibited the highest firmness, SSC, and TA. In contrast, the fruits of the vinyl house group exhibited the lowest values for these quality attributes.

Treatment	Fruit Weight (g)	L/D	Firmness (N)	SSC (°Brix)	TA (%)
Water spray	$232.6\pm24.75~^{\rm a}$	0.88	$66.10\pm2.96$ <sup>a</sup>	$14.3\pm0.42$ a	$0.19\pm0.02~^{\rm a}$
Vinyl house	$197.4\pm19.70~^{\mathrm{b}}$	0.88	$54.70\pm2.36~^{\rm c}$	$12.7\pm0.45$ $^{\rm c}$	$0.13\pm0.01~^{\rm c}$
Control	233.7 $\pm$ 17.41 $^{\rm a}$	0.88	$58.72\pm3.15^{\text{ b}}$	$13.7\pm0.61$ $^{\rm b}$	$0.17\pm0.01$ $^{\rm b}$

Table 1. Fruit quality attributes of the three groups.

L/D: Length-to-diameter ratio. All values are expressed as the mean  $\pm$  SD of 20 individual fruits. Distinct letters indicate statistically significant differences, as determined using Tukey's HSD test (p < 0.05).



**Figure 3.** Differences in the coloration patterns of 'Hongro' fruit under three different treatments based on the orchard management practices. (a) Color indexes ( $L^*$ ,  $a^*$ , and  $b^*$ ).  $L^*$ : light—dark scale.  $a^*$ : green—red scale.  $b^*$ : yellow—blue scale. Data are expressed as mean  $\pm$  SD. Different letters denote a significant difference (p < 0.05). (b) Different coloration patterns in the skin of the fruit from the three groups. Scale bars = 20 mm.

## 3.3. Fruit Disorders

Various common apple fruit disorders were visually inspected for the presence of symptoms such as cracking, spots, bitter rot, stink bug, and russet. Symptoms of injury (Figure 4) were observed in the fruits of all three groups. However, the degree of injury slightly differed among these groups (Table 2). The fruits of the vinyl house group exhibited a higher ratio of cracking, spots, and poor color compared with those of the other groups; however, no differences were observed for these disorders between the water spray and control groups. Furthermore, a higher russeting ratio was observed in water-sprayed fruits. The incidence of bitter rot was lower in the control group but higher in the water spray and vinyl house groups.



**Figure 4.** Visual images of the fruits with popular disorders (cracking, spots, bitter rot, stink bug, and russeting). These representative images were captured using the fruit of the vinyl house group. Scale bars = 20 mm.

Table 2. Fruit disorders among the three groups.

Treatment	Cracking	Spots	Poor Color	Russeting	Bitter Rot
Water spray	10.2 <sup>b</sup>	2.1 <sup>b</sup>	22.3 <sup>b</sup>	2.8 <sup>a</sup>	3.8 <sup>a</sup>
Vinyl house	24.4 <sup>a</sup>	3.2 <sup>a</sup>	32.4 <sup>a</sup>	0.8 <sup>b</sup>	4.0 <sup>a</sup>
Control	12.7 <sup>b</sup>	2.5 <sup>b</sup>	19.7 <sup>b</sup>	1.3 <sup>b</sup>	3.2 <sup>b</sup>

All values are expressed as percentages (%) of the number of disordered fruits per total number of assessed fruits. Distinct letters indicate statistically significant differences, as determined using Tukey's HSD test (p < 0.05).

### 3.4. Expression Profiles of Anthocyanin Biosynthesis-Related Genes

Various coloration patterns depend on the expression of the genes involved in anthocyanin biosynthesis. Gene expression was evaluated at the transcript level to explain the different coloration patterns. Figure 5 illustrates the expression profiles of anthocyanin biosynthesis genes in fruit skin. The expression of these genes significantly differed among the three groups. Gene expression was the lowest in the fruits of the vinyl house group but the highest in the fruits of the water spray group. Furthermore, the expression slightly differed between the water spray and control groups. In general, different treatments resulted in different expression profiles of the biosynthesis-related genes. The expression of these genes differed significantly among the three groups in the following order: water spray > control > vinyl house.

### 3.5. Expression Profiles of Carotenoid Biosynthesis-Related Genes

The expression profiles of carotenoid biosynthesis-related genes refer to the expression patterns of the genes involved in carotenoid biosynthesis. The resulting coloration patterns were investigated via qRT-PCR analysis of the transcript expression of carotenoid biosynthesis-related genes. The expression of these genes differed among the three groups (Figure 6). In the fruits of the vinyl house group, *MdGGPPS*, *MdCRTISO*, *MdLCY* $\varepsilon$ , *MdLCY* $\beta$ , *MdCRH* $\beta$ , and *MdZEP* were downregulated compared with their expression in the other groups. Furthermore, *MdZISO*, *MdPDS*, and *MdZDS* expression was similar in the vinyl house and control groups. When comparing the water spray and control groups, the expression of these genes was higher in the water spray group than in the control group. However, no significant differences were observed in *MdGGPPS* and *MdCRTISO* expression between the water spray and control groups. Only *MdPSY* expression was not significantly different among the three groups. In correlation with the coloration illustrated in Figure 3, the results obtained for gene expression were significantly different among the three groups.



**Figure 5.** The expression profiles of the anthocyanin biosynthesis-related structural genes: phenylalanine ammonia lyase (*MdPAL*), chalcone synthase (*MdCHS*), chalcone isomerase (*MdCHI*), cinnamate-4-hydroxylase (*MdC4H*), 4-coumarate: CoA ligase (*Md4CL*), flavanone 3-hydroxylase (*MdF3H*), dihydroflavonol 4-reductase (*MdDFR*), anthocyanidin synthase (*MdANS*), and UDP-glucose: flavonoid glucosyltransferase (*MdUFGT*) and the transcription factor *MYB10* in the skin tissues of 'Hongro' apples collected from the three treatment groups based on different climatic conditions. Relative expression was normalized to that of the reference gene (*MDP0000336547*). Data are expressed as the mean  $\pm$  SD (error bars) of three biological replicates. Different letters express significant differences (*p* < 0.05) based on Tukey's HSD test.



**Figure 6.** The expression profiles of the carotenoid biosynthesis-related genes: geranylgeranyl pyrophosphate synthase (*MdGGPS*), phytoene synthase (*MdPSY*), phytoene desaturase (*MdPDS*), zeta-carotene isomerase (*MdZISO*), zeta-carotene desaturase (*MdZDS*), carotenoid prolycopene isomerase (*MdCRTISO*), epsilon lycopene cyclase (*MdLCY* $\varepsilon$ ), beta lycopene cyclase (*MdLCY* $\beta$ ), and zeaxanthin epoxidase (*MdZEP*) in the skin tissues of 'Hongro' apples collected from the three treatment groups. Relative expression was normalized to that of the reference gene (*MDP0000336547*). Data are expressed as the mean  $\pm$  SD (error bars) of three biological replicates. Different letters express significant differences (p < 0.05) based on Tukey's HSD test.

## 3.6. Pigment Accumulation

Both anthocyanins and  $\beta$ -carotene are essential pigments that contribute to the coloration and nutritional value of apples. Various factors, including genetics, environmental conditions, and developmental stage, affect the accumulation of these pigments in ap-

ples [25,26]. The color level depends on the concentrations of anthocyanins and  $\beta$ -carotene present in the tissues, where they are synthesized via the anthocyanin and carotenoid biosynthesis pathways, respectively. These pigments are the downstream products of the biosynthesis pathways. In addition to investigating different coloration patterns via the expression of a given gene, we analyzed the accumulation of these pigments (Figure 7). Similar to the coloration patterns, anthocyanin (Figure 7a) and  $\beta$ -carotene (Figure 7b) accumulation was the lowest in the fruit skin of the vinyl house group. However, their accumulation did not significantly differ between the water spray and control groups.



**Figure 7.** Pigment accumulation in the skin of 'Hongro' apples. (a) Anthocyanin and (b)  $\beta$ -carotene. Data are expressed as the mean  $\pm$  SD of three biological replicates. Different letters represent significant differences (p < 0.05) based on Tukey's HSD test.

The lowest anthocyanin accumulation was observed in the fruit skin tissues of the vinyl house group, with a value of 32  $\mu$ g/g of FW. However, its accumulation was higher in the water spray and control groups (47.5 and 43.5  $\mu$ g/g FW, respectively). For  $\beta$ -carotene, the accumulation amount was significantly higher in the fruits of the water spray (3.35  $\mu$ g/g FW) and control (3.30  $\mu$ g/g FW) groups than in those of the vinyl house group (2.96  $\mu$ g/g of FW).

## 4. Discussion

High-temperature climates can significantly affect apple fruit coloration, leading to poor pigmentation and aesthetic quality. When apple trees are exposed to prolonged high-temperature periods, the natural pigmentation processes are disrupted. Excessive heat can interfere with the production of pigments such as anthocyanins and carotenoids which are responsible for the vibrant hues in apples, resulting in fruits having less attractive colors [7,8,27], even though they are induced at low temperatures [12,28]. Furthermore, elevated temperatures may accelerate fruit ripening, leading to premature color changes and uneven pigmentation across the surface of the fruit [29]. In the present study, the treatment of 'Hongro' apple fruits with different orchard-based heating systems resulted in on-tree fruits having different coloration patterns. In high-temperature climates, colorless apple fruits grew inside the vinyl house (Figures 3, S2 and S3). At the molecular level, coloration results from pigment accumulation, which is primarily driven by the anthocyanin and carotenoid biosynthesis pathways. Therefore, we evaluated the expression of the genes involved in the anthocyanin and carotenoid biosynthesis pathways in the fruit skin tissue to elucidate the vibrant coloration of the fruits collected from the three groups (Figures 5 and 6). We observed that the gene expression differed among the three groups, with most genes exhibiting the lowest expression in fruits of the vinyl house group.

Moreover, the accumulation of pigments such as anthocyanins and  $\beta$ -carotene was analyzed (Figure 7). Regarding fruit coloration patterns and transcript levels, the fruits grown under high temperatures (vinyl house group) accumulated the lowest amounts of anthocyanins and  $\beta$ -carotene. However, the amounts of these pigments were not significantly different in the fruits of the other two groups (water spray and control) and

were higher than those in the fruits of the vinyl house group. Similarly, previous studies have revealed that low temperatures induce fruit skin color by increasing the expression of anthocyanin biosynthesis genes and their accumulation in apples [30,31], pears [32], and grapes [33]. The application of the heating system to orchard-based on-tree apple fruits decreased color and anthocyanin concentrations [8]. In addition to anthocyanin synthesis-related structural genes, transcription factors are also involved in regulating anthocyanin biosynthesis and controlling fruit color [34,35]. High temperatures inhibit the activation of the MBW complex, weakening apple fruit color by downregulating the genes associated with the anthocyanin biosynthesis pathway [8]. In the present study, we discovered that together with the anthocyanin biosynthesis-related genes, the expression of the transcription factor MYB10 was decreased in the fruits cultivated in the vinyl house (Figure 5). Exposure to prolonged periods of high temperatures not only decreased the expression of the genes associated with the anthocyanin biosynthesis pathway and the accumulation of anthocyanin, but also the expression of carotenogenic genes and the accumulation of  $\beta$ -carotene. Taken together, these results suggest that high temperatures downregulate pigment biosynthesis-related gene expression and accumulation, resulting in poor coloration.

Elevated temperatures can have detrimental effects on various aspects of apple quality, including texture, flavor, and storability [36,37]. High temperatures accelerate ripening, resulting in the premature softening of the fruit and a decline in texture [38]. This can decrease crispness and firmness, which are often considered freshness attributes by consumers. Additionally, the flavor profile of apples may be compromised under high-temperature conditions, with some varieties experiencing decreased sweetness and overall flavor intensity. Furthermore, high temperatures can shorten the shelf life of apples, increase susceptibility to postharvest disorders, and reduce storability [37,39]. In the present study, in addition to the coloration patterns, the effects of orchard management on fruit quality attributes were investigated (Table 1). Apples cultivated under different conditions exhibited different fruit quality attributes. The fruits cultivated under high-temperature climates (vinyl house) had low quality attributes, including firmness, SSC, and TA, compared with the fruits of other groups. In contrast, under cool-down conditions with water spraying, the fruits exhibited a higher firmness index, SSC, and TA. Furthermore, during the experimental period, the average temperature inside the vinyl house was higher than the outside temperature by 5.3 °C during daytime; however, it was similar at nighttime (Figure 1).

The high temperature inside the vinyl house not only affected the color and quality of the fruit, but also tree growth. Notably, new shoots emerged from 'Hongro' apple trees cultivated inside the vinyl house during fruit maturing (Figure S4). This phenomenon is rare because buds or new shoots develop only during the flowering and fruiting stages. Furthermore, the trees cultivated in the vinyl house had more fruits than those in the other treatment groups (Table S3). This may explain why fruits cultivated in the vinyl house were smaller than those cultivated in the other conditions (Table 1). Another reason for the smaller fruit set in the vinyl house may be the development of new shoots; these trees share energy for new shoot growth instead of focusing on fruit development. A previous study revealed that the extremely high temperatures in plastic film greenhouses affected the aboard range, including coloration, quality, and shoot growth, as observed in mandarin fruit. The application of 8 °C-higher temperature than the atmospheric temperature decreased skin fruit coloration and total soluble sugar, total flavonoid, and phenolic contents [40].

Finally, to investigate the effect of different climates on fruit quality, fruit damage symptoms were categorized and documented. Fruit disorders encompass various symptoms that can affect the appearance, texture, flavor, and overall quality of apples. These disorders can arise from various factors, including environmental conditions, cultural practices, pests, diseases, and genetics. The appearance of injury symptoms in apples primarily affects fruit quality, hindering their marketability.

Overall, the different climatic conditions resulted in a broad range of apple qualities. Apples exposed to high-temperature climates exhibited poor color and more symptoms of injury, decreasing their overall quality. The avoidance of high temperatures by applying a water irrigation system can prevent excessive heat, induce coloration, and improve the quality of apples. Addressing these challenges may require implementing strategies, including avoiding high temperatures, developing cultivars more tolerant to warming, and moving production areas to adopt cultural practices to mitigate the effects of climate change-produced high temperatures on fruit production [5,41,42]. Furthermore, the importance of implementing mitigation strategies should be considered to ensure the continuous production of high-quality fruits in the face of changing climatic conditions.

#### 5. Conclusions

In conclusion, the study demonstrates that prolonged exposure to high-temperature climates significantly impacts apple fruit quality, particularly coloration and the overall aesthetic appeal. The disruption of pigment production pathways, including anthocyanins and carotenoids, at high temperatures leads to poor fruit pigmentation and compromised color development. This temperature-induced effect extends beyond coloration, affecting various quality attributes such as texture, flavor, firmness, and fruit disorder of apples. The observed decline in fruit quality under high-temperature conditions underscores the urgent need for mitigation strategies, including the use of water irrigation systems to cool orchards. We found that trees cultivated under a water mist spraying system exhibited higher fruit quality indicators (firmness of the flesh, SSC, and TA) with less cracking. Therefore, water mist spraying systems can be used in orchard management to prevent the effects of high temperatures caused by climate change. Addressing these challenges is essential to ensure continuous high-quality fruit production despite changing climatic conditions. Implementing adaptive cultural practices and selecting appropriate cultivars are critical steps toward mitigating the adverse effects of rising temperatures on apple fruit production and maintaining the overall orchard productivity and fruit marketability. By prioritizing these strategies, growers can navigate the impacts of climate change on apple production and sustainably meet consumer demands for high-quality fruit in the future.

**Supplementary Materials:** The following supporting information can be downloaded at: https://www. mdpi.com/article/10.3390/agriculture14050761/s1. Table S1: qRT-PCR primer list used for analysis the expression levels of anthocyanin biosynthesis genes. Table S2: qRT-PCR primer list used for analysis the expression levels of carotenoid biosynthesis genes. Table S3: The growth characteristics of apple tree grown under different orchard-based managements. Figure S1: Meteorological data collected at the experimental site of the Apple Research Center (Gunwi-gun, Daegu, Republic of Korea) in 2022. Figure S2: Apparent differences in the coloration pattern of on-tree fruits among the three groups were subjected to different orchard-based management. Figure S3: Variant coloration patterns of 'Hongro' fruit at different treatments among three groups based on orchard management practices at harvest. Figure S4: The emergence of new shoots was observed in 'Hongro' apple trees grown inside the vinyl house during fruit maturing.

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