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Changes in Sucrose and Sorbitol Metabolism Cause Differences in the Intrinsic Quality of Peach Fruits Cultivated in Field and Greenhouse Environments

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Abstract: Fruit trees grow in complex environments where various environmental factors are related to each other, exerting a comprehensive effect on fruit quality. In this study, diurnal variations in environmental indices in the field and greenhouse were recorded, and the changes of leaf photosynthetic assimilate metabolism and fruit soluble sugar accumulation in peach (*Prunus persica*) under the influence of a comprehensive environment were explored. The results showed that the field environment was more favorable for peach photosynthesis, and more sucrose, glucose and fructose could be accumulated compared with the greenhouse environment. In addition, more sorbitol was converted into glucose and fructose in field fruits. Therefore, field fruits exhibited a particularly greater increase in the fructose content, which greatly increased the sweetness of field fruits. This study revealed changes in the pattern of sucrose and sorbitol metabolism in peaches grown in the field and greenhouse, and analyzed the possible reasons and mechanisms of fruit intrinsic quality differences. This research will provide a theoretical basis and reference for the regulation of fruit quality in the greenhouse environment.

Keywords: peach; field; greenhouse; sugar metabolism; fruit quality

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1. Introduction

Fruit quality is a broad concept, including physical properties (size, shape and firmness), sensory properties (sweetness, acidity, texture and aroma), appearance factors (fruit surface smoothness and color), nutrition and characteristics related to food safety [1,2], all of which may affect consumers' purchase decisions and fruit sales. The growth cycle of fruit trees includes many important stages, such as shoot growth, flower bud differentiation, fruit development and dormancy. During these periods, numerous factors affect fruit quality, including fruit tree varieties, management measures and cultivation environment, among which the environment is a very important factor affecting the quality of fruit trees [3,4]. Karagiannis et al. [5] considered that high-altitude areas with large differences in diurnal temperature and ultraviolet radiation intensity might increase the accumulation of anthocyanin in peach peel, and increase the redness of peach peel. Lopez et al. [6] reported a significant relationship between water stress and soluble solid contents in fruit. High temperature conditions promoted fruit ripening and skin reddening in the early stages of fruit development in 'KU-PP2' peaches, while high temperatures negatively affected fruit quality in the later stages of fruit development [7]. The color, size, soluble solids concentration and dry matter content of peach fruit were closely related to light [8]. Canopy position changed fruit quality by affecting the specific environment of peach fruit growth (e.g., light, temperature, water, etc.). High canopy light was evenly distributed and effective for a long period of time, facilitating fruit ripening and the accumulation of sorbitol and sucrose [9].

Peach belongs to the Rosaceae family and is a small deciduous tree. It is one of the most economically valuable imported fruits in the world, particularly in temperate regions [10], including China, the Mediterranean region (Italy and Spain) and the United States [11]. With the development of modern agriculture, global agricultural production has begun to change from traditional methods to greenhouse cultivation, forming a multifunctional system integrating greenhouse manufacturing, production materials and environmental control [12]. Due to the small size of the peach tree, it is easy to cultivate and manage, and it has the characteristics of a high yield of fruits that can be eaten fresh. Therefore, protected cultivation may promote early maturity, facilitate market placement as soon as possible and improve economic benefits. Therefore, the peach is considered one of the most valuable protected cultivated tree species [13,14]. However, environmental factors directly related to fruit growth and development in facility cultivation are limited, such as light, temperature, water and carbon dioxide [15]. These features make facility-cultivated peach fruit lighter in flavor, lower in sugar content and poorer in intrinsic quality than the peach fruit grown in the field environment, which affects consumers' willingness to buy peaches cultivated in greenhouses and substantially affects sales [16]. Therefore, the differences in ecological factors between the field and greenhouse environments were analyzed in this study, and the changes in sugar metabolism and possible causes during the process of peach fruit growth and development were compared and explored.

The intrinsic quality of peach fruit is composed of firmness, sweetness, acidity, aroma and nutrients, and is mainly related to the composition of sugars and the ratio of sugars to acid. Soluble sugars that accumulate in fruit mainly include sucrose, fructose, glucose and sorbitol [17,18]. Among them, fructose has the highest sweetness, and sweetness affects consumers' satisfaction with peaches [19]. Sugar metabolism in the Rosaceae plant family is a complex regulatory network, including biosynthesis of carbohydrates in source tissues, long-distance transport of phloem solutes, metabolism and accumulation in fruit sinks [20–23]. In recent years, the peach industry has rapidly developed. Despite numerous reports on the fruit quality of greenhouse-grown peaches, few studies have examined the effects of comprehensive ecological factors between the greenhouse environment and field environment on fruit quality. As numerous environmental factors are responsible for the difference in the quality of fruits grown inside and outside the greenhouse, this study did not employ a single environmental factor treatment, but rather compared and analyzed the difference between the field environment and the greenhouse environment and explored the regular pattern of sugar metabolism during the process of fruit development in the two environments. The mechanisms involved in the differences in the intrinsic quality of fruit grown inside and outside the greenhouse provide a theoretical basis and reference for improving the fruit quality of greenhouse-cultivated fruit trees.

2. Materials and Methods

2.1. Plant Material

The 3-year-old peach tree 'Zhongyou 16' (*Prunus persica* var. *nectarina* cv. Zhongyou 16) was used as the experimental material. Trees were planted in the field and greenhouse in the same region (Linyi, Shandong Province; 118°83′ E, 35°18′ N). Plant spacing and row spacing were 0.8 m × 1.5 m. The tree height was 2 m, and the crown width was 1.5 m. Tree growth was consistent. Five trees were randomly selected.

The sampling stage was divided into the young fruit period, first expansion period, stone hardening period, second expansion period and mature period, according to the fruit development period. The sampling sites were leaves, fruits and the phloem of shoots. The material was washed with distilled water, dried with clean gauze, frozen in liquid nitrogen and stored in a −80 °C ultralow temperature freezer. The specific sampling times are shown in Table 1, and the fruit development curve is shown in Supplementary Figure S1.

Table 1. Sample collection times.

Treatment	Sampling Time	Days after Full Bloom	Corresponding Growth Period
Greenhouse	23 March	20	Young fruit period (YFP)
	6 April	35	The first expansion period (FEP)
	20 April	50	Stone hardening period (SHP)
	4 May	65	The second expansion period (SEP)
	18 May	80	The mature period (MP)
Field	2 May	20	Young fruit period (YFP)
	16 May	35	The first expansion period (FEP)
	30 May	50	Stone hardening period (SHP)
	13 June	65	The second expansion period (SEP)
	27 June	80	The mature period (MP)

2.2. Determination of Environmental Indices under Greenhouse and Field Conditions

The environmental conditions of the greenhouse and field were determined using a JL-18 air temperature and humidity light recorder, a JL-01 soil temperature and humidity recorder and a JL-28 carbon dioxide recorder (Qingyi Electronic Technology Co., Ltd., Handan, China). The temperature, humidity, light intensity and carbon dioxide concentration during the growth and development of plants were recorded.

2.3. Determination of Photosynthetic Parameters

On sunny days, we measured the photosynthetic parameters of functional leaves using a CIRAS-3 portable photosynthesis system (PP-Systems, Boston, MA, USA). An LED red and blue light source was used in the assessment, and the light intensity was set to 1200 μmol·m^{-2}·s^{-1}. The open-air path system was adopted, and the leaf chamber temperature was 25 °C.

2.4. Sugar Extraction and Measurements of the Sugar Contents

The 1 g samples of different plant parts were ground into powder, placed in a 50 mL centrifuge tube and 20 mL deionized water was added. After leveling, the supernatant solution was obtained by centrifugation at 4 °C and 10,000 rpm for 20 min. The supernatant was filtered with a 0.22 μm water system filter. The samples were degassed by ultrasonic before injection.

An American Beckman P/ACE high-performance capillary electrophoresis instrument 2000 system was used. The quartz capillary was 50 μm × 75 cm (Yongnian Chromatography Factory, Handan, China). The following electrophoretic conditions were employed: buffer solution, 10 mmol·L^{-1} sodium benzoate + 0.3 mmol·L^{-1} CTAB pH 12.5; voltage, 20 kV; negative electrode injection, 0.5 Psi pressure with a 3 s injection; temperature, 25 °C; and 214 nm indirect detection wavelength. The microinjection method was used [24].

For the qualitative and quantitative analyses of sugar components, all standards were purchased from Sigma-Aldrich. The qualitative analysis was performed by assessing the migration time of the sample and the standard. The single and mixed standards of sucrose, glucose, fructose and sorbitol were respectively detected, and the samples were compared with the standards. The external standard method was used for the quantitative analysis. The sugar standard sample was prepared as a raw solution with a mass concentration of 5 g·L^{-1}. The stock solution was diluted to 200, 400, 600, 800, 1000, 1400, 1800 and 2200 mg·L^{-1}, and then used to generate the standard curve. The soluble sugar content was calculated according to the standard curve of sucrose, glucose, fructose and sorbitol and the sample peak areas.

2.5. Sample Extraction and Enzyme-Linked Immunosorbent Assay (ELISA) Measurements

Frozen samples of peach leaves, fruits and phloem (0.5 g) were extracted in phosphate-buffered saline (PBS; pH 7.4; TransGen Biotech, Beijing, China) (1:9, *m*/*v*) and fully homog-

enized. The activities of sugar metabolism-related enzymes, including sucrose synthase (SUS), sucrose phosphate synthase (SPS), vacuolar acid invertase (VAINV), neutral invertase (NINV), sorbitol-6-phosphate dehydrogenase (S6PDH), sorbitol dehydrogenase (SDH), sorbitol oxidase (SOX), hexokinase (HK) and fructose kinase (FK), were determined using a Plant ELISA Kit according to the manufacturer's instructions (Jiangsu Kete Biotechnology Co., Ltd., Yancheng, China) [22].

2.6. RNA Extraction and Quantitative PCR

Total RNA was extracted from 500 mg of frozen (−75 °C) leaf, fruit and phloem samples using an RNAprep Pure Plant Kit (Polysaccharides and Polyphenolics-rich; Tiangen, Beijing, China). RNA was treated with RNase-free DNase (Takara, Dalian, China) to avoid DNA contamination. The single-stranded cDNAs were synthesized from 1 μg of RNA using a Prime Script RT reagent kit with gDNA Eraser (Takara, Dalian, China). qRT-PCR was performed with a gene-specific primer pair, and the peach GAPDH gene was used as an internal control [25]. Quantitative reverse transcription-PCR (qRT-PCR) was performed using SYBR Premix Ex Taq (Takara, Dalian, China) with a CFX96 real-time PCR detection system (Bio-Rad, Hercules, CA, USA). Three biological replicates were used for each analysis. The relative expression levels were calculated using the $2^{-\Delta\Delta CT}$ method. Detailed information on the genes and their specific primers is provided in Supplementary Table S1.

2.7. Statistical Analyses

Each treatment was measured with three biological replicates, and the data were presented as the means ± standard deviations (SDs). The data were analyzed using analysis of variance and significance was determined with IBM SPSS Statistics 21.0 (SPSS Inc., Chicago, IL, USA). Values with $p < 0.05$ were considered statistically significant.

3. Results

3.1. Diurnal Variation in Environmental Indices in the Field and Greenhouse

During the fruit maturation period under field and greenhouse conditions (May in the greenhouse and July in the field), diurnal changes in environmental indices were recorded. According to Figure 1, the trends of the environmental indices in the field and greenhouse were similar in the fruit maturation period, but the numerical values were quite different. The soil temperature showed a single peak curve. The peak temperature in the greenhouse was 25.9 °C, which was significantly lower than the field peak temperature of 37.9 °C and appeared earlier than that in the field (Figure 1A). Soil humidity was very stable, approximately 21% in the greenhouse and 30% in the field, with almost no fluctuation (Figure 1B). The air temperature first increased and then decreased. The highest temperature in the greenhouse was 28.6 °C at 12:00, and that in the field was 34.7 °C at 14:00; the difference was significant (Figure 1C). The trend for air humidity was opposite to that of air temperature, showing a trend of first decreasing and then increasing. The air humidity ranged from 40–90% in the greenhouse and 30–60% in the field, and the fluctuation range in the field was less than that in the greenhouse (Figure 1D). In addition, concentrations of CO_2 in the two environments alternately changed. The CO_2 concentration range in the greenhouse was 620–800 ppm. The concentration was lower in the daytime and higher at night. The CO_2 concentration range in the field was 650–750 ppm. The concentration exhibited the opposite trend compared to that in the greenhouse. Specifically, the concentration was higher in the daytime and lower at night (Figure 1E). The maximum light intensity in the field environment was approximately 60,000 l×, whereas that in the greenhouse was less than 40,000 l×. The difference was significant. The light intensity in the field was also higher than that in the greenhouse (Figure 1F).

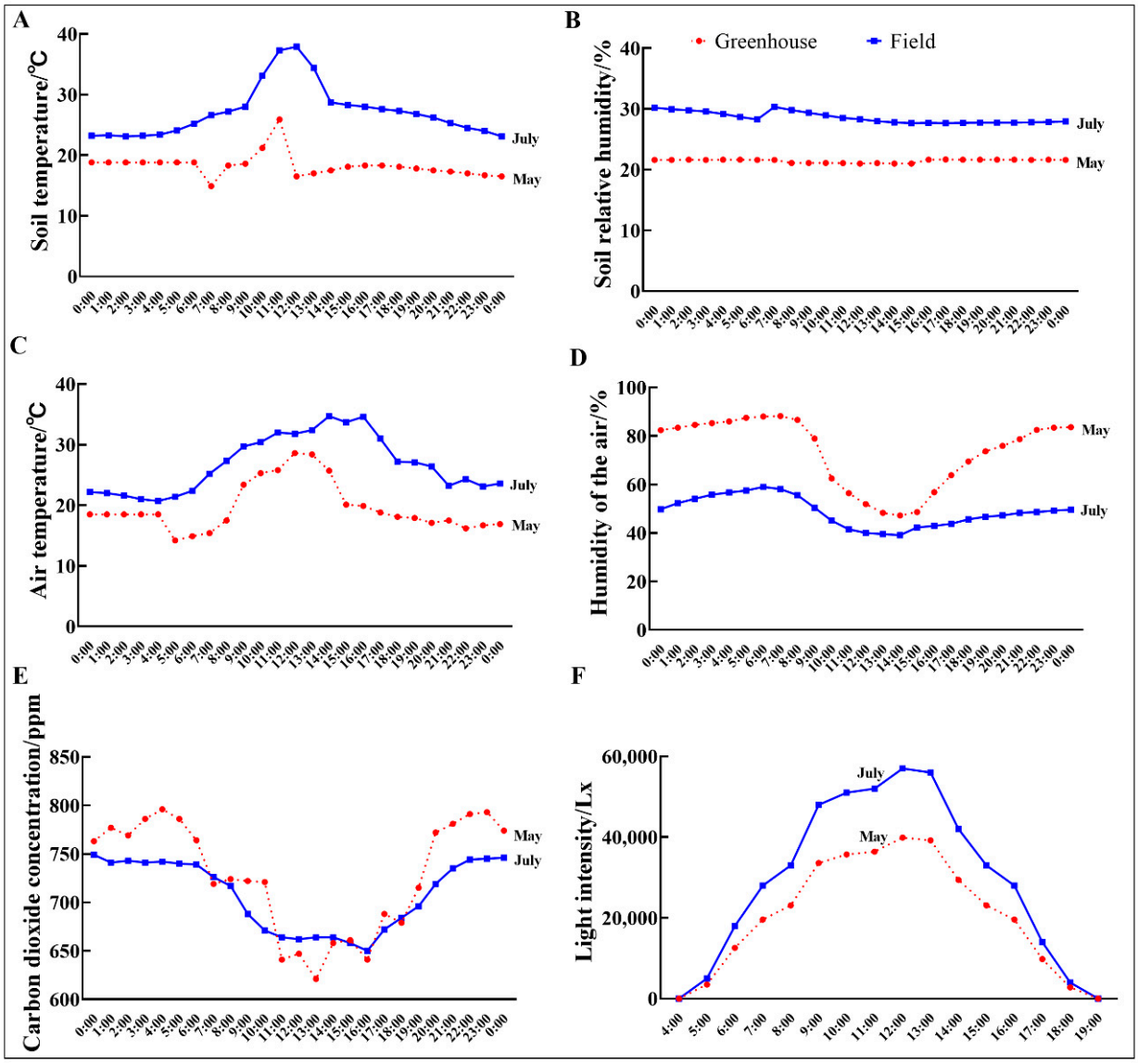

Figure 1. Diurnal variation in the field and greenhouse environments during fruit ripening. (**A**–**F**) present data on soil temperature, soil relative humidity, air temperature, air humidity, CO_2 concentration and light intensity, respectively. The measurement date in the greenhouse environment was 18 May, and that in the field environment was 27 July. Due to the similar diurnal variations in field and greenhouse environmental indicators at maturity, only the diurnal variation at harvest time is presented.

3.2. Changes in the Photosynthetic Parameters of Leaves

We measured the photosynthetic parameters of leaves under field and greenhouse conditions. As shown in Figure 2, the Pn of leaves fluctuated between 15–17 $\mu mol \cdot m^{-2} \cdot s^{-1}$ in the field environment, whereas the Pn of leaves in the greenhouse was approximately 14 $\mu mol \cdot m^{-2} \cdot s^{-1}$. In the five stages of fruit development, Pn in the field increased by 9.69%, 5.42%, 13.62%, 9.21% and 12.71%, respectively, compared with that in the greenhouse, and the differences were significant. In addition, Gs and Tr in the field environment were also significantly higher than those in the greenhouse, whereas Ci showed the opposite trend.

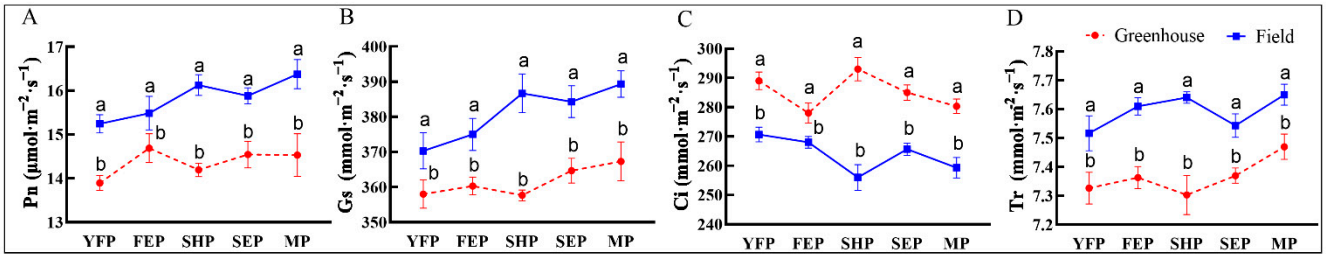

Figure 2. Photosynthetic parameters of peach leaves in the field and greenhouse. (**A**–**D**) show the photosynthetic rate (Pn), stomatal conductance (Gs), intercellular CO_2 concentration (Ci) and transpiration rate (Tr), respectively. The measurement dates were 3.23–5.18 in the greenhouse environment and 5.2–7.27 in the field environment. The data are presented as the means ± SDs from three biological replicates. Different lowercase letters indicate significant differences ($p < 0.05$).

3.3. Changes in Soluble Sugar Contents

As shown in Figure 3A, the trend for changes in the sucrose content in leaves was similar in the two environments, initially increasing and subsequently decreasing. The peak occurred during the stone hardening period in the greenhouse compared with the second expansion period that occurred in the field. No significant differences were noted in other periods, except in the second expansion period. Throughout the fruit development period, the change in sorbitol content in the leaves was relatively stable under both environmental conditions. In addition, glucose and fructose levels decreased in the leaves of plants grown in the field and the greenhouse. However, the fructose content in leaves of plants grown in the field was significantly higher than that in the greenhouse. In addition, the sorbitol content in leaves was maintained at approximately 15 mg·g^{-1} FW under both environmental conditions, which was higher than that in the other two parts of the plant assessed.

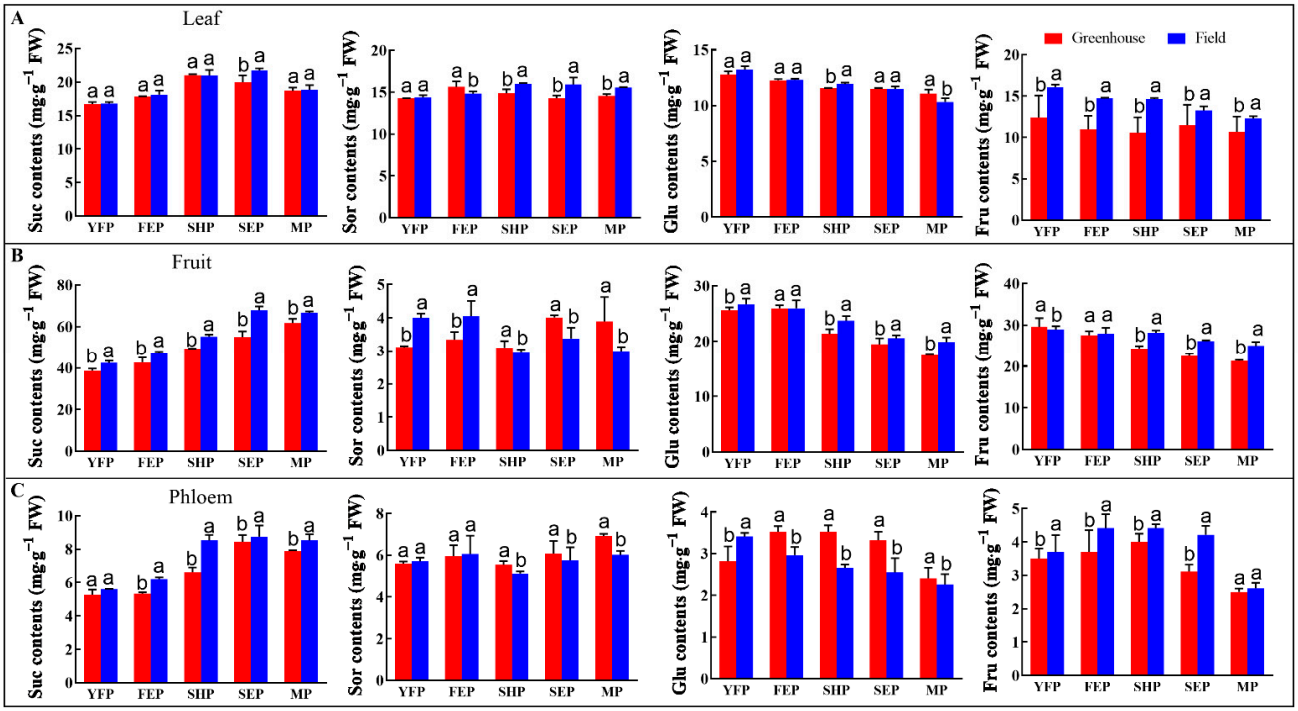

Figure 3. Sugar contents in different parts of peach fruit grown in the field and greenhouse. (**A**–**C**) show the contents of sugar components in leaves, fruits and phloem, respectively. Sucrose, glucose, fructose and sorbitol are abbreviated as Suc, Glu, Fru and Sor, respectively. The data are presented as the means ± SDs obtained from three biological replicates. Different lowercase letters indicate significance ($p < 0.05$).

As illustrated in Figure 3B,C, the sucrose content continuously increased during fruit development and ripening, and a similar trend was also observed in the phloem. The sucrose content in fruit and phloem in the field was higher than that in the greenhouse. The sucrose content in peach fruit from trees grown in the field and greenhouse was 66.74 mg·g^{-1} FW and 61.76 mg·g^{-1} FW, respectively. The sorbitol content in fruit and phloem was maintained at a low range of 3–6 mg·g^{-1} FW, and the pattern of changes was similar. Specifically, in the early stage of fruit development, the sorbitol content in fruit and phloem from trees grown in the field was greater than that in the greenhouse, whereas the opposite result was obtained in the later stage of fruit development. However, glucose and fructose contents in fruits gradually decreased, which was opposite to the trend for sucrose. In addition, the contents of glucose and fructose in fruits from trees grown in the field were significantly higher than those grown in the greenhouse in the late stage of fruit development.

3.4. Changes in the Activities of Key Enzymes Involved in Sugar Metabolism

3.4.1. Activities of Key Enzymes Involved in Sucrose Metabolism

Regarding sucrose metabolism, we mainly analyzed the activities of sucrose synthase (SUS), sucrose phosphate synthase (SPS), vacuolar acid invertase (VAINV) and neutral invertase (NINV). SUS and SPS activities in leaves from plants grown in the field environment were significantly higher than those in plants grown in the greenhouse. At the fruit maturation stage, the VAINV activity in leaves from trees in the field was significantly higher than that in leaves from trees cultivated in the greenhouse, whereas the NINV activity in leaves from trees grown in the greenhouse was significantly higher than that in leaves from plants grown in the field (Figure 4A). Regardless of the greenhouse or field environment, the SPS activity in the fruit increased, whereas the VAINV and NINV activities in the fruit continued to decrease. In the mature stage of the fruit, the difference in NINV activity between the field and the greenhouse conditions was not significant (Figure 4B). Opposite trends for changes in SPS and VAINV activities in phloem were observed, but the enzyme activities detected in plants grown in the field were greater than those in plants grown in the greenhouse (Figure 4C).

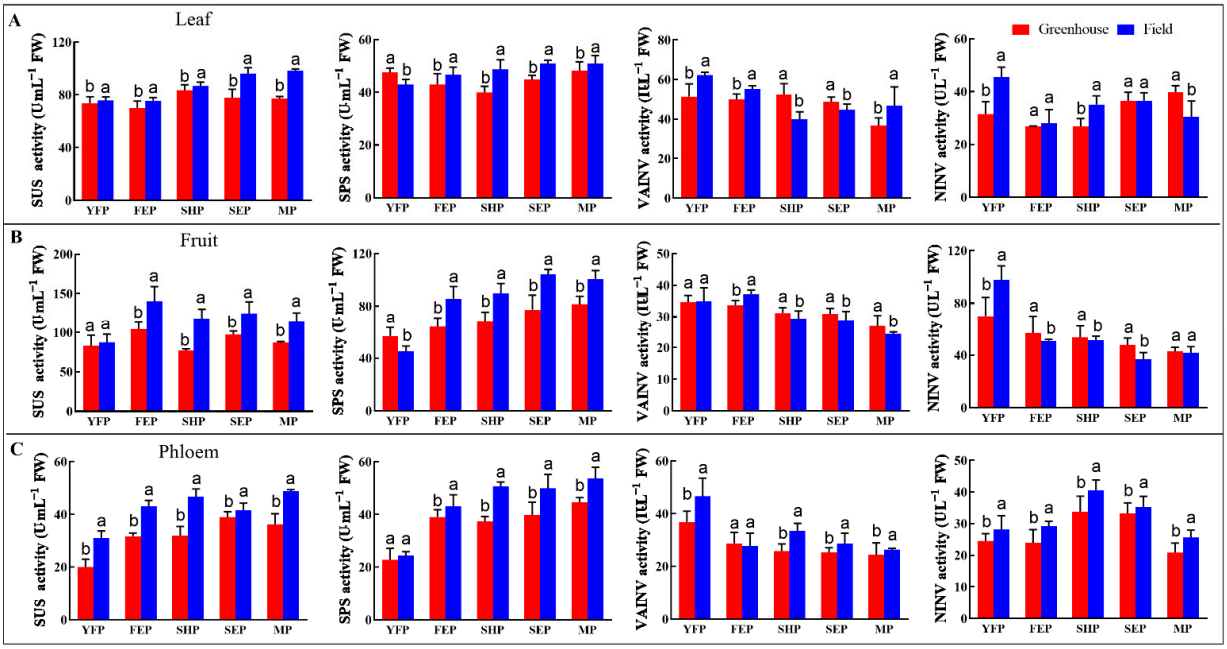

Figure 4. Activities of enzymes related to sucrose metabolism in different parts of peach trees grown in the field and greenhouse. (**A**–**C**) show the activities of sucrose metabolic enzymes in leaves, fruits and phloem, respectively. These enzymes include SUS, SPS, VAINV and NINV. The data are presented as the means ± SDs from three biological replicates. Different lowercase letters indicate significant differences ($p < 0.05$).

3.4.2. Activities of Key Enzymes Involved in Sorbitol Metabolism

Regarding sorbitol metabolism, sorbitol-6-phosphate dehydrogenase (S6PDH) and sorbitol dehydrogenase (SDH) activities were mainly analyzed. Figure 5A shows that the S6PDH activity in leaves from trees grown in the field was significantly greater than that detected in plants grown in the greenhouse during the second expansion and mature periods, whereas the SDH activity showed the opposite trend. SDH activity in fruit from plants grown in the greenhouse was higher than that of plants grown in the field in the early stage of fruit development, whereas SDH activity in fruit from plants grown in the field was significantly higher than that in the greenhouse in the stages of fruit development up to maturity (Figure 5B). The trends for changes in SDH activity in phloem in plants grown in the field and greenhouse first decreased and then increased during the maturation period, and the SDH activity was higher in fruits from plants grown in the field than in those grown in the greenhouse during the fruit development period (Figure 5C).

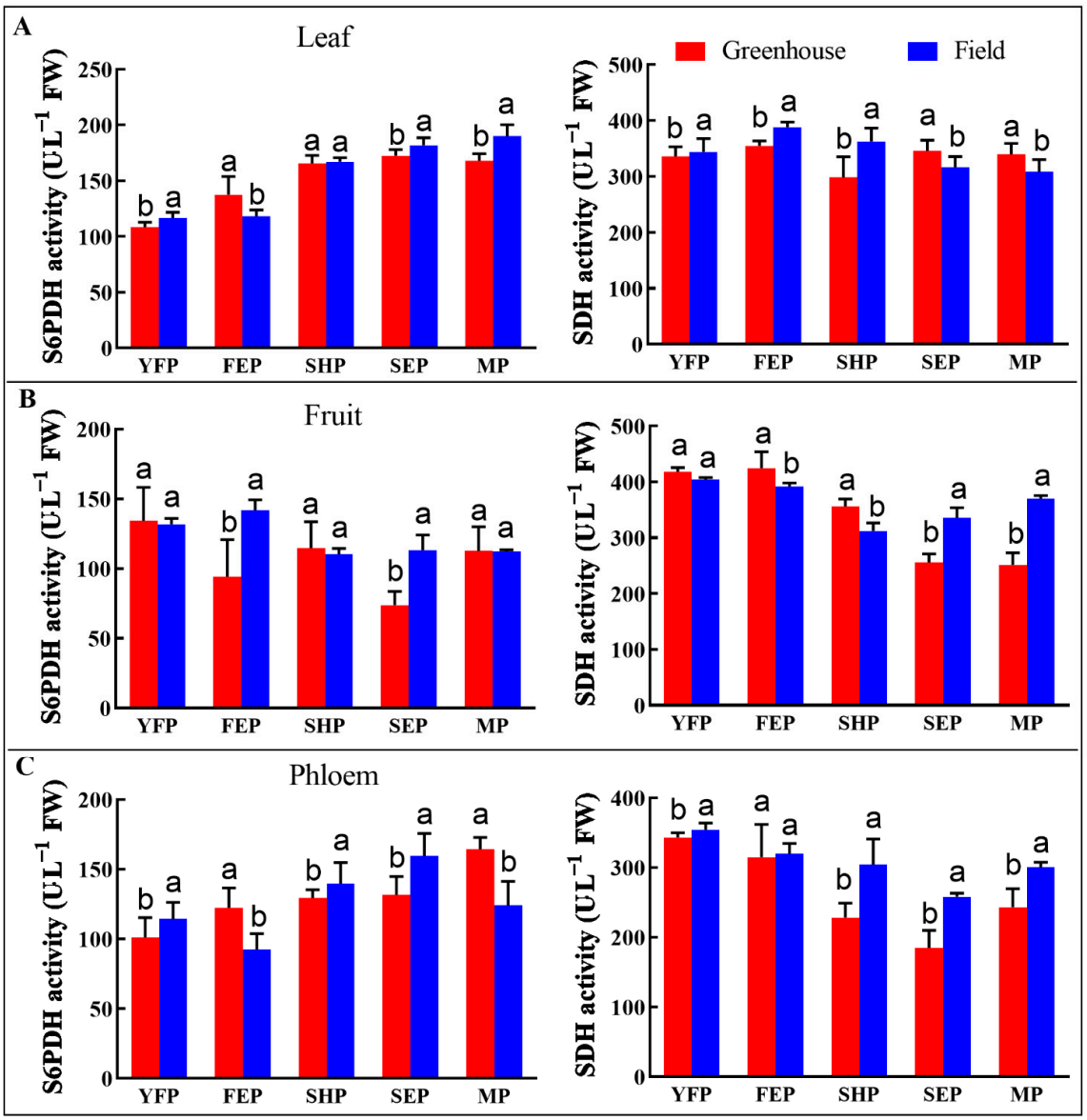

Figure 5. Activities of sorbitol metabolic enzymes in different parts of peach trees grown in the field and greenhouse. (**A**–**C**) show the activities of sorbitol metabolic enzymes in leaves, fruits and phloem, respectively. These enzymes include S6PDH and SDH. The data are presented as the means ± SDs from three biological replicates. Different lowercase letters indicate significance ($p < 0.05$).

3.4.3. Activities of Key Enzymes Involved in Hexose Metabolism

The activities of hexokinase (HK) and fructose kinase (FK) were mainly analyzed to assess hexose metabolism. Figure 6A shows significantly greater FK activity in leaves obtained from trees grown in the field environment than those obtained from plants grown in the greenhouse during the whole fruit development period. HK activity was always higher in plants grown in the greenhouse than in the field in the periods of fruit development to maturity. The FK activity observed in plants grown in the greenhouse was significantly higher than that in the field, except during the second expansion period (Figure 6B). HK and FK activities in phloem were lower than those in leaves and fruits, and steadily changed (Figure 6C).

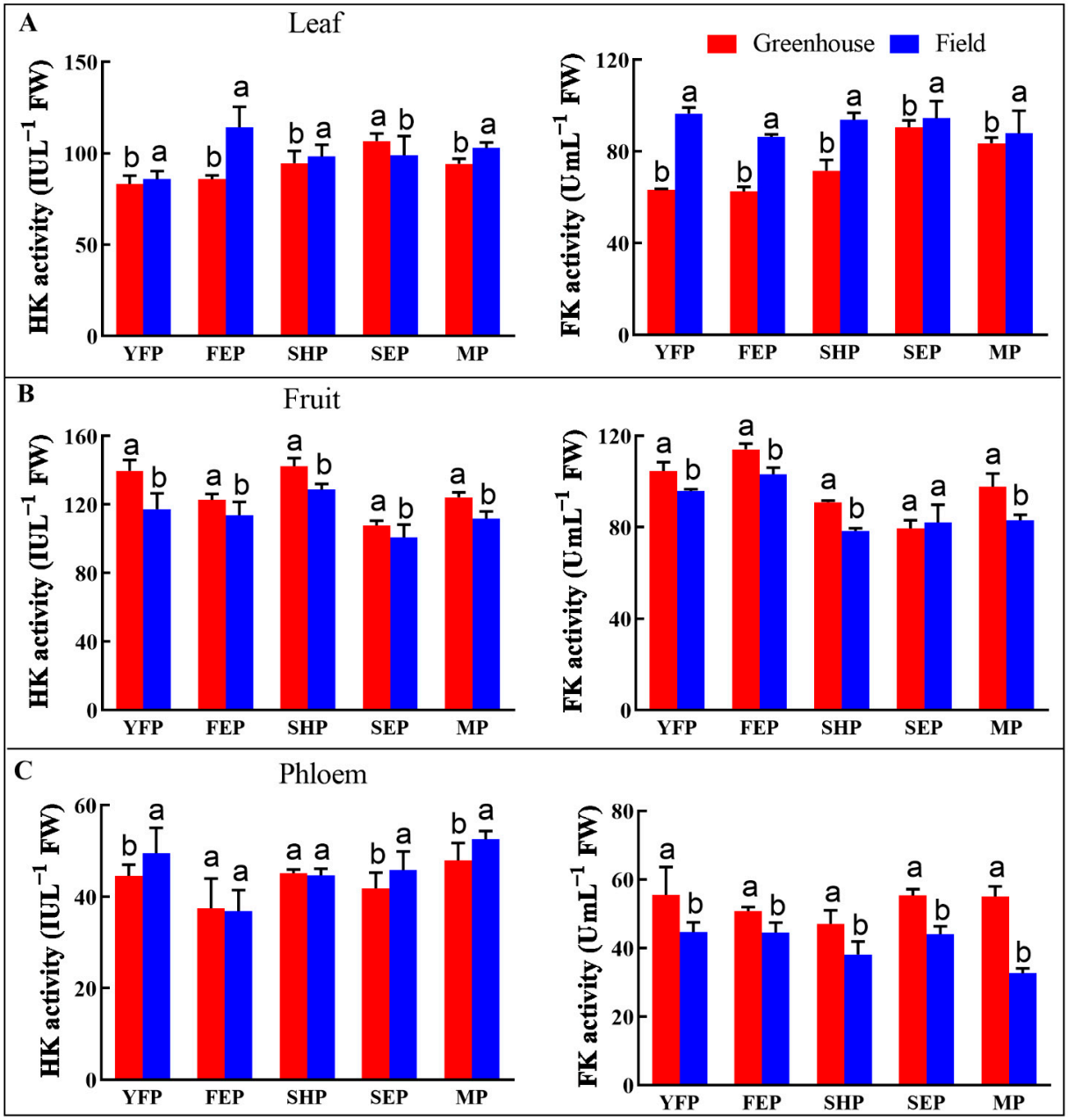

Figure 6. Activities of hexose metabolic enzymes in different parts of peach trees grown in the field and greenhouse. (**A**–**C**) show the activities of hexose metabolic enzymes in leaves, fruits and phloem, respectively. These enzymes include HK and FK. The data are presented as the means ± SDs from three biological replicates. Different lowercase letters indicate significant differences ($p < 0.05$).

3.5. Expression Patterns of Genes Encoding Key Enzymes Involved in Sugar Metabolism

3.5.1. Expression Patterns of Genes Encoding Key Enzymes Involved in Sucrose Metabolism

In this study, the expression of some genes encoding key enzymes in the sucrose metabolism pathway was analyzed. The relative expression levels of *PpSUS4*, *PpSUS5* and *PpSUS6* in leaves from trees cultivated in the field environment were significantly higher than those in the trees grown in the greenhouse at the later stage of fruit development (Figure 7A). In addition, *PpSPS1* expression levels in leaves suddenly increased during the second expansion stage and then decreased during the mature period, and the expression level in the field environment was significantly higher than that in the greenhouse (Figure 7B). The opposite expression patterns were noted for the two *PpVAINVs*. *PpVAINV1* expression gradually decreased, whereas *PpVAIN2* expression gradually increased (Figure 7C). Although the expression levels of *PpSUSs* in fruits from trees grown in the two environments continued to decrease, the expression level detected in most periods in the greenhouse environment was higher than that noted in the field environment (Figure 8A). *PpSPS2* expression levels continued to increase, and *PpSPS3* expression showed the opposite trend (Figure 8B). In addition, with fruit development, the expression levels of *PpVAINs* and *PpNINVs* in plants cultivated in the two environments decreased as a whole, and the expression levels of *PpVAIN1*, *PpVAIN2*, *PpNINV4* and *PpNINV7* reached their lowest levels during fruit ripening (Figure 8C). Interestingly, *PpNINV3* expression levels in the phloem of plants grown in the greenhouse were increased compared with those obtained in the field, whereas *PpNINV4* expression levels were increased in the phloem of plants grown in the field compared with those grown in the greenhouse (Figure 9C).

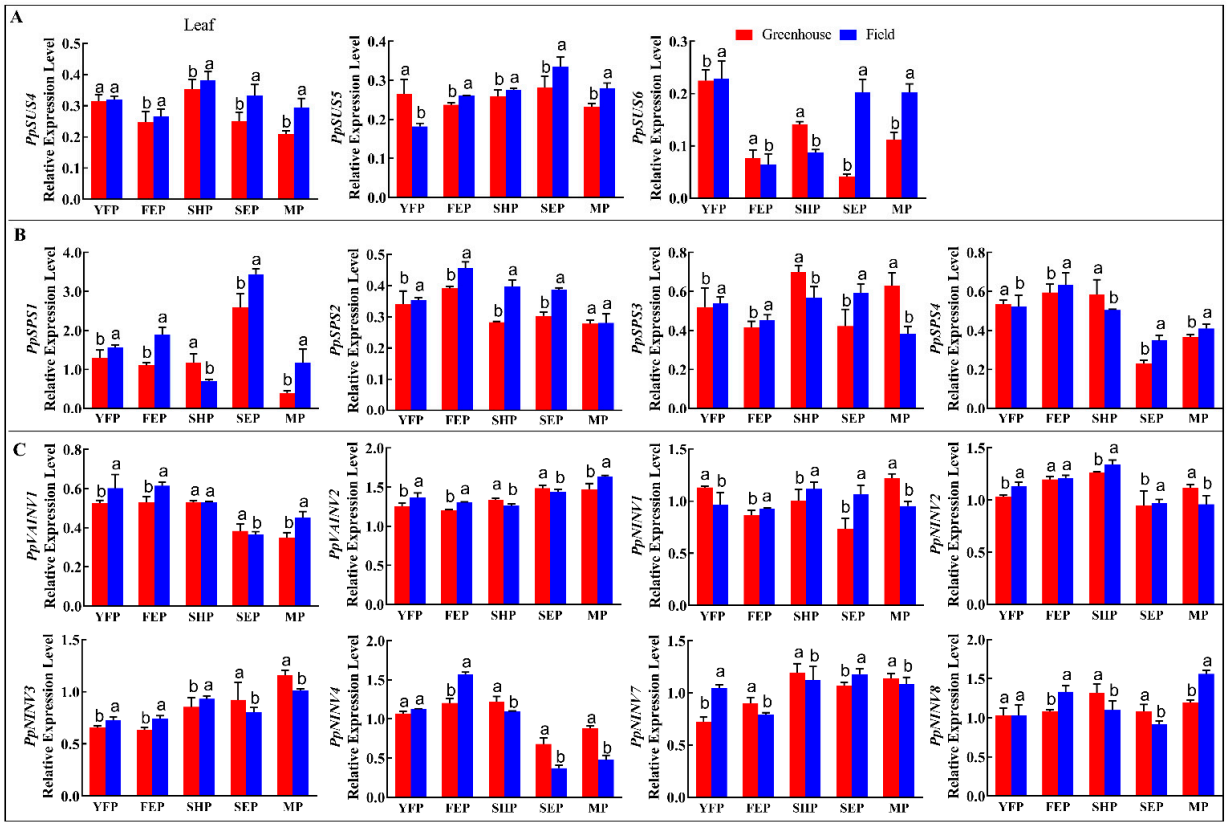

Figure 7. Expression levels of sucrose metabolism-related genes in leaves of plants grown in field and greenhouse environments. (**A**–**C**) show the expression levels of SUS, SPS and INV-related genes, respectively. The data are presented as the means ± SDs from three biological replicates. Different lowercase letters indicate significant differences ($p < 0.05$).

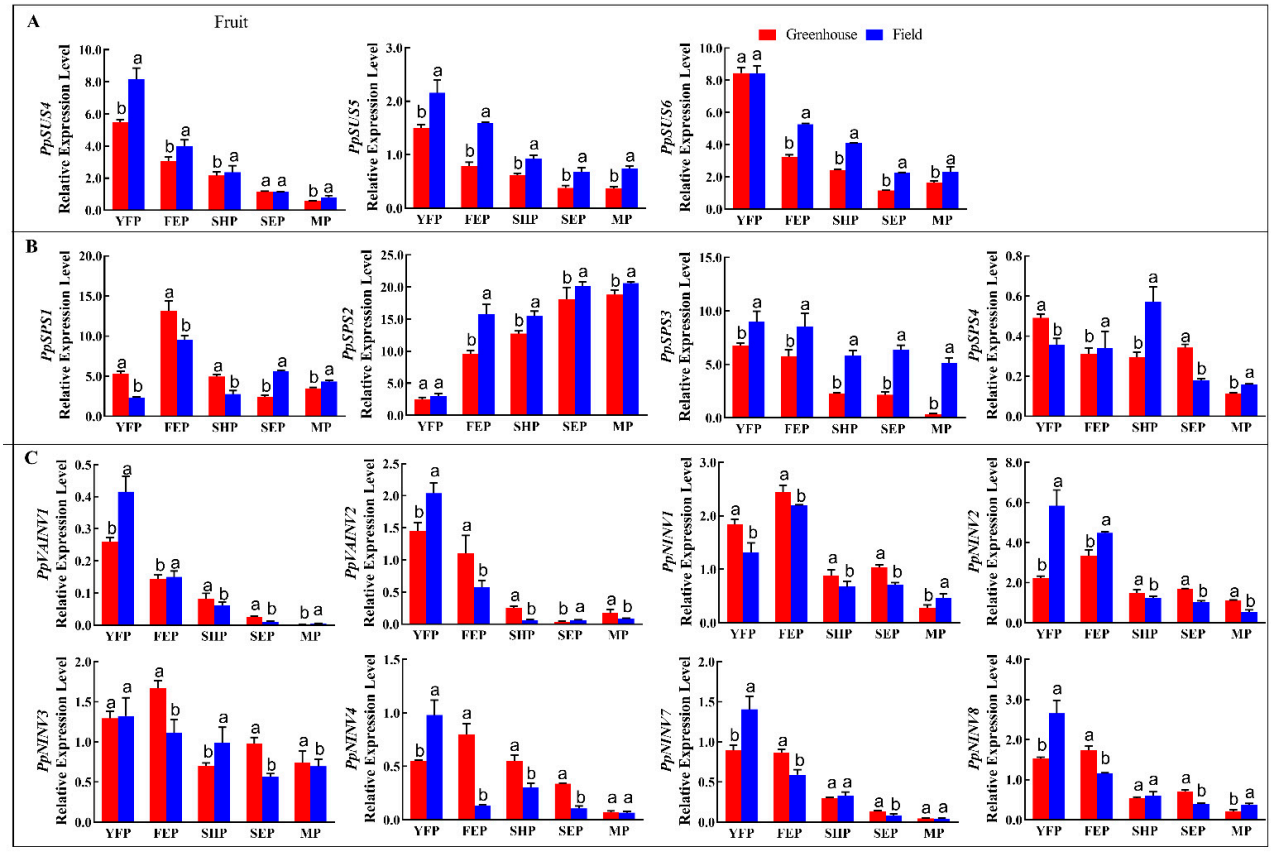

Figure 8. Expression levels of sucrose metabolism-related genes in fruits from plants grown in field and greenhouse environments. (**A**–**C**) show the expression levels of SUS, SPS and INV-related genes, respectively. The data are presented as the means ± SDs from three biological replicates. Different lowercase letters indicate significant differences ($p < 0.05$).

3.5.2. Expression Patterns of Genes Encoding Key Enzymes Involved in Sorbitol Metabolism

In the sorbitol metabolism pathway, *PpSDH1* expression levels in leaves were significantly higher in plants grown in the greenhouse than in those grown in the field, except at the young fruit stage. In leaves, *PpSDH2* and *PpSDH3* expression levels were similar throughout the whole fruit development stage. In the greenhouse environment, *PpSDH2* and *PpSDH3* levels in leaves initially increased and subsequently decreased. The trend for changes in the field environment was classified as the 'M' type, and the relative expression levels of *PpSDH2* and *PpSDH3* in leaves from plants cultivated in the greenhouse environment were significantly higher than those in plants cultivated in the field at maturity. *PpS6PDH* expression levels in leaves of plants grown in the field environment were increased compared with those in the greenhouse at the late stage of fruit development (Figure 10A). In the later stages of fruit development, the expression levels of *PpSDHs* in field fruit were higher than those noted in greenhouse fruit, and the expression level of *PpS6PDH* in fruit was lower than that in other parts (Figure 10B). The expression level of *PpSDHs* in the phloem of plants grown in the field environment was greater than that in the greenhouse phloem, and the difference was significant in the young fruit period (Figure 10C).

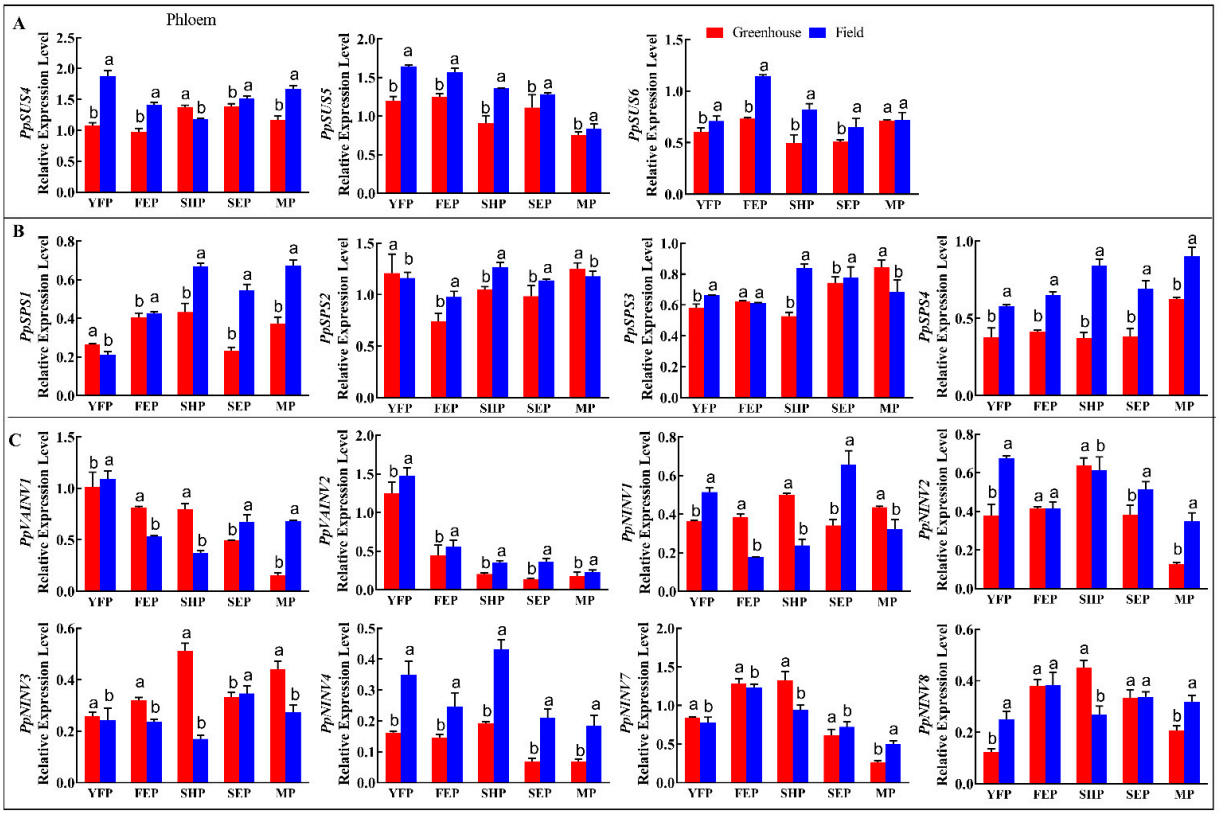

Figure 9. Expression levels of sucrose metabolism-related genes in the phloem of plants grown in field and greenhouse environments. (**A**–**C**) show the expression levels of SUS, SPS and INV-related genes, respectively. The data are presented as the means ± SDs from three biological replicates. Different lowercase letters indicate significant differences ($p < 0.05$).

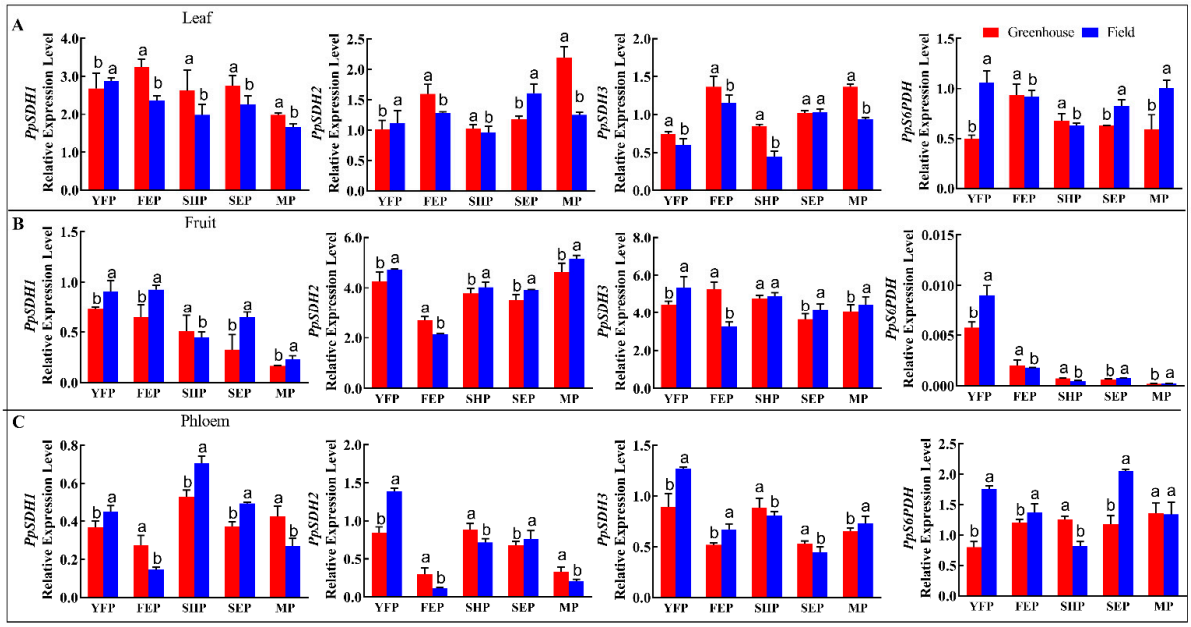

Figure 10. Expression levels of genes involved in sorbitol metabolism in different parts of peach trees grown in the field and greenhouse. (**A**–**C**) show the expression levels of sorbitol metabolism-related genes in leaves, fruits and phloem, respectively. The data are presented as the means ± SDs from three biological replicates. Different lowercase letters indicate significant differences ($p < 0.05$).

3.5.3. Expression Patterns of Genes Encoding Key Enzymes Involved in Hexose Metabolism

Figure 11 shows that the expression levels of *PpHKs* in leaves of plants grown in the greenhouse environment were increased compared with that in the field at most fruit tree development stages. The *PpFK3* expression levels in plants grown in the greenhouse were always significantly increased compared with those in plants grown in the field (Figure 11A). The expression level of *PpFKs* in fruit gradually increased and was stable in the late stage of fruit development, whereas the expression levels of *PpHK2, PpHK3* and *PpHK6* in the early stage of fruit development were higher than those in the later stage of fruit development (Figure 11B). The expression levels of *PpHKs* in greenhouse phloem were significantly higher than those in field phloem at the mature period (Figure 11C).

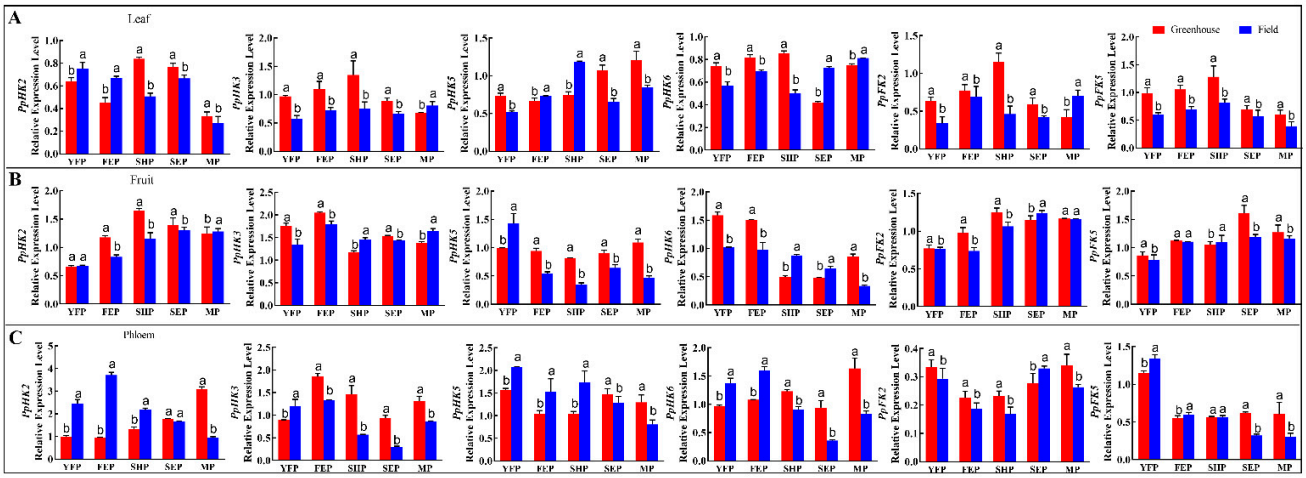

Figure 11. Expression levels of genes related to hexose metabolism in different parts of peach trees grown in the field and greenhouse. (**A**–**C**) show the expression levels of hexose metabolism-related genes in leaves, fruits and phloem, respectively. The data are presented as the means ± SDs from three biological replicates. Different lowercase letters indicate significant differences ($p < 0.05$).

4. Discussion

4.1. Differences between How Field and Greenhouse Environments Affect the Photosynthetic Capacity and Fruit Sugar Accumulation in Peach

The quality of fruit cultivated in greenhouses, particularly the intrinsic quality, is inferior to that of fruits obtained from trees grown in the field. A solution to this key problem in the cultivation and production of protected fruit trees is urgently needed. The contents and proportions of soluble sugars in fruit are the main factors determining the intrinsic quality, which originates from the transport of photosynthetic assimilates in leaves and is affected by the photosynthetic capacity of leaves. Greenhouse environmental conditions, such as temperature, humidity, light intensity and CO_2 concentration, are very different from those observed in the field. All of these factors affect leaf photosynthesis and photosynthetic assimilate metabolism. In this study, diurnal variations in environmental indices in the field and greenhouse were noted during the mature period. The trends for changes in various indices in the two environments were similar, but the specific values were significantly different because the fruit maturation period occurred in May in the greenhouse and July in the field. Temperature affects the activities of key enzymes regulating soluble sugars in the metabolic pathway of assimilates. A higher temperature within the suitable temperature range increases the activities of sucrose synthase (SUS) and vacuolar acid invertase (VAINV) in fruit, decomposes sucrose in fruit, and promotes the continuous transportation of sucrose in leaves to fruit sinks [26,27]. The soil and air temperatures in the field were higher than those in the greenhouse (Figure 1A), indicating that the plants in the field had a higher metabolic capacity of assimilates than those in the greenhouse. In addition, the light intensity in the field environment was also greater

than that noted in the greenhouse (Figure 1F). Previous studies have found that fruits from high light intensity areas tend to have stronger photosynthetic activity. Thus, the sugar/organic acid ratio is higher, and the flavor and sweetness are better [28]. As a raw material, environmental CO_2 is closely related to photosynthesis. In the dark period, CO_2 concentrations were higher in the greenhouse, and the concentration of CO_2 in the greenhouse and field changed with increasing light intensity (Figure 1E). Previous studies have suggested that increasing the CO_2 concentration may increase the soluble sugar content in plant tissue [29].

The dynamic changes in environmental conditions are closely related to photosynthesis in plants. The Pn, Gs and Tr in the field environment were significantly increased compared with those noted in the greenhouse (Figure 2), indicating that the leaves had stronger light energy capture ability and assimilate synthesis ability, which was beneficial to the accumulation, transport and distribution of photosynthetic assimilates [30]. In summary, the difference between the greenhouse and field environments mainly affected photosynthesis in peach trees grown under the two environmental conditions, which subsequently affected sugar accumulation and the metabolic properties of the fruit.

4.2. The Metabolic Characteristics of Assimilates in Different Environments Affect the Intrinsic Quality of Peach Fruit

The intrinsic quality of fruit is mainly determined by the contents and proportions of soluble sugars, which are regulated by various factors, such as genetic properties and natural environmental and cultivation measures. These factors do not exist in isolation but interact with each other to form a complex metabolic regulatory network that affects enzymatic activities, genetic changes, sugar content and sugar metabolism [3,10]. The accumulation of sugars in fruit is closely related to leaf photosynthesis and phloem transport. Sucrose, fructose, glucose and sorbitol accumulate in peach fruit, leaves and phloem [31]. Therefore, in this study, the dynamic changes in sugar components in peach leaves, phloem and fruit were determined to analyze the differences in soluble sugar contents between field and greenhouse peach fruits, and the possible causes were also assessed.

Sucrose and sorbitol are the main carbohydrates synthesized in source leaves and are transported over long distances to fruit sinks in a phloem-loaded manner, where they are metabolized and accumulate [32]. In this study, no significant difference in the sucrose content in leaves was noted between field and greenhouse environments, and a decreasing trend was noted at the fruit ripening stage. Interestingly, sucrose phosphate synthase (SPS) and SUS activities and the expression levels of *PpSS4*, *PpSS5*, *PpSS6*, *PpSPS1* and *PpSPS2* in the leaves of plants grown in the field environment were increased compared with those obtained in the greenhouse (Figures 3A, 4A and 7A,B). This finding indicates that the sucrose content synthesized in the leaves under the field environment should be greater than that obtained in the greenhouse. However, why was no significant difference in the indigenous experimental results noted? By comparing the sucrose contents in fruit and phloem, as well as the activities of key enzymes involved in sucrose synthesis, we hypothesized that most of the sucrose synthesized by leaves was transported to fruit through phloem in the field environment, whereas less of the sucrose synthesized by leaves in plants grown in the greenhouse was transported to fruit (Figure 3A,B and Figure 4B). VAINV and neutral invertase (NINV) irreversibly decompose sucrose into glucose and fructose [33]. The sucrose content in leaves gradually decreased after reaching a peak during the stone hardening period, and the sucrose content in fruit gradually increased with the development of fruit (Figure 3A,B). These findings were consistent with the findings of previous studies [34]. The sucrose content in field fruit was significantly higher than that in greenhouse fruit, suggesting the possibility of increased sugar accumulation in field fruit (Figure 3B). In addition, VAINV and NINV activities in fruit continuously decreased (Figure 4B), and the expression levels of *PpVAINV1*, *PpVAINV2*, *PpNINV2*, *PpNINV7* and *PpNINV8* were also very low in the later stages of fruit development (Figure 8C), which was also an important reason for the continuous increase in fruit sucrose content [35].

Sorbitol accounts for approximately 60–70% of the photosynthetic products produced in source leaves, consistent with the results of this study. The sorbitol content, sorbitol-6-phosphate dehydrogenase (S6PDH) activity and *PpS6PDH* expression level were increased in leaves compared with fruit and phloem (Figures 3A, 5A and 10A). Subsequently, we compared the sorbitol content in leaves from both the field and the greenhouse environments. After the stone hardening period, the sorbitol content in leaves from plants grown in the field environment was increased compared with that in the greenhouse, whereas the sorbitol content in fruits from trees cultivated in the greenhouse was significantly increased compared with that in the field (Figure 3A,B). We hypothesized that the sorbitol synthesized in leaves was transported to fruits in large quantities through the phloem in the greenhouse environment, and thus, the amount of sorbitol retained in leaves in the greenhouse environment was low. In addition, this experiment also revealed similar sucrose and sorbitol levels in leaves, ranging from 14 $mg \cdot g^{-1}$ FW to 25 $mg \cdot g^{-1}$ FW during the whole fruit development period. However, in the fruit, the sucrose content increased (38–70 $mg \cdot g^{-1}$ FW) and the sorbitol content decreased (3–5 $mg \cdot g^{-1}$ FW) (Figure 3A,B). Specifically, less sucrose is decomposed when transported to the fruit, whereas most of the sorbitol transported to the fruit is decomposed. We hypothesized that the glucose and fructose in the fruit may be mainly converted from sorbitol. In addition, significantly less sorbitol was detected in the field fruit than in the greenhouse fruit, and the activities of sorbitol oxidase (SOX) (Supplementary Figure S2) and sorbitol dehydrogenase (SDH) and the expression levels of *PpSDH1*, *PpSDH2* and *PpSDH3* were also increased in plants grown in the field compared with those grown in the greenhouse at the later stage of fruit development (Figures 3B, 5B and 10B). Based on these results, more sorbitol is transported to the cytoplasm and is decomposed into glucose and fructose in field fruit compared with greenhouse fruit [36–38], and the sucrose content in field fruit was also significantly higher than that in greenhouse fruit. This difference may explain why the intrinsic quality of the fruit from trees grown in the field is better than that of fruit from trees grown in the greenhouse. By comprehensively analyzing the sucrose content, sorbitol content, related metabolic enzyme activities and coding gene expression in four parts of the plant, we hypothesize that glucose in peach fruit is mainly decomposed from sucrose and sorbitol, and fructose is mainly decomposed from sorbitol [35].

Sugar is the essential component contributing to the edible quality of fruit and affects consumers' satisfaction regarding sweetness [39]. The flavor of peach fruit is related to the content and proportion of various sugar components. Sucrose is the main sugar present in mature peach fruits, followed by fructose and glucose, and sorbitol is present at the lowest levels [40,41]. Of these sugars, fructose is the sweetest, as it is approximately 1.7 times as sweet as sucrose [42]. Breeding these varieties is also one of the goals of breeders [21]. In the present study, the highest contents of glucose and fructose were detected in the fruit compared with other parts, and gradually decreased as the fruit matured. This finding differs from the trend for sucrose content (Figure 3B). The contents of glucose and fructose in field peach fruit were significantly higher than those in greenhouse fruit throughout the fruit development period, especially in the later stages of fruit development (Figure 3B). More importantly, the content of fructose in field fruit was significantly higher than that of glucose, whereas the contents of fructose and glucose were approximately the same in greenhouse fruit. The possible explanation for this difference in sugar content was that the higher activity of hexokinase (HK) and fructose kinase (FK), and higher expression of *PpHK2*, *PpHK5*, and *PpFK5* in greenhouse peach fruit, accelerated the phosphorylation of glucose and fructose (Figures 6B and 11B) [43]. Therefore, we hypothesize that the relatively high contents of soluble sugars, such as sucrose, glucose and fructose, particularly fructose, may be one of the main reasons why the fruit flavor and intrinsic quality of fruits obtained from plants grown in the field environment are better than those of plants grown in the greenhouse environment.

5. Conclusions

The field environment from May to July was more beneficial to photosynthesis and fruit sugar accumulation in peach trees than the greenhouse environment from March to May. The photosynthetic compounds synthesized in the leaves in the field were present at higher levels than those in the greenhouse, enabling sufficient assimilates to be retained or transported out of the leaves. The sucrose, glucose and fructose levels in the field fruits were higher than those in the greenhouse fruits. The sucrose and sorbitol present in the fruit were mainly derived from the transport of photosynthetic assimilates in leaves, and glucose and fructose in the fruit were mainly derived from the decomposition and conversion of sorbitol. The main reason why the intrinsic quality of field fruit is better than that of greenhouse fruit is that more sorbitol in field fruit is converted into glucose and fructose than in greenhouse fruit. Notably, the significant increase in fructose content increases the sweetness and flavor of field fruit (Figure 12).

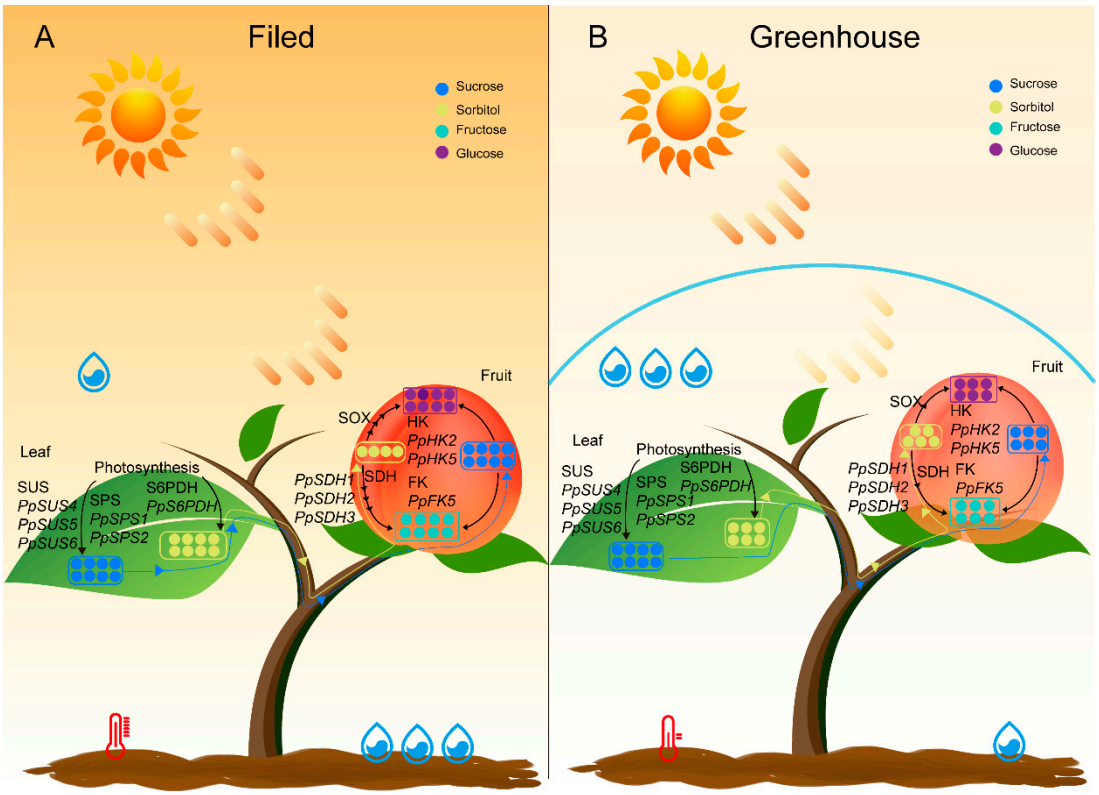

Figure 12. Proposed model for the differences in sugar transport in peaches from trees grown in field and greenhouse environments. (**A**) presents the field environment, and (**B**) presents the greenhouse environment, where the blue curve represents the greenhouse film. For environmental indicators, the deeper the background color, the higher the temperature and light intensity. The more water droplets that are present, the greater the humidity. The greater the thermometer index, the higher the temperature. The blue ball represents sucrose, the yellow ball represents sorbitol, the cyan ball represents fructose, and the purple ball represents glucose. The greater the number of balls, the greater the content. The greater the number of arrows, the greater the transport or decomposition.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/agronomy12112877/s1, Figure S1: Fruit development curve; Figure S2: SOX activity in different parts of peach grown in the field and greenhouse. (A–C) show the SOX activities in leaves, fruits and phloem, respectively. The data are presented as the means ± SDs from three biological replicates. Different lowercase letters indicate significant differences ($p < 0.05$); Table S1: Primers used for qRT-PCR.

Author Contributions: G.X. performed most of the experiments, analyzed the data, and compiled the original manuscript. C.L. assisted with the qRT-PCR experiments. D.L. designed the experiments and helmed the project. W.X., X.F., S.Q. and X.C. contributed suggestions and discussion. L.L. and D.L. designed the experiments and reviewed and edited the manuscript. All authors have read and agreed to the published version of the manuscript.

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References

1. Zhang, C.; Hao, Y.-J. Advances in genomic, transcriptomic, and metabolomic analyses of fruit quality in fruit Crops. *Hortic. Plant J.* **2020**, *6*, 361–371. [CrossRef]
2. Génard, M.; Gibert, C.; Bruchou, C.; Lescourret, F. An intelligent virtual fruit model focussing on quality attributes. *J. Hortic. Sci. Biotechnol.* **2009**, *84*, 157–163. [CrossRef]
3. Lopresti, J.; Goodwin, I.; McGlasson, B.; Holford, P.; Golding, J. Variability in size and soluble solids concentration in peaches and nectarines. *Hortic. Rev.* **2014**, *42*, 253–312.
4. Minas, I.S.; Tanou, G.; Molassiotis, A. Environmental and orchard bases of peach fruit quality. *Sci. Hortic.* **2018**, *235*, 307–322. [CrossRef]
5. Karagiannis, E.; Tanou, G.; Samiotaki, M.; Michailidis, M.; Diamantidis, G.; Minas, I.S.; Molassiotis, A. Comparative physiological and proteomic analysis reveal distinct regulation of peach skin quality traits by altitude. Front. *Plant Sci.* **2016**, *7*, 1689.
6. Lopez, G.; Behboudian, M.H.; Vallverdu, X.; Mata, M.; Girona, J.; Marsal, J. Mitigation of severe water stress by fruit thinning in 'o'henry' peach: Implications for fruit quality. *Sci. Hortic.* **2010**, *125*, 294–300. [CrossRef]
7. Sikhandakasmita, P.; Kataoka, I.; Mochioka, R.; Mochioka, R.; Beppu, K. Impact of temperatures during fruit development on fruit growth rate and qualities of 'KU-PP2' Peach. *Hortic. J.* **2022**, *91*, 152–156. [CrossRef]
8. Anthony, B.M.; Minas, I.S. Optimizing peach tree canopy architecture for efficient light use, increased productivity and improved fruit quality. *Agronomy* **2021**, *11*, 1961. [CrossRef]
9. Anthony, B.M.; Chaparro, J.M.; Sterle, D.G.; Prenni, J.E.; Minas, I.S. Metabolic signatures of the true physiological impact of canopy light environment on peach fruit quality. *Environ. Exp. Bot.* **2021**, *191*, 104630. [CrossRef]
10. Vimolmangkang, S.; Zheng, H.; Peng, Q.; Jiang, Q.; Wang, H.; Fang, T.; Liao, L.; Wang, L.; He, H.; Han, Y. Assessment of sugar components and genes involved in the regulation of sucrose accumulation in peach fruit. *J. Agric. Food Chem.* **2016**, *64*, 6723–6729. [CrossRef]
11. Front 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]
12. Wang, T.; Wu, G.; Chen, J.; Cui, P.; Chen, Z.; Yan, Y.; Zhang, Y.; Li, M.; Niu, D.; Li, B.; et al. Integration of solar technology to modern greenhouse in china: Current status, challenges and prospect. *Renew. Sustain. Energy Rev.* **2017**, *70*, 1178–1188. [CrossRef]
13. Layne, D.R.; Wang, Z.; Liang, N. Protected cultivation of peach and nectarine in china industry observations and assessments. *J. Am. Pomol. Soc.* **2013**, *67*, 18–28.
14. Martínez-Gómez, P.; Rahimi Devin, S.; Salazar, J.A.; López-Alcolea, J.; Rubio, M.; Martínez-García, P.J. Principles and prospects of prunus cultivation in greenhouse. *Agronomy* **2021**, *11*, 474. [CrossRef]
15. Tewolde, F.T.; Lu, N.; Shiina, K.; Maruo, T.; Takagaki, M.; Kozai, T.; Yamori, W. Nighttime supplemental LED inter-lighting improves growth and yield of single-truss tomatoes by enhancing photosynthesis in both winter and summer. *Front. Plant Sci.* **2016**, *7*, 448. [CrossRef]
16. Xi, W.; Zhang, Q.; Lu, X.; Wei, C.; Yu, S.; Zhou, Z. Improvement of flavour quality and consumer acceptance during postharvest ripening in greenhouse peaches by carbon dioxide enrichment. *Food Chem.* **2014**, *164*, 219–227. [CrossRef]
17. Yu, X.; Ali, M.M.; Li, B.; Fang, T.; Chen, F. Transcriptome data-based identification of candidate genes involved in metabolism and accumulation of soluble sugars during fruit development in 'Huangguan'plum. *J. Food Biochem.* **2021**, *45*, e13878. [CrossRef]
18. Yang, J.; Zhang, J.; Niu, X.-Q.; Zheng, X.L.; Chen, X.; Zheng, G.-H.; Wu, J.-C. Comparative transcriptome analysis reveals key genes potentially related to organic acid and sugar accumulation in loquat. *PLoS ONE* **2021**, *16*, e0238873. [CrossRef]
19. Kelley, K.M.; Primrose, R.; Crassweller, R.; Hayes, J.E.; Marini, R. Consumer peach preferences and purchasing behavior: A mixed methods study. *J. Sci. Food Agric.* **2016**, *96*, 2451–2461. [CrossRef]
20. Teo, G.; Suzuki, Y.; Uratsu, S.L.; Lampinen, B.; Ormonde, N.; Hu, W.K.; DeJong, T.M.; Dandekar, A.M. Silencing leaf sorbitol synthesis alters long-distance partitioning and apple fruit quality. *Proc. Natl. Acad. Sci. USA* **2006**, *103*, 18842–18847. [CrossRef]
21. Cirilli, M.; Bassi, D.; Ciacciulli, A. Sugars in peach fruit: A breeding perspective. *Hortic. Res.* **2016**, *3*, 15067. [CrossRef] [PubMed]

22. Wang, X.; Fu, X.; Chen, M.; Huan, L.; Liu, W.; Qi, Y.; Gao, Y.; Xiao, W.; Chen, X.; Li, L.; et al. Ultraviolet B irradiation influences the fruit quality and sucrose metabolism of peach (*Prunus persica* L.). *Environ. Exp. Bot.* **2018**, *153*, 286–301. [CrossRef]
23. Falchi, R.; Bonghi, C.; Drincovich, M.F.; Famiani, F.; Lara, M.V.; Walker, R.P.; Vizzotto, G. Sugar metabolism in stone fruit: Source-sink relationships and environmental and agronomical effects. *Front. Plant Sci.* **2020**, *11*, 573982. [CrossRef]
24. Yang, G.; Xu, H.; Zou, Q.; Zhang, J.; Jiang, S.; Fang, H.; Wang, Y.; Su, M.; Wang, N.; Chen, X. The vacuolar membrane sucrose transporter MdSWEET16 plays essential roles in the cold tolerance of apple. *Plant Cell Tissue Organ Cult. (PCTOC)* **2020**, *140*, 129–142. [CrossRef]
25. Tong, Z.; Gao, Z.; Fei, W.; Zhou, J.; Zhen, Z. Selection of reliable reference genesfor gene expression studies in peach using real-time PCR. *BMC Mol. Biol.* **2009**, *10*, 1–13. [CrossRef]
26. Rolland, F.; Baena-Gonzalez, E.; Sheen, J. Sugar sensing and signaling in plants: Conserved and novel mechanisms. *Annu. Rev. Plant Biol.* **2006**, *57*, 675–709. [CrossRef]
27. Moore, C.E.; Meacham-Hensold, K.; Lemonnier, P.; Slattery, R.A.; Benjamin, C.; Bernacchi, C.J.; Lawson, T.; Cavanagh, A.P. The effect of increasing temperature on crop photosynthesis: From enzymes to ecosystems. *J. Exp. Bot.* **2021**, *72*, 2822–2844. [CrossRef]
28. Mikulic-Petkovsek, M.; Schmitzer, V.; Slatnar, A.; Stampar, F.; Veberic, R. A comparison of fruit quality parameters of wild bilberry (*Vaccinium myrtillus* L.) growing at different locations. *J. Sci. Food Agric.* **2015**, *95*, 776–785. [CrossRef]
29. Dong, J.; Gruda, N.; Lam, S.K.; Li, X.; Duan, Z. Effects of elevated CO_2 on nutritional quality of vegetables: A Review. *Front. Plant Sci.* **2018**, *9*, 924. [CrossRef]
30. Kaiser, E.; Morales, A.; Harbinson, J.; Kromdijk, J.; Heuvelink, E.; Marcelis, L.F. Dynamic photosynthesis in different environmental conditions. *J. Exp. Bot.* **2015**, *66*, 2415–2426. [CrossRef]
31. Ludewig, F.; Flügge, U.-I. Role of metabolite transporters in source-sink carbon allocation. *Front. Plant Sci.* **2013**, *4*, 231. [CrossRef] [PubMed]
32. Moing, A.; Carbonne, F.; Zipperlin, B.; Svanella, L.; Gaudille're, J.P. Phloem loading in peach: Symplastic or apoplastic? *Physiol. Plant.* **1997**, *101*, 489–496. [CrossRef]
33. Moriguchi, T.; Yamaki, S. Purification and characterization of sucrose synthase from peach (*Prunus persica*) fruit. *Plant Cell Physiol.* **1988**, *29*, 1361–1366.
34. Aslam, M.M.; Deng, L.; Wang, X.B.; Wang, Y.; Pan, L.; Liu, H.; Niu, L.; Lu, Z.H.; Cui, G.C.; Zeng, W.F.; et al. Expression patterns of genes involved in sugar metabolism and accumulation during peach fruit development and ripening. *Sci. Hortic.* **2019**, *257*, 108633. [CrossRef]
35. Li, M.; Feng, F.; Cheng, L. Expression patterns of genes involved in sugar metabolism and accumulation during apple fruit development. *PLoS ONE* **2012**, *7*, e33055. [CrossRef]
36. Zhou, R.; Cheng, L.; Dandekar, A.M. Down-regulation of sorbitol dehydrogenase and up-regulation of sucrose synthase in shoot tips of the transgenic apple trees with decreased sorbitol synthesis. *J. Exp. Bot.* **2006**, *57*, 3647–3657. [CrossRef]
37. Li, M.; Li, D.; Feng, F.; Zhang, S.; Ma, F.; Cheng, L. Proteomic analysis reveals dynamic regulation of fruit development and sugar and acid accumulation in apple. *J. Exp. Bot.* **2016**, *67*, 5145–5157. [CrossRef]
38. Yang, J.; Zhu, L.; Cui, W.; Zhang, C.; Li, D.; Ma, B.; Cheng, L.; Ruan, Y.L.; Ma, F.; Li, M. Increased activity of MdFRK2, a high-affinity fructokinase, leads to upregulation of sorbitol metabolism and downregulation of sucrose metabolism in apple leaves. *Hortic. Res.* **2018**, *5*, 71. [CrossRef]
39. Orazem, P.; Stampar, F.; Hudina, M. Quality analysis of 'redhaven' peach fruit grafted on 11 rootstocks of different genetic origin in a replant soil. *Food Chem.* **2011**, *124*, 1691–1698. [CrossRef]
40. Moriguchi, T.; Ishizawa, Y.; Sanada, T. Differences in sugar composition in prunus persica fruit and the classification by the principal component analysis. *J. Jpn. Soc. Hortic. Sci.* **1990**, *59*, 307–312. [CrossRef]
41. Brooks, S.J.; Moore, J.N.; Murphy, J.B. Quantitative and qualitative changes in sugar content of peach genotypes [*Prunus persica* (L.) Batsch.]. *J. Am. Soc. Hortic. Sci.* **1993**, *118*, 97–100. [CrossRef]
42. Kroger, M.; Meister, K.; Kava, R. Low-calorie sweeteners and other sugar substitutes: A review of the safety issues. *Compr. Rev. Food Sci. Food Saf.* **2006**, *5*, 35–47. [CrossRef]
43. Desnoues, E.; Gibon, Y.; Baldazzi, V.; Signoret, V.; Génard, M.; Quilot-Turion, B. Profiling sugar metabolism during fruit development in a peach progeny with different fructose-to-glucose ratios. *BMC Plant Biol.* **2014**, *14*, 336. [CrossRef] [PubMed]