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Optimizing Biochemical and Phytochemical Attributes in Peaches through Foliar Applications of Silicon and Zinc

Syed Tanveer Shah [1,2,†], Naseer Ahmad [1,†], Abdul Basit [3], Muhammad Sajid [2], Aftab Jamal [4,*], Muhammad Farhan Saeed [4], Waleed Iqbal [5], Mahmoud F. Seleiman [6], Emanuele Radicetti [7] and Roberto Mancinelli [8,*]

[1] Department of Horticulture, The University of Agriculture, Peshawar 25130, Pakistan; dr.syedtanveershah@hu.edu.pk (S.T.S.); na8297052@gmail.com (N.A.)

[2] Department of Agriculture, Faculty of Biological and Health Sciences, Hazara University, Mansehra 21120, Pakistan; drsajid_1@yahoo.com

[3] Floricultural Biotechnology Laboratory, Department of Horticultural Science, Kyungpook National University, Daegu 41566, Republic of Korea; abdulbasit97_lily@knu.ac.kr

[4] Department of Environmental Sciences, COMSATS University Islamabad, Vehari Campus, Vehari 61100, Pakistan; farhansaeed@cuivehari.edu.pk

[5] Department of Agronomy, Faculty of Crop Production Sciences, The University of Agriculture, Peshawar 25130, Pakistan; wiqbal@aup.edu.pk

[6] Department of Plant Production, College of Food and Agriculture Sciences, King Saud University, P.O. Box 2460, Riyadh 11451, Saudi Arabia; mseleiman@ksu.edu.sa

[7] Department of Chemical, Pharmaceutical and Agricultural Sciences (DOCPAS), University of Ferrara, 44121 Ferrara, Italy; rdcmnl@unife.it

[8] Department of Agricultural and Forestry Sciences, University of Tuscia, Via San Camillo de Lellis snc, 01100 Viterbo, Italy

* Correspondence: aftabses98@gmail.com (A.J.); mancinel@unitus.it (R.M.)

† These authors contributed equally to this work.

Abstract: Peach production faces significant pre-harvest challenges, including low moisture, nutrient deficiencies, flower drop, physical damage, and surface discoloration, which can limit yield and fruit quality. To mitigate these issues, the present study hypothesized that foliar applications of silicon and zinc could enhance peach growth, yield, and quality due to their known roles in improving stress tolerance, nutrient uptake, and antioxidant activity. Therefore, this research aimed to identify optimal concentrations of silicon and zinc for quality peach production. Ten-year-old peach trees of uniform size were sprayed with four levels of silicon (0%, 0.1%, 0.2%, and 0.3%) and zinc (0%, 0.25%, 0.50%, and 0.75%) for two consecutive growing seasons, at the berry and pit hardening stages, using a Randomized Complete Block Design (RCBD) with three replications. The averaged data from the two years showed that the pre-harvest foliar application of silicon significantly improved all yield and quality attributes of peaches. The foliar application of silicon at 0.3% notably enhanced fruit growth, yield, and biochemical attributes. Additionally, the highest fruit growth, yield, and quality of peach fruits were observed at the 0.75% zinc concentration. Maximum antioxidant activity, flavonoid content, proline content, and catalase activity were observed in fruits from plants treated with 0.3% silicon, which were statistically on par with 0.2% silicon. However, peroxidase activity was highest at 0.2% silicon. Regarding zinc levels, antioxidant activity, flavonoid content, proline content, and peroxidase activity were highest in fruits treated with 0.75% zinc, while catalase activity was superior when fruits were sprayed with 0.50% zinc. The interaction between silicon and zinc concentrations was found to be non-significant for most parameters, except for titratable acidity, TSS–acid ratio, ascorbic acid content, antioxidant activity, flavonoid content, and peroxidase activity. In conclusion, the foliar application of 0.3% silicon and 0.75% zinc independently enhanced all yield and quality characteristics of peaches. For the agro-climatic conditions of Peshawar, 0.2% silicon and 0.50% zinc are recommended for optimal peach production.

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1. Introduction

Peach (*Prunus persica* L.), a member of the Rosaceae family, is native to China, where it has been domesticated and cultivated for centuries [1]. Peaches have significant nutritional value, being rich in fiber, potassium, minerals (important during pregnancy), and antioxidants (which strengthen the immune system), with high contents of vitamins C and A. They are commonly used in salads and juices. Peach fruit can help prevent cancer, hypokalemia, high cholesterol, obesity, and neurological diseases [2]. In Pakistan, peaches are cultivated on an area of 15.29 thousand hectares, producing 110.76 thousand tons annually. The area under peach cultivation in Khyber Pakhtunkhwa is 10,081 hectares, yielding 69,417 tons, followed by Baluchistan, where the area and production are 4973 hectares and 39,457 tons, respectively. In Punjab, the area under peach cultivation is 241 hectares, with a production of 1890 tons [3].

Peaches are climacteric in nature and are considered among the most perishable fruits [4]. They ripen quickly, resulting in a short postharvest life. Low moisture and nutrient unavailability are key factors contributing to reduced yield and quality. These factors can negatively affect fruit production and quality. Issues such as pollen germination failure, non-functional flowers, and reproductive organ deficiencies lead to flower drop. Physical damage during harvesting and handling can cause surface discoloration and other problems in peach fruit. The quality of peaches is influenced by growth patterns, harvesting practices, and post-harvest factors, including chilling injury [5]. Due to their perishable nature, peaches cannot be stored for extended periods. Pre-harvest fertilizer applications, biopesticides, and micronutrient sprays are some techniques used to improve yield, maintain quality, and extend the post-harvest life to meet market demands [6]. The softening of peach flesh during transportation increases susceptibility to diseases, which can reduce the market value of the fruit [7].

Silicon is often an overlooked element with the potential to enhance fruit development, production, and quality. Being readily available, silicon constitutes about one-fourth (29%) of the Earth's surface [8]. It improves photosynthetic activity, increases the potassium–sodium ratio, promotes the action of specific enzymes, enhances the solubility of chemicals in the plant's xylem, and strengthens the plant's defense system [9]. It can be utilized as a fertilizer in agriculture, because it improves the yield, quality, and other attributes of fruit trees [10]. Silicon has been shown to have significant positive effects on plant development, growth, yield, insect and disease resistance, and tolerance to environmental stresses in various crops. The primary role of silicon in plant biology is to mitigate multiple stresses (both abiotic and biotic) by stimulating natural defenses, such as increasing active compounds like peroxidase, chitinase, flavonoid phytoalexins, and polyphenol oxidase. These compounds also help protect plants against various fungal diseases.

Zinc is a key nutrient involved in a wide range of enzymatic activities essential for optimal plant growth and development. It plays a crucial role in the synthesis of carbohydrates and proteins [11] and in gene expression [12]. In alkaline soils with high pH, zinc is available in very limited amounts. Zinc is also essential for the structure and function of about 10% of proteins in living systems [13]. Over three hundred enzymes require zinc as a cofactor [14]. In plant cells, zinc aids in the detoxification of reactive oxygen species, which are extensively produced during germination [15]. Zinc is vital for enhancing yield as it improves fruit quality, reduces fruit drop, and activates the plant's natural defenses [16]. Zinc foliar treatments have been shown to improve the chemical (TSS and vitamin C content), physical (yield and fruit juice), and sensory (taste) characteristics of fruit [16]. Considering the perishable nature of peaches and the postharvest losses caused by various biotic and abiotic stresses, as well as the often-underappreciated role of silicon in plant processes, its potential in mitigating these stresses, and the deficiency of zinc in high-pH soils, this study hypothesized that silicon would improve peach fruit development and stress resistance. Additionally, it was proposed that the optimal concentration of zinc would enhance fruit quality, and the interaction between silicon and zinc would have a synergistic effect on overall peach quality and yield. The objectives of this research were to (i) investigate the influence of silicon on peach fruit growth, yield, and quality; (ii) determine the optimal concentration of zinc for enhancing peach fruit production; and (iii) examine the interaction between silicon and zinc on the yield, biochemical, and phytochemical attributes of peaches.

2. Materials and Methods

2.1. Experimental Site and Plant Materials

A two-year research study was conducted at the Horticulture Research Farm, The University of Agriculture Peshawar (UAP), Pakistan, which is situated in a subtropical climate at 34.01° N latitude and 71.35° E longitude, at an elevation of 350 m above sea level. The average temperatures range from a minimum of 5 °C in winter to a maximum of 45 °C in summer. March is the wettest month, while June is the driest. The soil type was Calcaric Luvisols (FL ca), according to the World Reference Base (WRB) system of soil taxonomy, and had a silt loam texture which comprised sand 19.4%, silt 71.6%, and clay 8.96% with a pH range of 7.8–8.2 [17]. Specific meteorological data for the experimental site is provided in Figure 1. The experiment was conducted in an existing peach orchard at the Horticulture Research Farm, The University of Agriculture Peshawar. Disease-free, uniformly sized peach trees of the Florida King cultivar, approximately 10 years old, were selected for this experiment. These trees were grafted onto the local Peshawar peach rootstock [18].

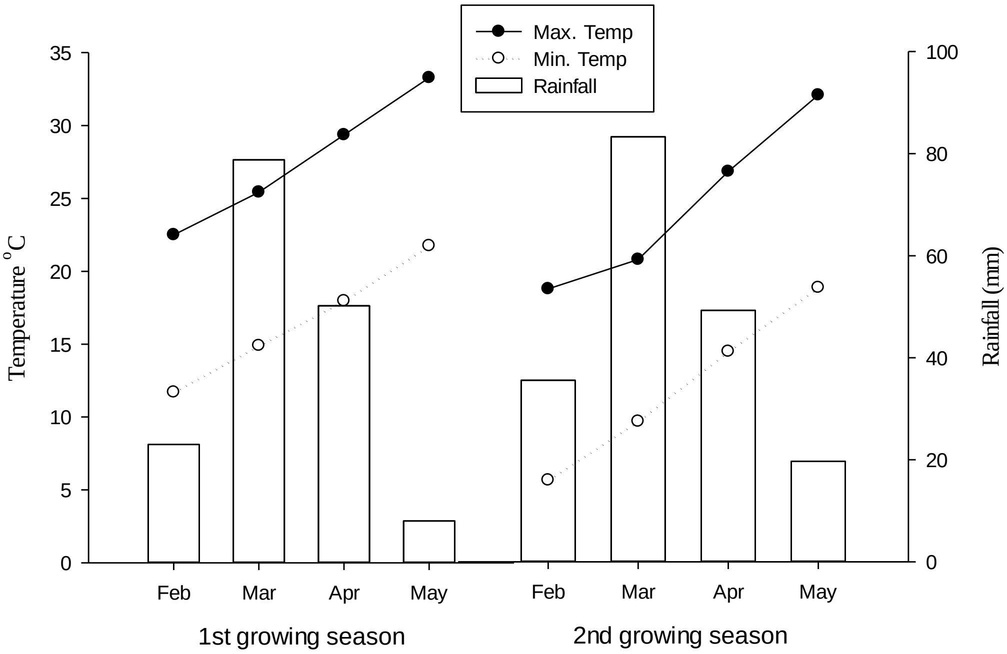

Figure 1. The climatic data of the experimental site during the two growing seasons of peaches.

2.2. Experimental Design and Treatment Combinations

This research study was conducted using a Randomized Complete Block Design (RCBD) with a two-factorial arrangement, replicated three times. Peach trees were foliar sprayed with four concentrations of silicon (0, 0.1%, 0.2%, and 0.3%) and four concentrations of zinc (0, 0.25%, 0.50%, and 0.75%) over two consecutive growing seasons. Each replication consisted of 16 treatments, resulting in a total of 48 experimental units/treatments in all three replications. Two trees were assigned to each treatment per replication, leading to 96 trees per replication and a total of 288 trees for the entire experiment.

2.3. *Preparation and Application of Silicon and Zinc Sprays*

For the preparation of various concentrations (0.1, 0.2, and 0.3%) of silicon solutions, 4.34, 8.69, and 13.04 g of sodium silicate (Na_2SiO_3) were dissolved in 1 L of distilled water. In order to make different concentrations of zinc (0.25, 0.50, and 0.75%) solution, 7.1, 14.2, and 21.4 g of zinc sulphate ($ZnSO_4$) were dissolved in 1 L of distilled water. Each peach tree required 3 L of water, so the amounts of silicon and zinc were adjusted for the preparation of a 3 L solution for each treatment, which was applied with a hand sprayer. Both silicon and Zn were applied two times, i.e., the first spray was carried out at the berry stage, and then, the second one was performed at the pit hardening stage. Peach fruit plants were sprayed with silicon and zinc during the morning, keeping many of the environmental factors in consideration. During the spray procedure, large plastic sheets were wrapped with bamboo sticks spread around the tree so that other plants were not affected with the specific dose applied to it.

2.4. *Harvesting and Postharvest Procedure*

Peach fruits of uniform shape and size, with no signs of any disease or bruise, and collected at physiological maturity were brought to the Post harvest Laboratory, Department of Horticulture, The University of Agriculture Peshawar. The fruits were cleaned, washed, and dried for determining various physicochemical, phytochemical, and enzymatic attributes.

2.5. *Data Collection*

2.5.1. *Fruit Growth and Yield Attributes*

Single fruit weight was measured by randomly selecting 20 fruits (10 fruits from each tree) from each treatment in each replication and weighing them using a Sartorius Laboratory balance (Model Cubis ii, Sartorious manufacturers, Otto-Brenner-Str. 20, 37079 Göttingen, Germany). The average weight was then calculated. The volume of peach fruit was measured using the water displacement method. A 1 L beaker was filled completely with water, and 10 randomly selected peach fruits were fully immersed. The displaced water was collected and measured in mL using a conical flask. Since 1 mL is equivalent to 1 cm^3, the fruit volume was recorded in cubic centimeters (cm^3). To determine the fruit yield $tree^{-1}$, all of the fruits on each tree were counted. A sample of 10 fruits from each branch was taken, weighed using a digital balance, and the average fruit weight was calculated. The total fruit yield $tree^{-1}$ was then determined by multiplying the average fruit weight by the number of fruits on the tree at the time of harvest.

2.5.2. *Pulp Stone Ratio*

To determine the pulp-to-stone ratio, the pulp was removed from the selected fruits of each treatment, and the average pulp weight was calculated. The stones from the fruits of each treatment were then extracted, and the average stone weight was determined. The pulp-to-stone ratio was calculated by dividing the average pulp weight by the average stone weight.

$$Pulp\ stone\ ratio = \frac{average\ pulp\ weight}{average\ stone\ weight}$$

2.5.3. *Biochemical Attributes*

Fruit firmness was measured following the procedure of Pocharski et al. [19] using a handheld penetrometer (FT-011 model, Effigi manufacturer, 1185 Pineridge Road Norfolk, VA 23502, USA) with an 8 mm probe. A digital hand refractometer (model: R9500, manufacturer: Reed instruments, REED Instruments 16975 Leslie St Newmarket, ON L3Y 9A1, Ontario, Canada) was used to evaluate the total soluble solids (TSS) of the peach fruit.

A small portion of the peach was peeled to expose the flesh, and a drop of fruit juice from the peeled area was placed on the refractometer prism. The reading in °Brix was noted by pointing the refractometer toward a light source. The titration method was used to determine titratable acidity. A 10 mL sample of peach juice was diluted to 100 mL using distilled water, and 2-3 drops of phenolphthalein were added. The prepared sample was then titrated against 0.1 N NaOH solution [20]. The results were expressed as a percentage of citric acid. Titratable acidity was calculated using the following formula:

$$Titratable\ acidity = \frac{N \times T \times F \times 100}{D \times S} \times 100$$

where

N = the normality of NaOH;
T = NaOH used in mL;
S = diluted sample in mL;
D =sample taken for dilution;
F = constant factor 0.0064.

The TSS-to-acid ratio was calculated using the following formula:

$$TSS\ to\ acid\ ratio = \frac{Total\ soluble\ solids}{Titratable\ acidity}$$

To find out the ascorbic acid content, 0.4% oxalic acid was added to a 100 mL volumetric flask containing 10 mL of juice sample. An aliquot sample (5 mL) was calculated and titrated against 2, 6-dichlorophenol indophenol dye until the appearance of a light pink color. The estimation of vitamin C was measured with the given formula [20].

$$vitamin\ C\ (\text{mg}/\ 100\ \text{g}) = \frac{F \times T \times 100}{D \times S} \times 100$$

where

F = dye factor constant (0.064);
T = dye solution utilized from burette (mL);
S = juice sample for titration (10 mL);
D = fruit juice for dilution (10 mL).

Reducing and non-reducing sugars were determined using the procedure given by AOAC [20].

2.6. Sample Preparation and Quantification of DPPH Free Radicals Antioxidant Activity (%), Flavonoid Content (mg g^{-1} FW), and Proline Content (μg g^{-1} FW)

Samples from each treatment were collected and air-dried. The samples were then converted into a fine powder with the help of a pestle and mortar. For sample preparation, a known quantity of sample (10 mg) and ethanol (10 mL) in a 1:1 ratio were mixed and incubated for 7 days. The incubated samples were then centrifuged for almost 15 min at ten thousand rpm. For the quantification of flavonoids, and antioxidant activities, supernatants were collected and further subjected to quantification using the methods of Ahmad et al. [21]. For the quantification of proline content, peach mesocarp flesh tissue (1 g) was homogenized with 8 mL of 80% (*v:v*) ethanol and extracted for 1 h in the dark, then centrifuged at $12{,}000\times\ g$ and 4 °C for 10 min and the supernatant was collected. The proline content was measured according to the method of Sanchez et al. [22], and the result was expressed as μg g^{-1} FW.

2.7. Enzymatic Activities

The catalase activity of peach fruit was ascertained by the method as described by Abbasi et al. [23]. To accomplish the reaction buffer solutions, one (Buffer A) was prepared by adding 2.9 mL, 15 M K_3PO_4 buffer at pH 7.0 in a cuvette and the other solution (Buffer B) was made by adding 2.9 mL of 12.5 mM H_2O_2 in 15 M K_3PO4 buffer (pH 7.0) in another cuvette. Two cuvettes were separately filled with 100 µL of enzyme extract. Subsequently, the two cuvettes were placed in a gloomy box. The optical densities at 240 nm for these cuvettes were noted at 45 and 60 s and this time was noted when the cuvettes were filled with extract. The optical density difference at 45 and 60 s intervals was recorded using the spectrophotometer Optima® 3000 plus (model name: SP 3000, Manufacturer: Optima Inc., OPTIMA Bldg. 1-6-8 Yamatocho, Nakano-ku, Tokyo 165-0034 JAPAN) and the readings were used to compute the catalase activity. The results were expressed as catalase unit g^{-1} protein.

The peroxidase activity of peach fruit was determined as described by Abbasi et al. [23] with slight modification. The reaction mixture consisted of 1.7 mL, 15 mM $NaKO_4P^-$ buffer (pH 6.0). The two substrates consisted of 500 µL 0.1 mM guaiacol and 500 µL 1 mM H_2O_2 and 300 µL enzymes extracted in a 3 mL cuvette. Peroxidase activities were noted for OD (optical density) change over a 3 min time at 470 nm and the results were expressed as unit g^{-1} protein.

2.8. Statistical Analysis and Data Visualization

The data recorded were arranged with a 2-factorial RCB design and a two-way Anova technique using the F-Test was applied to check the significance of the data. Statistix-8.1 (statistical software version 8.1) was used to analyze the data. In the case of significance, the Least Significance Difference (LSD) test was applied to check differences among the means [24].

3. Results

3.1. Fruit Growth and Yield Attributes

The individual application of silicon and zinc significantly influenced the fruit weight, volume, and yield $tree^{-1}$ of the peaches. However, the interaction between silicon and zinc on these attributes was found to be non-significant. The mean data indicated that increasing the silicon concentration from 0 to 0.3% significantly increased fruit weight from 88.63 to 111.33 g, representing a 26% increase compared to the control. Similarly, fruit volume increased from 95.83 to 117.90 cm^3, a 23% rise compared to the control, and yield $tree^{-1}$ rose from 97.38 to 137.61 kg, a 44% increase compared to the control (Figure 2A). Regarding zinc, there was a significant increase in fruit weight from 95.37 to 112.60 g, fruit volume from 104.59 to 131.06 cm^3, and yield $tree^{-1}$ from 104.40 to 127.27 kg $tree^{-1}$ with increasing zinc concentration from control to 0.75% (Figure 2D). These results suggest an 18% increase in fruit weight, 25% increase in fruit volume, and 22% increase in fruit yield with the application of 0.75% zinc compared to the control.

3.2. Quality or Biochemical Attributes

The results of data showed that fruit firmness (Table 1), total soluble solids (Figure 2B,E), titratable acidity (Figure 2B,E), TSS-to-acid ratio (Figure 2B,E), ascorbic acid (Table 1), reducing sugars (Figure 2C,F), and non-reducing sugars (Figure 2C,F) were significantly affected by alone application of silicon and zinc concentrations. Furthermore, the interaction between silicon and zinc (S × Z) was found to be non-significant for fruit firmness, total soluble solids, reducing sugars, and non-reducing sugars, but significant for the titratable acidity (Figure 3A), TSS-to-acid ratio (Figure 3B), and ascorbic acid (Figure 4A) of peach fruits.

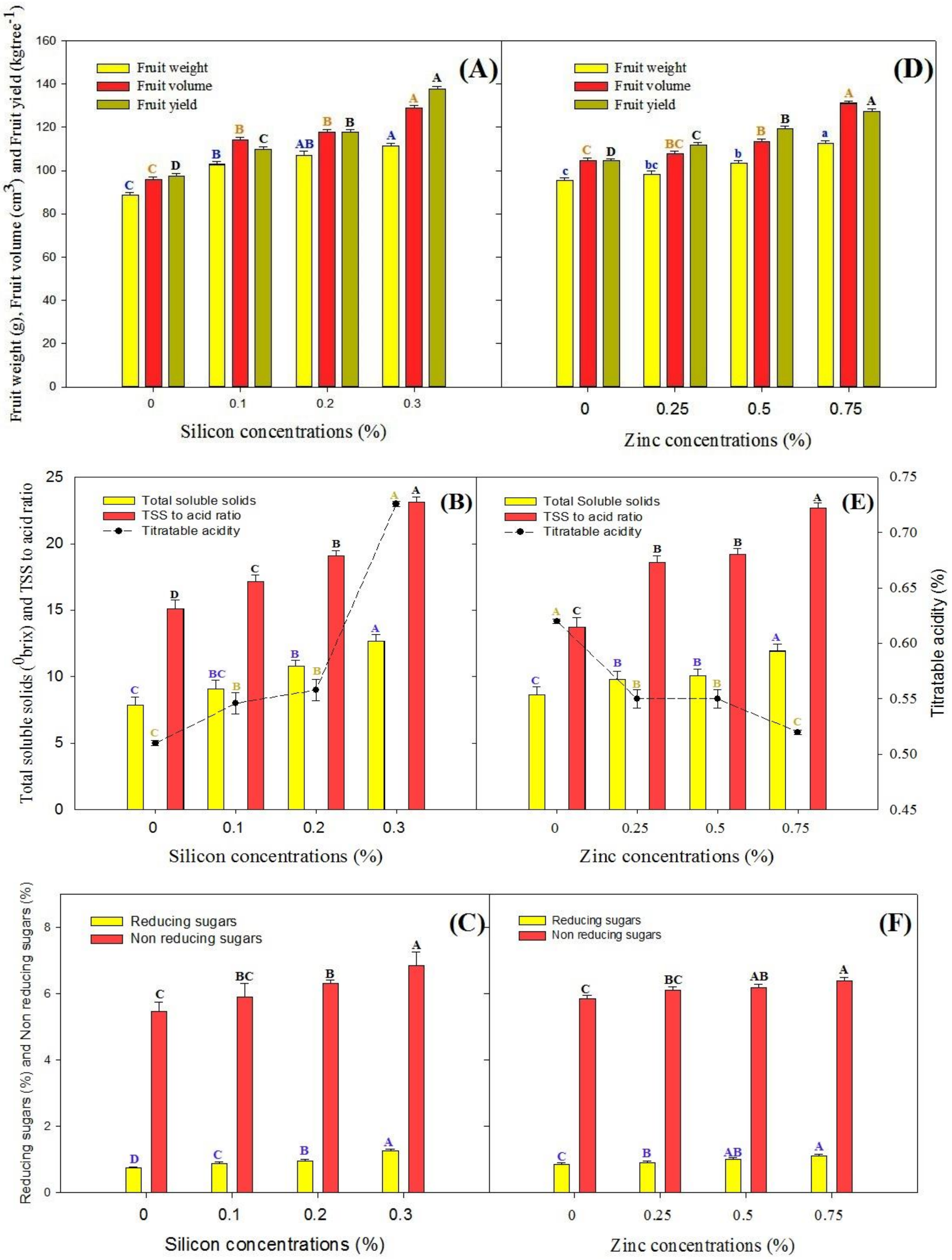

Figure 2. Fruit weight, fruit volume, fruit yield, total soluble solids, titratable acidity, TSS-to-acid ratio, and reducing and non-reducing sugars of peach fruits as affected by silicon (**A**–**C**) concentrations and zinc (**D**–**F**) concentrations. Vertical bars show standard error. Capital (color) and small letters show data significance at $p \leq 0.01$ and $p \leq 0.05$, respectively. The means are an average of two years.

The mean data regarding silicon concentrations showed that fruit sprayed with 0.3% silicon concentration showed maximum fruit firmness (1.53 kg cm^{-2}), TSS (12.69 °Brix), titratable acidity (0.68%), TSS-to-acid ratio (23.13), vitamin C (6.27 mg 100 g^{-1}), reducing sugars (1.26%), and non-reducing sugars (6.85%), while minimum fruit firmness (0.95 kg cm^{-2}), TSS (7.89 °Brix), titratable acidity (0.50%), TSS-to-acid ratio (13.5), vitamin C (4.72 mg 100 g^{-1}), reducing sugars (0.75%), and non-reducing sugars (5.85%) were observed in the control treatment (Table 1, Figure 2B,C). This suggests that the highest dose of silicon (0.3%) resulted in a 61% increase in fruit firmness, 62% in TSS, 36% in TA, 71% in TSS-to-acid ratio, 32% in vitamin C, 68% in reducing sugars, and 17% in non-reducing sugars as compared to the control treatment. Regarding the means for zinc concentration, fruit sprayed with 0.75% zinc concentration recorded the maximum fruit firmness (1.50 kg cm^{-2}), TSS (11.93 °Brix), titratable acidity (0.52%), TSS-to-acid ratio (22.7), vitamin C (6.14 mg 100 g^{-1}), reducing sugars (1.11%), and non-reducing sugars (6.39%), while the minimum fruit firmness (0.99 kg cm^{-2}), TSS (8.66 °Brix), titratable acidity (0.62%), TSS-to-acid ratio (13.75), vitamin C (4.77 mg 100 g^{-1}), reducing sugars (0.86%), and non-reducing sugars (5.58%) were observed in fruit sprayed with 0% zinc (Table 1, Figure 2E,F). This means that fruit firmness, TSS, TSS-to-acid ratio, vitamin C, reducing sugars, and non-reducing sugars were increased by 52, 37, 65, 28, 29, and 15% when zinc was applied at 0.75% compared with control treatment. The TA was decreased by 16% by the application of the highest level of zinc compared to the control treatment. Regarding the interaction of silicon and zinc, the titratable acidity tends to decrease at all levels of silicon and zinc; however, a steady decrease was observed at 0.3% silicon at all levels of zinc, whereas a sudden decrease in the titratable acidity of peach fruits was observed when the concentration of zinc increased from 0.25 to 0.5%, and with silicon concentrations at 0.1 and 0.2%, respectively. Control fruits showed a linear decrease in titratable acidity at all concentrations of silicon and zinc (Figure 3A). Regarding the S × Z interaction, thee TSS-to-acid ratio of peaches increased at all levels of silicon and zinc. However, a linear increase was observed in silicon concentrations at 0.1 and 0.2% at all levels of zinc, whereas the TSS-to-acid ratio initially showed a slow increase when plants were sprayed with silicon at 0.3%, with an increase in zinc concentration from 0 to 0.50%; however, a sudden increased was observed when zinc concentration increased to 0.75% at all levels of silicon. Control fruits showed a steady and slow increase in the TSS-to-acid ratio of peach fruits (Figure 3B). The S × Z interaction was also found to be significant for the ascorbic acid content of peach. The data showed that a linear increase in ascorbic acid was observed at all levels of silicon and zinc (Figure 3C).

Table 1. Fruit firmness (kg cm^{-2}) and ascorbic acid (mg 100 g^{-1}) of peach fruits as affected by the foliar application of silicon and zinc.

Treatments	Parameters	
Silicon (S) Levels (%)	**Fruit Firmness**	**Vitamin C**
0	0.95 C	4.72 C
0.1	1.18 B	5.00 BC
0.2	1.25 B	5.69 AB
0.3	1.53 A	6.27 A
LSD ($p \leq 0.01$)	0.14	0.72
Zinc (Z) concentrations (%)		
0	0.99 C	4.77 C
0.25	1.17 B	5.14 BC
0.50	1.26 B	5.63 AB
0.75	1.50 A	6.14 A
LSD ($p \leq 0.01$)	0.14	0.72
Interaction ($p \leq 0.01$)		
S × Z	NS	**

Means followed by similar letters are statistically at par with each other at the 1% level of significance. Capital letters show that the data are significant at $p \leq 0.01$. The means are an average of two years. NS: non significant; ** data significant at $p \leq 0.05$.

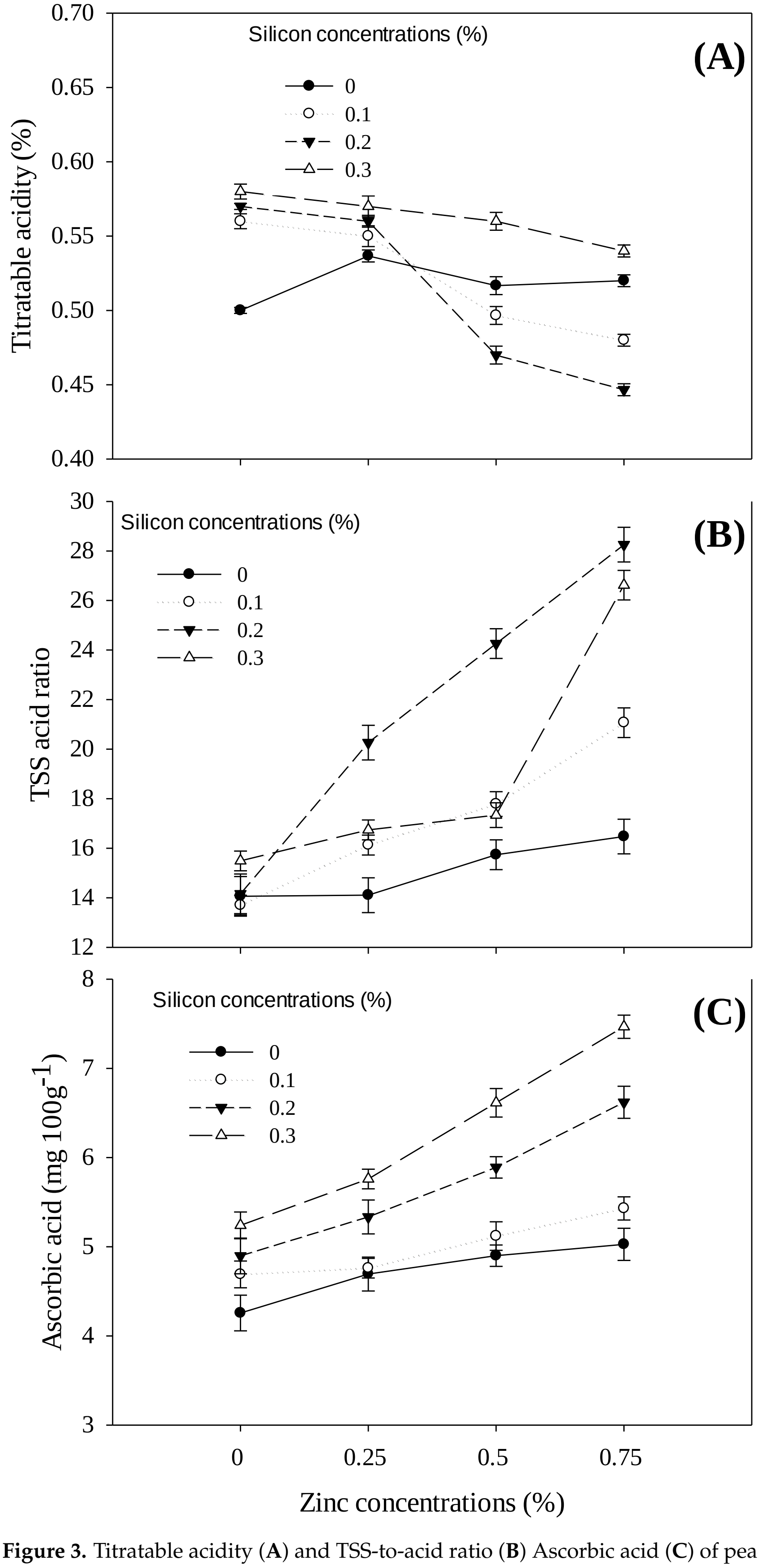

Figure 3. Titratable acidity (**A**) and TSS-to-acid ratio (**B**) Ascorbic acid (**C**) of peach fruits as affected by the interaction of silicon and zinc. Error bar shows standard error. The means are an average of two years.

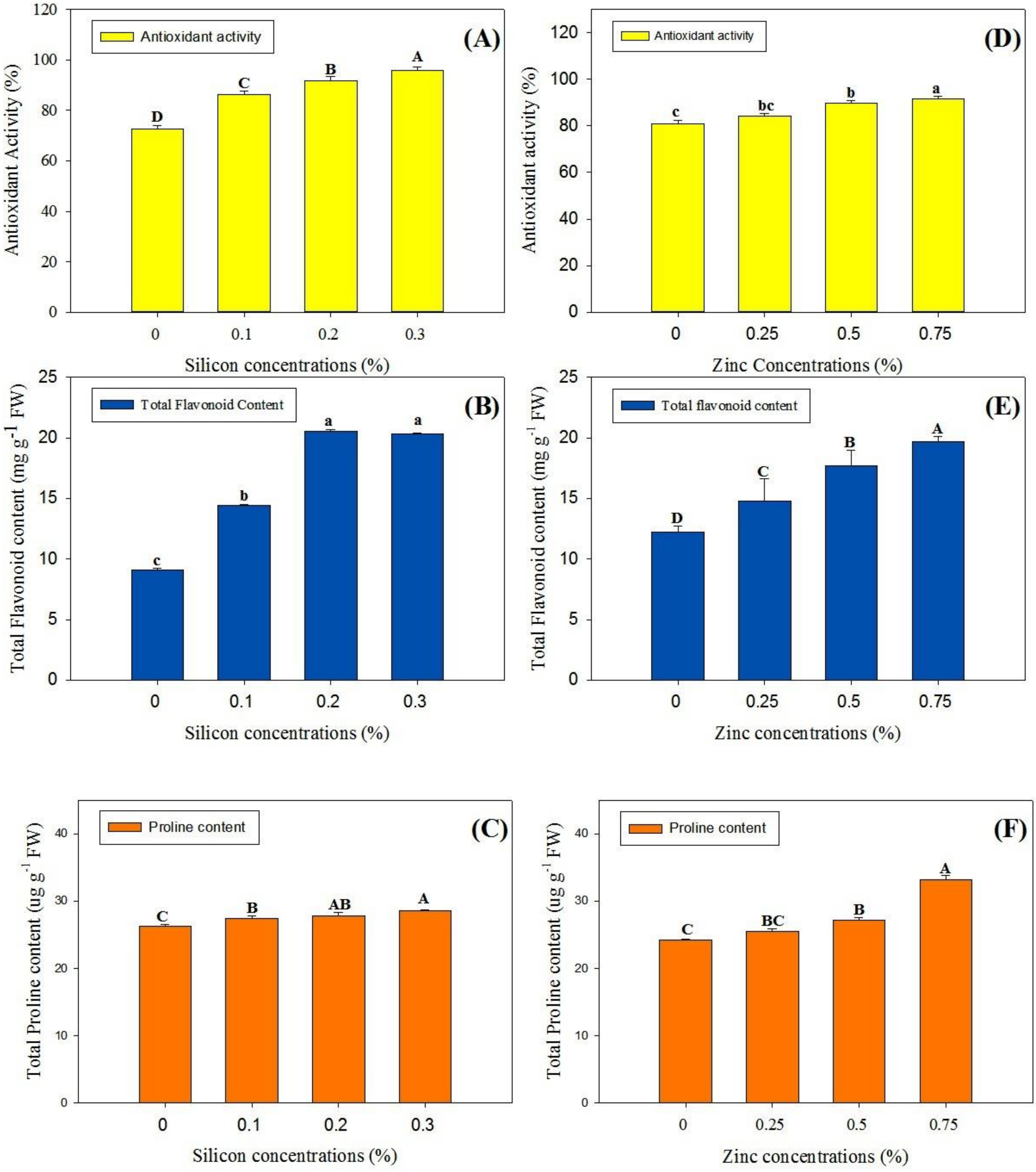

Figure 4. Antioxidant activity (%) (**A**,**D**), total flavonoid content (mg g^{-1} FW) (**B**,**E**), and total proline content (µg g^{-1} FW) (**C**,**F**) of peach fruits as affected by silicon and zinc concentrations, respectively. Error bar shows standard deviation. Capital and small letters show data significance at $p \leq 0.01$ and $p \leq 0.05$, respectively. The means are an average of two years.

3.3. Phytochemical Analysis and Enzymatic Activities

The analysis of data showed that the antioxidant activity, flavonoid content, and catalase (CAT), peroxidase (POD), and proline levels in the fruit were significantly ($p \leq 0.01$) affected by the application of different concentrations of silicon and zinc. Furthermore, the interaction between silicon and zinc (S × Z) was found to be significant for antioxidant activity, flavonoid content, and peroxidase activity.

Fruits sprayed with a 0.3% silicon concentration showed the maximum antioxidant activity (95.93%) and catalase activity (35.76 U g^{-1} protein), which were statistically different from the other treatments. In contrast, the minimum antioxidant activity (72.77%) and catalase activity (29.10 U g^{-1} protein) were observed in control plants untreated with silicon (Figures 4A and 5B). This represents a 32 and 23% increase in antioxidant and catalase activity, respectively, when silicon was applied at 0.3% compared to the control. Moreover, the maximum peroxidase activity (41.37 U g^{-1} protein) was observed in peach plants sprayed with 0.1 and 0.2% silicon, while the minimum peroxidase activity (40.94 U g^{-1} protein) was recorded in control plants untreated with silicon (Figure 5A). This indicates that peroxidase activity increased with the application of 0.2% silicon compared to the control. A similar trend was observed for total flavonoid content, proline, and peroxidase activity. The maximum flavonoid (20.55 mg g^{-1} FW) content was observed in plants sprayed with 0.2% silicon, which was statistically at par with the flavonoid content (20.31 mg g^{-1} FW) in plants sprayed with 0.3% silicon. In contrast, the lowest flavonoid content (9.12 mg g^{-1} FW) was recorded in untreated control plants (Figure 4B). An increase of 122% in flavonoid content was recorded in fruits sprayed with 0.2% silicon compared to the control treatment. The maximum proline content (28.52 µg g^{-1} FW) was observed in peach plants foliar sprayed with 0.3% silicon, which was statistically at par with the proline content (27.78 and 27.42 µg g^{-1} FW) in plants sprayed with 0.2 and 0.1% silicon, respectively. The lowest proline content (26.27 µg g^{-1} FW) was observed in untreated peach plants (Figure 4C). This shows that peach plants sprayed with 0.3% silicon exhibited a 9% increase in proline content compared to the control treatment. Regarding zinc concentration, peach fruit plants sprayed with 0.75% zinc recorded the highest antioxidant activity (91.47%), flavonoid content (19.69 mg g^{-1} FW), proline content (33.17 µg g^{-1} FW), and peroxidase activity (41.46 U g^{-1} protein). In contrast, the lowest antioxidant activity (81.08%), flavonoid content (12.21 mg g^{-1} FW), proline content (24.17 µg g^{-1} FW), and peroxidase activity (41.05 U g^{-1} protein) were observed in the control treatment (untreated plants) (Figures 4D–F and 5C). This shows an increase of 13% in antioxidant activity, 61% in flavonoid content, 37% in proline content, and 1% in peroxidase activity in plants sprayed with 0.75% zinc compared to the control treatment where no zinc was applied.

Moreover, the highest catalase (34.94 U g^{-1} protein) activity was recorded in fruit plants sprayed with 0.50% zinc, while the lowest catalase activity (29.66 U g^{-1} protein) was observed in plants sprayed with distilled water (0% zinc) (Figure 5D). The data indicate an 18% increase in catalase activity in peach plants sprayed with 0.50% zinc compared to the control treatment.

For the interaction effect on antioxidant activity, control fruits showed a non-significant increase at all levels of silicon and zinc. However, a significant increase in antioxidant activity was observed at 0.1%, 0.2%, and 0.3% silicon when zinc levels increased from 0 to 0.25%, while it remained steady at 0.50% and 0.75%, respectively (Figure 6).

Regarding flavonoid content, there was a steady increase at 0% and 0.2% silicon across all zinc levels. At 0.3% silicon, the flavonoid content initially did not increase from 0 to 0.25% zinc, but then showed a sharp increase from 0.25% to 0.75% zinc (Figure 7A). For peroxidase activity, a steady increase was observed across all levels of silicon and zinc. However, a sharp increase occurred in peach fruits treated with 0.3% silicon and 0.75% zinc (Figure 7B). The S × Z interaction for proline content and peroxidase activity was found to be non-significant.

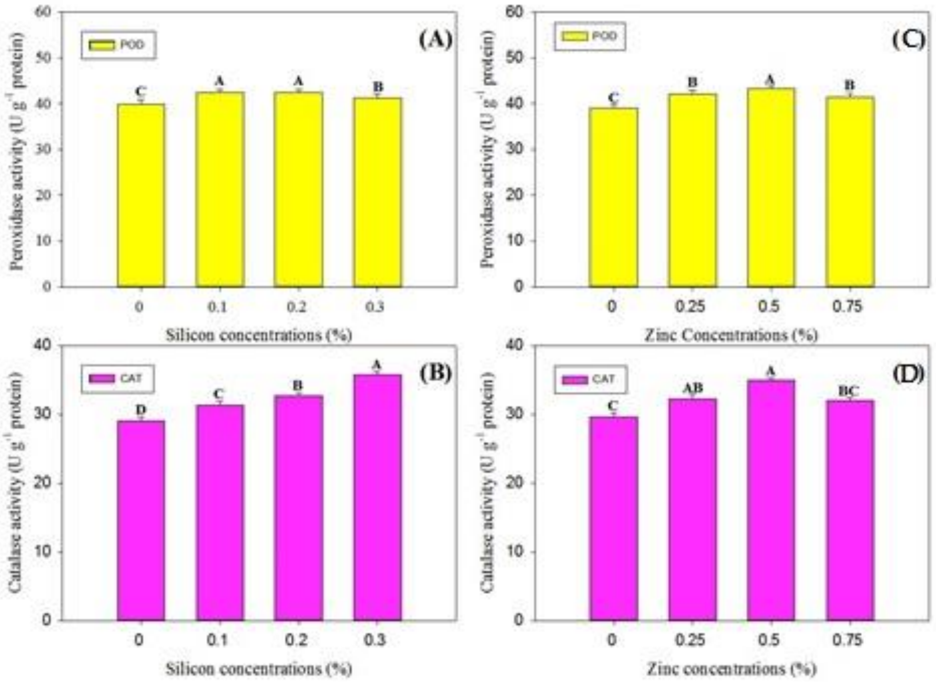

Figure 5. Peroxidase activity (U g^{-1} protein (**A**,**C**) and catalase activity (U g^{-1} protein) (**B**,**D**) of peach fruits as affected by silicon and zinc concentrations, respectively. Error bar shows standard deviation. Capital letters show data significance at $p \leq 0.01$. The means are an average of two years.

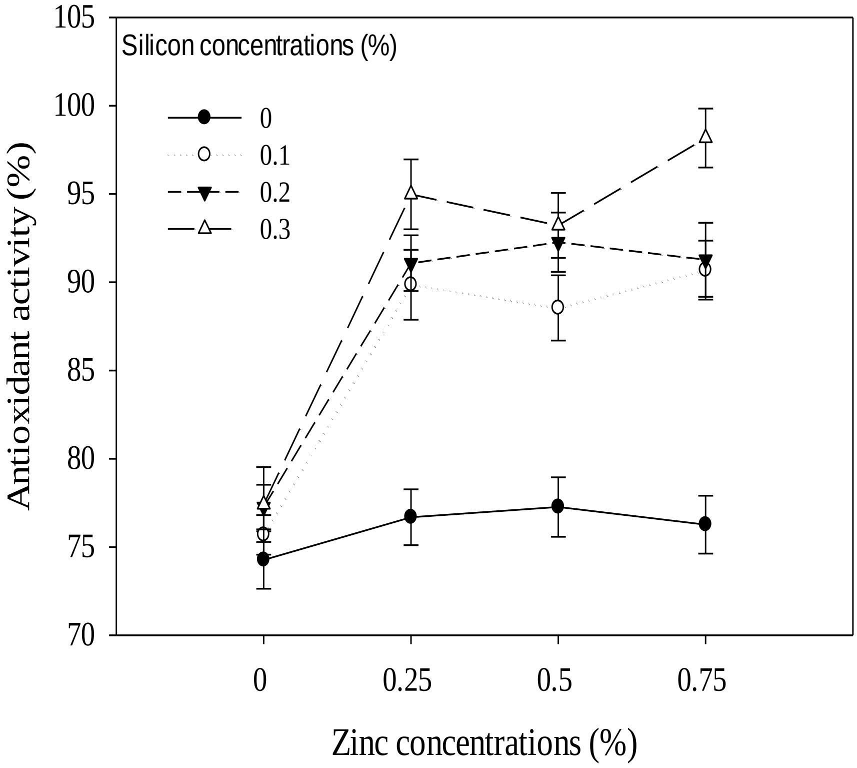

Figure 6. Antioxidant activity (%) of peach fruits as affected by the interaction of silicon and zinc concentrations. Error bar shows standard deviation. The means are an average of two years.

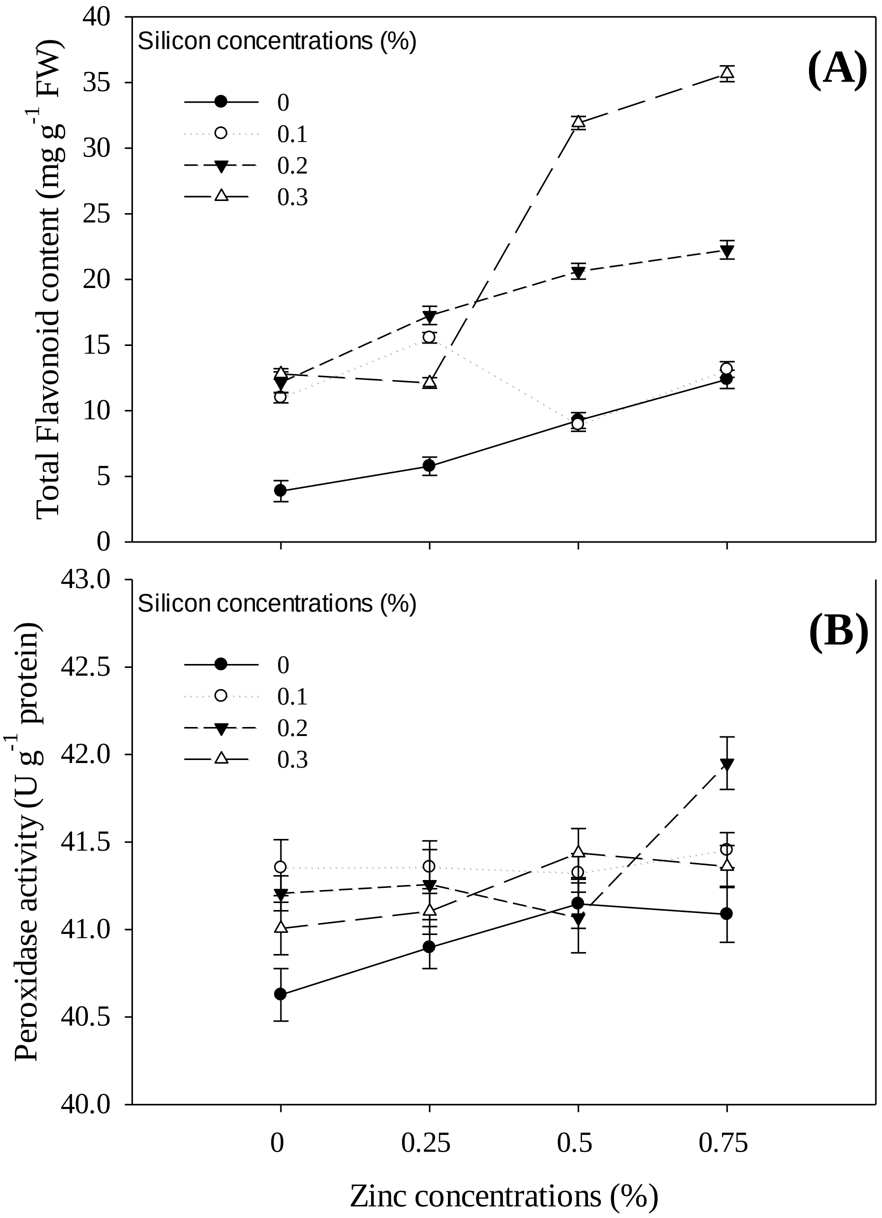

Figure 7. (**A**) Total flavonoid content (mg g^{-1} FW) and (**B**) peroxidase activity (U g^{-1} protein) of peach fruits as affected by the interaction of silicon and zinc concentrations. Error bar shows standard deviation. The means are an average of two years.

3.4. Correlation Analysis

The heatmap in Figure 8 illustrates the correlation coefficients among the various studied traits, demonstrating significant interrelationships. Notably, there is a strong positive correlation between disease incidence (DI) and the number of fruits (NF) ($r \approx 0.90$), indicating a high degree of interdependence between these traits. Similarly, fruit weight (FW) and NF exhibit a strong positive correlation ($r \approx 0.85$), as do fruit yield plant^{-1} (FYpT) and fruit firmness (FR) ($r \approx 0.80$), and pulp-to-stone ratio (PSR) and FR ($r \approx 0.78$).

Moderate positive correlations are observed between TSS and TA ($r \approx 0.65$), and between PH and FR ($r \approx 0.60$), suggesting that these trait pairs also share considerable commonality. Conversely, a weak negative correlation is observed between DM and FW ($r \approx -0.30$), highlighting an inverse relationship between these traits. Similarly, a moderate negative correlation exists between DM and DI ($r \approx -0.50$), and between PH and TA ($r \approx -0.45$).

Interestingly, the TSS-to-acid ratio exhibits a strong negative correlation with both TSS ($r \approx -0.75$) and reducing sugars (RD) ($r \approx -0.70$), suggesting these traits have significant opposing trends. Additionally, weak to moderate negative correlations are present between antioxidant activity (AnOX) and RD ($r \approx -0.40$) and between flavonoid content (FLD) and fruit volume (FV) ($r \approx -0.35$), further underscoring the complex interdependencies among the studied traits.

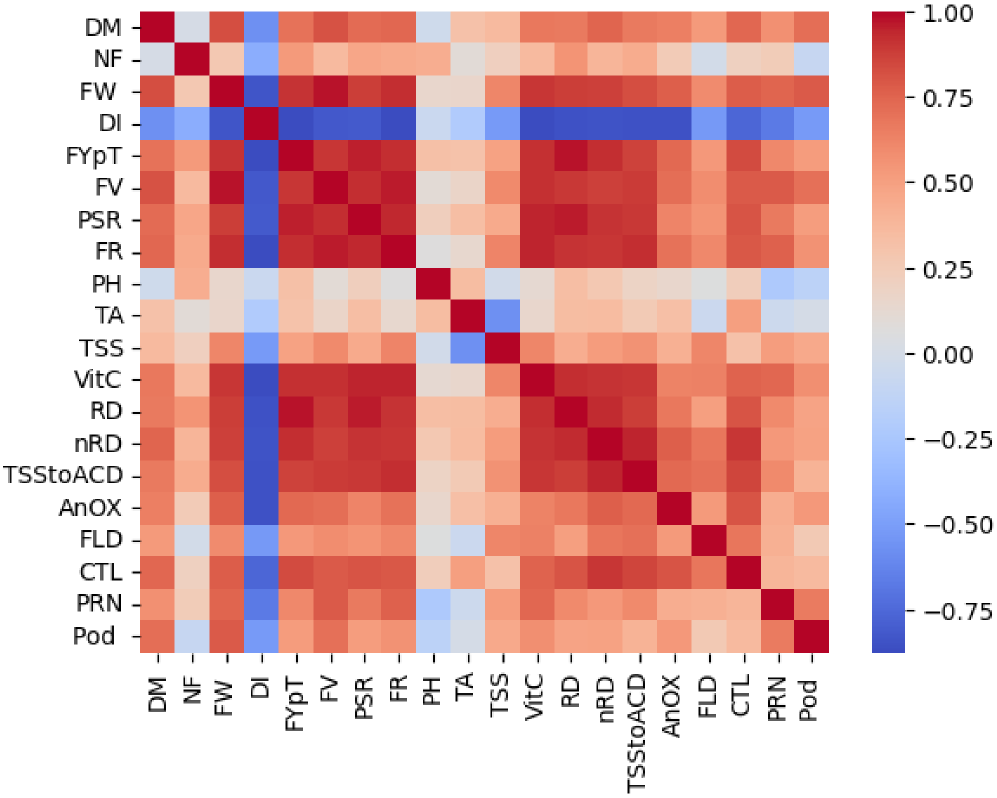

Figure 8. Heatmap depicting the correlation coefficients among various studied traits. The color gradient from blue to red represents the range of correlation values, with blue indicating negative correlations and red indicating positive correlations. FW: fruit weight; DI: disease incidence; FYpT: fruit yield plant^{-1}; FV: fruit volume; PSR: pulp-to-stone ratio; FR: fruit firmness; TA: titratable acidity; TSS: total soluble solids; VitC: ascorbic acid; RD: reducing sugars; nRD: non-reducing sugars; TSStoACD: TSS-to-acid ratio; AnOX: antioxidant activity; FLD: flavonoid content; CTL: catalase activity; PRN: proline content; Pod: peroxidase activity.

3.5. Principal Component Analysis

The PCA biplot in Figure 9 elucidates the relationships among various traits and their contributions to the first two principal components (PC1 and PC2). PC1 captures 64.63% of the total variance, while PC2 accounts for 11.19%, together explaining 75.82% of the variability within the dataset.

Traits such as TSS, Pod, PRN, FLD, and DM exhibit strong positive loadings on PC1, indicating their significant contribution to the primary axis of variation. Conversely, DI shows a strong negative loading on PC1, suggesting an inverse relationship with the positively loading traits. Similarly, traits like TA, PH, and NF have substantial positive loadings on PC2, indicating their prominent role in the secondary source of variation.

The clustering of traits like RD, FYpT, PSR, and CTL near the origin signifies their lesser influence on the first two principal components, suggesting that these traits do not contribute significantly to the primary axes of variation in this dataset.

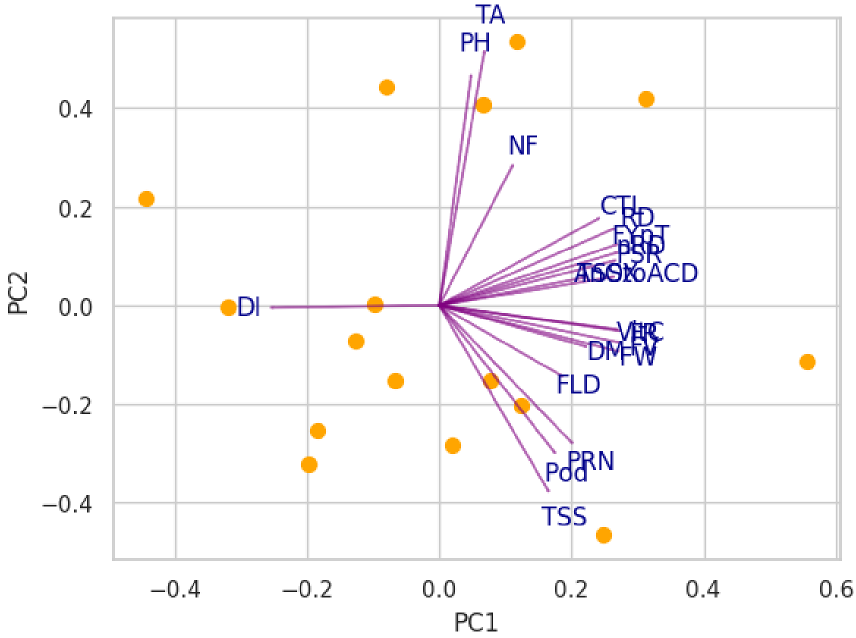

Figure 9. PCA biplot illustrating the relationships between different traits and their contributions to the first two principal components (PC1 and PC2). PC1 accounts for 64.63% of the total variance, while PC2 explains 11.19%, cumulatively capturing 75.82% of the variability in the dataset. Arrows indicate the direction and magnitude of each trait's contribution to the principal components. FW: fruit weight; DI: disease incidence; FYpT: fruit yield plant^{-1}; FV: fruit volume; PSR: pulp-to-stone ratio; FR: fruit firmness; TA: titratable acidity; TSS: total soluble solids; VitC: ascorbic acid; RD: reducing sugars; nRD: non-reducing sugars; TSStoACD: TSS-to-acid ratio; AnOX: antioxidant activity; FLD: flavonoid content; CTL: catalase activity; PRN: proline content; Pod: peroxidase activity.

4. Discussion

4.1. Fruit Growth and Yield Attributes

The weight of the fruit is directly linked to consumer preference and farmers' profit, making it a vital quality feature [25]. The application of silicon not only increased the number of fruits [26] but also enhanced the weight of individual fruits (Figure 2A). The increase in fruit weight with silicon application is attributed to the uptake of nutrients and water from the soil, which boosts the rate of photosynthesis [27]. The photosynthates produced by the plant are then transported to the developing fruit [28], resulting in increased fruit weight. Furthermore, the increment in fruit weight may be due to cell division during the early growth stages, followed by cell expansion, which is facilitated by silicon's role in transporting water and other metabolites within the cell [29]. A similar effect of silicon on fruit weight was reported by Ravishankar [30] in bananas. Likewise, Jarosz [31] also observed a significant increase in tomato fruit weight with the application of silicon.

Zinc is essential for regulating osmotic activities, maintaining cell water balance, and preserving cell membrane structural stability [32]. The application of micronutrients, particularly zinc, is beneficial for enhancing fruit growth and plant metabolism [33]. Dhotra et al. [34] reported similar effects of zinc in improving the weight of peach fruits. Foliar zinc treatments have also been shown to increase fruit weight in both oranges [35] and peaches [1], with studies highlighting significant increases in peach fruit weight due to zinc application.

The increased fruit volume may be attributed to the application of silicon, which promotes cell division and expansion. This process facilitates the transfer of metabolites and water into the cells, ultimately leading to an increase in both total fruit volume and weight [29]. Silicon aids in stimulating the uptake of nutrients by plants, which in turn enhances the process of photosynthesis [27]. Shehata and abdelgawad [36] also reported that silicon application increased the fruit length, fruit volume, and total fruit yield of squash. The increase in fruit volume due to zinc application may be attributed to its stimulatory effects on plant metabolism, as observed by Rawat et al. [37] in guava. Additionally, increases in fruit volume were reported with zinc sprays in kinnow [38] and lychee [39].

As a beneficial plant nutrient, silicon is used to enhance the development and productivity of various crops [40]. It increases yield tree^{-1} by improving the plant's photosynthetic efficiency, leading to a higher concentration of solutes in the leaves. These photosynthates are actively transported to the fruit, thereby boosting overall productivity and yield tree^{-1} [41]. The application of silicon has also been shown to increase fruit yield in apricot [42]. Additionally, the positive effects of potassium silicate in enhancing yield and fruit characteristics in sapota were reported by Lalithya et al. [43]. Zinc plays a key role in regulating enzymatic activity [44] and promotes chlorophyll production, which boosts photosynthesis, and contributes to increased yield [45]. The yield-enhancing effects of zinc have also been observed in mango, as reported by Singh et al. [46].

4.2. Pulp-to-Stone Ratio

The pulp-to-stone ratio increased with a higher concentration of silicon, which might be due to the translocation of produced photosynthates to growing fruits [47]. This improvement in growth and yield-related attributes has also been observed in bananas [30]. Similarly, the application of silicon has been found to enhance fruit quality, increase the pulp-to-stone ratio, and enlarge fruit size in sapota [43].

Zinc significantly increased the pulp-to-stone ratio, likely due to enhanced activity and the accumulation of photosynthates in plant parts, facilitating the conversion and translocation of minerals and sugars during fruit development and maturation [48]. The foliar application of zinc increased its concentrations in the fruit pulp, which act as a stronger sink compared to other fruit parts [49]. Furthermore, zinc's direct role in translocating nutrients and minerals throughout the fruit development process contributed to the increased pulp-to-stone ratio. These findings align with those reported by Shukla et al. [50].

4.3. Biochemical Attributes

Silicon enhances tissue strength and rigidity, which likely contributed to the observed increase in fruit firmness. This effect may be attributed to silicon's role in binding pectin molecules within the cell wall, thereby stabilizing its structure and improving firmness [51]. Silicate fertilization has been shown to enhance firmness in tomatoes [51]. Similarly, zinc foliar sprays improve fruit firmness by regulating biological membranes, potentially due to zinc's role in enhancing lignification and membrane strength [52]. A similar trend in fruit firmness was observed in zinc-sprayed litchi plants, as reported by Cronje et al. [39].

Total soluble solids (TSS) are an indirect indicator of sugar concentration in fruit and one of the most fundamental tools for determining fruit quality. Furthermore, sugars serve as the primary source of energy [53]. Another key role of silicon is to enhance the metabolism of fruit sugars, leading to an increase in raffinose, sucrose, and soluble sugar content, thereby raising the TSS of the fruit [54]. Lalithya et al. [43] also recorded higher TSS in sapota with the foliar application of potassium silicate. Zinc increases the TSS content in peach fruits as it contributes to the conversion of polysaccharides into simple sugars [37]. The increase in total soluble solids and total sugars with zinc sulphate might be due to the fact that zinc plays a crucial role in photosynthesis, leading to the accumulation of carbohydrates. Additionally, zinc regulates enzymatic activity, including enzymes involved in metabolizing carbohydrates into simple sugars [50]. Chandra and Singh [48] also reported an increase in TSS due to the foliar application of zinc in aonla fruit.

Malic acid contributes to the titratable acidity in peaches and serves as a measure of total acidity. Among other organic acids (fumaric, citric, and quinic acid), malic acid is the primary organic acid present in mature fruits [55]. Silicon-treated fruits exhibited higher TA at the time of harvest, likely due to slower metabolic activity and respiration rates, which also increased shelf life [56]. Furthermore, the application of potassium silicate significantly improved both total acidity and the TSS-to-acid ratio in loquat [57]. The reduction in the acidity of fruit may be attributed to an increase in fruit zinc concentration, which converts malic acid into sugars [37]. Another explanation for the decrease in TA is that zinc application increases TSS content by breaking down polysaccharides into simple sugars [37]. Similar findings were reported by Anees et al. [58], who noted that the application of micronutrients, especially zinc, decreased TA in Valencia oranges and mango cv. Dusehri.

Silicon had an excellent effect on the TSS–acid ratio of fruit. A decrease in the TSS–acid ratio is typically associated with increased maturity. Another reason for the increase in the TSS–acid ratio may be the conversion of starch into sugars, as leaf starch content decreases and sugars accumulate in the fruit. This gradual decrease in acidity, along with the increase in TSS, raises the TSS–acid ratio of fruits [59]. El-Kholy et al. [57] also observed increased acidity and a higher TSS–acid ratio in peach and loquat. The increase in the TSS–acid ratio of peaches with zinc application could be due to zinc-enhancing TSS content (Figure 2B) while reducing titratable acidity (Figure 2A) in the fruit. Another explanation for the increased TSS–acid ratio is that zinc contributes to the breakdown of complex sugars into simple sugars, thereby raising TSS and, consequently, the TSS–acid ratio [37].

Additionally, zinc plays a crucial role in photosynthesis, leading to carbohydrate accumulation, and it regulates enzymatic activity, including the enzymes responsible for converting carbohydrates into simple sugars [50], which further elevates TSS and the TSS–acid ratio of the fruit.

Vitamin C (ascorbic acid), a potent water-soluble antioxidant, helps boost the immune system and combat plant diseases. It scavenges reactive oxygen species (ROS) produced in the plant body [60]. Silicon application has been shown to increase vitamin C content in fruit by reducing metabolic activity. The addition of silicon to hydroponic solutions enhanced vitamin C levels in tomato fruit [61]. In apples, silicon application increased soluble solids and ascorbic acid content, though it did not affect fruit firmness [62]. Potassium silicate applications at 2% improved various chemical characteristics of peach fruits, such as SSC, acidity, SSC–acid ratio, and vitamin C content [57]. Zinc detoxifies ROS, mitigating the damage caused by membrane sulfides and lipids [63]. Furthermore, zinc positively influences vitamin C levels and the antioxidant capacity of fruits by activating enzymes that strengthen the plant's immune system [64]. Similar results were reported by Aglar et al. [65] in Zn (0.3% $ZnSO_4$)-treated Jersey Mac apples.

Silicon increases reducing sugars by enhancing plant tolerance to various stresses and boosting the antioxidant defense system. Additionally, silicon plays a key role in the biosynthesis of organic compounds, which contributes to the increase in reducing sugars [66]. Martichenkov and Calvert [54] reported that the application of silicon increased reducing sugars, soluble sugars, sucrose, and raffinose in citrus. Similarly, Ahmed et al. [66] observed an increase in TSS, reducing sugars, non-reducing sugars, acidity, and total sugars in date palm through the foliar application of potassium silicate.

Sugars produced during photosynthesis are transported to sink organs as sucrose and sorbitol. In fruit, sucrose and sorbitol are transformed into reducing sugars, such as fructose and glucose, by various enzymes (e.g., AI, NI, SS, SDH, and SOX) [67]. The increase in fruit sugar content is associated with higher zinc levels due to zinc's crucial role in sugar synthesis. Zinc facilitates sugar synthesis by activating enzymes like sorbitol oxidase and dehydrogenase, as well as enhancing IAA activity, which ultimately increases both total and reducing sugars in fully matured fruits [68]. Meena et al. [9] also reported an increase in reducing sugars in aonla with foliar zinc application.

Silicon foliar spray increased non-reducing sugars, total sugar, soluble sugars, sucrose, and raffinose with higher levels of silicon [54]. A similar observation was made by Ahmed et al. [66], who reported an increase in non-reducing sugars in date palm with the foliar application of potassium silicate.

The increase in non-reducing sugars from foliar zinc spray may be attributed to its involvement in the photosynthesis of metabolites and the rapid translocation of sugars from other parts of the plant to the developing fruit [69]. Zinc fertilization acts as a cofactor (both regulatory and structural) for various enzymes, including carbonic anhydrase, which plays a key role in enhancing the plant's photosynthetic capacity [68]. Additionally, enzymes such as indole acetic acid, sorbitol oxidase, and dehydrogenase, which are activated by zinc, contribute to the increase in non-reducing sugars in fully matured fruits. The activity of the aldolase enzyme is also positively influenced by zinc fertilization, aiding in the synthesis of assimilates, including total sugars, reducing sugars, and non-reducing sugars [70].

4.4. Phytochemical Attributes

Antioxidant capacity refers to the ability to inhibit the oxidation process, making it a highly desirable property in fruits due to its role in various diseases [71]. The application of silicon regulates antioxidant enzyme activities and maintains reactive oxygen species at non-toxic levels, thereby protecting plants against oxidative damage under stress conditions [72]. Silicon's beneficial influence on enhancing the activities of catalase, peroxidase, polyphenol oxidase, and glutathione reductase, along with its role in eliminating superoxide and hydrogen peroxide, is likely due to its ability to manage oxidative stress by alleviating ion toxicity and the accumulation of nucleoproteins, which contribute to plant resistance against stress factors [73]. Liang et al. [74] strongly suggest that silicon may play a role in the metabolic, physiological, and structural activities of higher plants exposed to abiotic and biotic stresses. Abdelaal et al. [75] recorded an increase in antioxidant content in sweet pepper due to silicon application.

The foliar application of zinc increased the concentrations of N, P, K, Cu, and Zn in the leaves and fruits, while simultaneously reducing sodium concentrations in both [76]. Zinc has the ability to inhibit the uptake, translocation and transport of Na+ ions in plants, thereby improving growth [77]. Zinc application not only enhances nutrient balance but also plays a direct or indirect role in boosting physiological functions and metabolism, which increases the levels of assimilates and their derivative macromolecules, thereby enhancing antioxidant activity [78]. They further reported a significant increase in the antioxidant capacity of eggplant, confirming the present results.

Silicon is known to enhance the activity of reactive oxygen species (ROS)-scavenging antioxidant enzymes, primarily ascorbate peroxidase (APX), superoxide dismutase (SOD), catalase (CAT), monodehydroascorbate reductase (MDHAR), and glutathione reductase (DHAR), thereby protecting cells against oxidative damage [79]. Additionally, silicon increases the content of non-enzymatic antioxidant compounds such as flavonoids and phenols [80]. The regulatory roles of silicon on the concentrations of phenolics and flavonoids vary among different plants. These silicon-mediated increases in flavonoid and phenolic levels contribute to the nutritional quality of fruit. Increased flavonoid content following silicon application has been reported in tomatoes [81].

The application of zinc induced oxidative stress in the shoots, leading to an increase in non-enzymatic antioxidant molecules such as phenolic compounds and flavonoids due to the accumulation of reactive oxygen species (ROS) [82]. Phenolic compounds play a crucial role in the detoxification of ROS [83], as they can directly eliminate active oxygen species primarily due to their redox properties. They act in the absorption and neutralization of free radicals, extinction of singlet and triplet oxygen, or decomposition of peroxides. This behavior explains the high accumulation of phenols and flavonoids in kinnow fruits from plants subjected to zinc treatments [84].

Proline plays a central role in scavenging reactive oxygen species (ROS), maintaining cell redox homeostasis, and supplying energy [85]. Silicon reduces Na^+ uptake while increasing K^+ concentrations and uptake, which enhances the activities of several enzymes, promotes photosynthesis, improves proline and water status, and inhibits chlorophyll degradation [86]. The increase in proline is a typical response that plays a pivotal role in protecting chlorophyll pigments from degradation [87]. An increase in proline content in peas due to silicon application has been reported [86], and similar results were found in sorghum following silicon application, as noted by Yin et al. [87]. Proline is an important amino acid that accumulates in plants in response to various environmental stresses [88].

The results indicate that exposure to zinc concentration leads to a significant increase in proline content in the test plants [89].

4.5. Enzymatic Activities

Catalase (CAT) plays a significant role in scavenging free radicals produced during metabolic processes. Superoxide dismutase (SOD) converts OH^- into H_2O_2, which is then converted into H_2O and O_2 by CAT [90]. Silicon helps regulate enzymatic activities and maintains reactive oxygen species levels, thus protecting plants against oxidative damage under stress conditions [72]. Silicon enhances the activity of catalase and promotes the elimination of superoxide and hydrogen peroxide by helping to control oxidative stress through the alleviation of ion toxicity and the accumulation of nucleoproteins, which assist plants in stressful conditions [73]. Abdelaal et al. [75] recorded that the foliar application of silicon successfully up-regulated catalase activity in sweet pepper.

Zinc increased the activities of SOD, CAT, and POD by enhancing cellular protection against ROS. This is achieved through the regulation of antioxidant systems, which include both enzymatic components [e.g., superoxide dismutase (SOD), catalase (CAT), peroxidase (POD), and ascorbate peroxidase (APX)] and non-enzymatic constituents [91]. Similarly, Salim et al. [78] observed an increase in catalase activity in eggplant.

POD is an important antioxidant enzyme with a role similar to that of CAT [92]. One of the beneficial effects of silicon is its ability to trigger a range of natural defenses. For instance, the presence of silicon has been shown to stimulate the activity of compounds such as chitinase, peroxidase, polyphenol oxidases, and flavonoids, all of which protect against fungal pathogens [93]. Silicon also helps alleviate metal toxicity in plants, particularly through the activity of peroxidase [94]. The foliar application of silicon successfully up-regulated peroxidase activity in sweet pepper, as recorded by [75]. Similarly, Vieria et al. [95] reported that silicon significantly increased peroxidase content in citrus.

Zinc application enhances physiological functions and metabolism, which increases the levels of assimilates and their derivative macromolecules, thereby boosting the activity of the antioxidant enzyme peroxidase (POD) [78]. Zinc also increases the activities of SOD, CAT, and POD by enhancing cellular protection against ROS. This protection is typically achieved through the regulation of antioxidant systems, which consist of both enzymatic components [e.g., superoxide dismutase (SOD), catalase (CAT), peroxidase (POD), and ascorbate peroxidase (APX)] and non-enzymatic constituents [91]. Similarly, Salim et al. [78] observed an increase in peroxidase activity in eggplant when sprayed with zinc.

5. Conclusions

This study advances our understanding of the role of micronutrients, particularly silicon and zinc, in enhancing not only the agronomic performance of peach trees but also their resilience to environmental stresses. The findings highlight the pivotal role these elements play in modulating biochemical and physiological processes that are crucial for fruit quality and overall productivity. The demonstrated improvements in fruit attributes and stress-related enzymatic activities suggest that targeted nutrient management can significantly contribute to sustainable fruit production, especially in regions facing similar agro-climatic challenges. These results also highlight the potential for silicon and zinc to be integrated into broader horticultural practices, offering a pathway toward optimizing fruit quality and yield while enhancing the nutritional value and storability of the produce. The implications of this research extend beyond peach cultivation, suggesting avenues for applying these insights to other fruit crops, thereby contributing to the broader goal of sustainable agriculture and food security. Future studies should investigate the molecular mechanisms of silicon and zinc, assess their long-term impacts on soil health, and apply these findings to other fruit crops and diverse environmental conditions to refine nutrient management practices. The only limitation of the study is the availability of zinc and silicon in the soil, as both of these elements are greatly dependent on soil type and pH.

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