

Article



# **Quality Responses of Table Grapes 'Flame Seedless' as Effected by Foliarly Applied Micronutrients**

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**Abstract**: Micronutrient (iron, zinc and boron) deficiencies are a basic and prominent factor affecting grape quality and yield in the Pothwar region. To overcome these deficiencies, different levels of micronutrients were applied foliarly on grapevines at five different berry developmental stages during two consecutive growing seasons (2018 and 2019). The data suggested that foliar treatment of micronutrients significantly increased the yield, number of bunches per vine, bunch weight, yield per vines, bunch length, berry number per cluster, berry diameter, berry weight and cluster compactness. The biochemical quality attributes of berries, including sugars (reducing, non-reducing as well as total sugars), ascorbic acid content, pH and TSS values, were at their highest levels in grapevines supplemented with Fe, Zn and B treatment at 200 ppm, respectively, i.e., the highest concentrations used. Biochemical leaf values, including chlorophyll a and b and leaf micronutrient content (Fe, Zn and B), were also highest in grapevines that were sprayed with Fe, Zn and B at 200 ppm. Overall, the results revealed that the performance of grapevine cv. 'Flame Seedless' growing in agroclimatic conditions of the Pothwar region was improved as a result of the foliar application of Fe, Zn and B at 200 ppm. The results also suggested that a further increase in the concentration of each nutrient might be helpful to obtain berries of improved quantity and quality.

Keywords: fruit quality; grapes; micronutrients; foliar spray; ascorbic acid

# 1. Introduction

Grape (*Vitis vinifera* L.) belongs to the family Vitaceae and is native to the riverbanks of Asia, North Ameria and Europe. It is considered the highest ranked fruit in the world because of its manifold benefits [1] and has gained significant importance due to its high nutritional value, taste, multiple uses and the superior returns obtained by farmers [2]. It is one of the dominant commercial fruit crops of temperate to tropical regions and covers more land than any other fruit, representing more than half of total fruit production of the world [3].

In Pakistan, the production of grapes is gaining attention, where mostly European grapes are being produced because of their harmony with the local climate [4]. Approximately 70% of the country's grapes are produced in Baluchistan, with minor amounts



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**Copyright:** © 2021 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). in other provinces, including Khyber Pakhtunkhwa and Gilgit-Baltistan [4]. In recent years, the Pothwar region (Northern Punjab) has gained momentum in terms of grapevine cultivation. However, the yield of grapes in Pakistan is just four tons per hectare, which is very low when compared with other high-yielding countries including Brazil, which produces more than 25 tons per hectare. This minor level of production as compared to developed countries is due to poor management practices, a poor nutritional profile, the unavailability of improved crop materials and the lack of research work in the region, affecting the production of grapes.

The Pothwar region has extensive land for crop production, with the limitations of low rainfall and less fertile lands. The area is vulnerable to drought, with average rainfall ranging from 30 to 200 mm and with temperatures up to 47 °C. As a result, salts amass over the soil surface, subsequently resulting in soil salinity [5]. Hot summers in the region result in a high rate of evapotranspiration, leading to higher electrical conductivity, i.e.,  $>4 \text{ mScm}^{-1}$  [6]. Sodicity (the deposition of surplus Na<sup>+</sup> as a result of salinity) in soil has several adverse effects on its physiochemical properties, including disruption of the structure, hydraulic properties and nutrient availability of soil [7]. On the other hand, soil fertility is reduced due to continuous cropping without adequate fertilization [8]. Likewise, the loss of nutrients due to soil erosion causes a decline in soil productivity [9]; erosion may affect soil properties including its organic matter, tilth, and water holding capacity, as well as the structure and texture of the soil [10,11]. Soils of the Pothwar region are mostly calcareous in nature (having large concentrations of calcium carbonate) with more than 7.5 soil pH. The reason behind this calcareous soil is that these properties are inherited from their parent soils, i.e., calcareous alluvial and loess [12]. Increases in soil erosion elevate calcareousness, which raises pH [13]. With each increase in unit pH, nutrient availability (especially micronutrients) reduces manyfold [14]. Micronutrients (Cu, Fe, Mn, Ni and Zn) are strongly bound with soil particles at high pH and, as a result, they are unavailable to the plants [15]. Soil erosion also has a negative impact on soil organic matter content. Most of the organic matter loss from fields is associated with eroded sediments [16].

Due to these limitations, the soils of the Pothwar region are deficient in micronutrients, including iron, zinc and boron [11,12]. Therefore, grapevines need proper nutrition management to fulfill their needs. As basic mineral nutrients, micronutrients (Fe, Zn, B, etc.) are also measured as essential nutrients for grapevine growth, metabolism, fruit development and quality because, as a co-factor, they activate many metabolic enzymes [17]. Micronutrients, especially Fe, Zn and B, are essential elements utilized by plants for healthy growth. Each of these elements has a significant role in plant growth [18,19]. Poor soil conditions bind these nutrients, ultimately affecting their availability and uptake. Under deficient conditions, the exogenous supply of these substances through an integrated approach can effectively regulate the viability of plants. These supplements compensate for the reduced supply of nutrients from the soil throughout different stages of development [20].

To overcome these requirements, at present, foliar application of these nutrients is gaining traction because of their multiple benefits, including their immediate effects and the low amount of fertilizer required in the solution [21]. Foliar fertilizers are more effective under high nutrient demands, especially when the soil supply and root uptake may be inadequate [22]. Therefore, in the current study, we planned to investigate the role of foliar sprays of micronutrients on the growth, development, yield and quality of the table grape cv. 'Flame seedless'.

#### 2. Materials and Methods

This experiment was conducted in a private vineyard at 'Muradi Janjeel' Tehsil 'Gujjar Khan', 'Rawalpindi' District (33°14' N, 73°08' E), Pakistan. It is semi-arid during the winter season and sub-humid during the summer season. Sixty to 70% of the rainfall takes place during the rainy season viz. mid-July to mid-September. The physical and chemical properties of the soil are shown in Table 1. This trial was carried out at the facility of the

Department of Horticulture, PMAS-Arid Agriculture University Rawalpindi, Pakistan. Thirty-nine disease-free, 4-year-old, and uniform size grape vines (*Vitis vinifera*) of 'Flame Seedless' were selected for this study. Vines of 'Flame seedless' grapes were planted in east–west direction on the Y-trellis system, with 8 feet vine to vine and 10 feet row-to-row distance. Standard cultural practices were applied to all experimental vines during the year 2018. Micronutrient treatments, i.e., Fe-EDTA (50, 100, 150 and 200 ppm), ZnSO<sub>4</sub> (50, 100, 150 and 200 ppm) and Boric acid (50, 100, 150 and 200 ppm), were sprayed at five different developmental stages viz. (i) before sprouting, (ii) during sprouting, (iii) after 10 days of sprouting, (iv) during blooming, and (v) after 10 days of blooming, i.e., a total number of 5 sprays. Tween-20 was used as surfactant for the sprays. The nutrient solution was sprayed thoroughly on the leaves and branches; hence, 1.5 L of spray per tree were used during the 1st two times, while 3 L per tree were used for the rest of sprays. The plant was sprayed thoroughly.

Soil Characteristics	0–15 cm	30–45 cm
Texture	Silt	loam
pH	$7.94\pm0.20$	$7.98\pm0.09$
ECs dS m <sup>-1</sup>	$0.54\pm0.26$	$0.42\pm0.08$
Organic matter (%)	$0.87\pm0.10$	$0.50\pm0.09$
CaCO <sub>3</sub> (%)	$6.30\pm4.89$	$7.50\pm3.82$
Nitrate-N (mg kg <sup>-1</sup> )	$2.75\pm0.57$	$2.40\pm2.15$
Phosphorus (mg kg <sup>-1</sup> )	$0.20\pm0.26$	$0.32\pm0.13$
Potassium (mg kg <sup>-1</sup> )	$80\pm18$	$77\pm13$
Copper (mg kg <sup>-1</sup> )	$2.71\pm0.42$	$3.36\pm0.93$
Iron (mg kg <sup>-1</sup> )	$4.81\pm0.32$	$3.64 \pm 1.89$
Manganese (mg kg <sup>-1</sup> )	$5.46 \pm 1.24$	$4.32\pm2.79$
Zinc (mg kg <sup>-1</sup> )	$0.28\pm0.05$	$0.19\pm0.75$
Boron (mg kg <sup>-1</sup> )	$0.21\pm0.17$	$0.04\pm0.02$

Table 1. The physical and chemical properties of the experimental soil.

 $\pm$  Standard deviation of the mean.

Grape bunches were taken from all possible locations on grapevines so that the samples represent correct yield and quality attributes.

### 2.1. Morphological Analysis

Different physical parameters were calculated manually, including number of clusters per vine, average bunch weight (g), average bunch length (cm), average berry number per cluster, average berry diameter (mm), and average berry weight (g). Average yield per vine and cluster compactness were calculated by using the following formulas:

Yield per vine = Clusters number per vine × Mean weight of bunches  

$$Cluster compactnes = \frac{Average \ berry \ number \ per \ cluster \ per \ vine}{Average \ cluster \ length}$$
(1)

## 2.2. Biochemical Quality Analysis

A sample of 50 berries was selected from bunches of each replicate and juice was extracted. Chemical attributes such as soluble solids contents (SSC), pH, titratable acidity (TA), ascorbic acid (Vitamin C), reducing sugars, total sugars and non-reducing sugars were determined from the juice. Soluble solid contents were determined [23] using a handheld refractometer (Model: SG-103) at room temperature, while pH was measured with a digital pH meter at  $18 \pm 2$  °C. To determine titratable acidity, extracted juice (10 mL) was mixed with 40 mL distilled water and 4–5 drops of phenolphthalein were added in the

juice. A 10 mL aliquot was placed in a titration flask and titrated against 0.1 normal (N) NaOH until a permanent light pink color appeared. After titration, titratable acidity was calculated by the given formula:

$$T.A (\%) = \frac{NaOH \text{ used } \times \text{ N of } NaOH \times \text{Equivalent weight of Tartaric acid}}{\text{Volume of juice used for titration (ml)}} \times 100$$

Ascorbic acid contents were calculated [24]. Five grams of pulp of grapes from 30 berries were ground, using a mortar and pestle, with 5 mL 1.0% HCL and the mixture was centrifuged for 10 min at 10,000 rpm. Absorbance of the extracted supernatant was noted at a wavelength of 243 nm by a spectrophotometer (sp3000 plus model, Optima Japan). Sugar content (total, reducing and non-reducing sugars) in the fruits was determined [25]. A 10 mL (juice) sample was taken in a 250 mL volumetric flask, to which 100 mL distilled water, 25 mL lead acetate solution (430/1000 mL) and 10 mL of (20%) potassium oxalate were added. In a conical flask, 10 mL of Fehling's (5 mL of both Fehling A and B) solution was taken. Sample aliquots were placed in a burette and left to run dropwise into the conical flask containing Fehling's solution. During titration, slow boiling continued until the appearance of a brick red color appeared again. The reading of the sample aliquot used was noted and the percent of reducing sugar was calculated as below:

Reducing sugar (%) = 
$$6.25 (X/Y)$$

where

X = mL of standard sugar solution reading used against 10 mL Fehling's solution.

Y = mL of sample aliquot used against 10 mL Fehling's solution.

For total sugars, 25 mL of aliquot already prepared for reducing sugars was taken in a 100 mL flask in which 20 mL distilled water and 5 mL of concentrated hydrochloric acid were added to convert non-reducing sugars to reducing sugars. To complete the conversion process, the solution was kept at ordinary temperature for a day. It was then neutralized with about 1 N NaOH solution using 2–3 drops of phenolphthalein as an indicator, and again, neutralized with HCL and made up to the volume of 100 mL with distilled water. The prepared solution was taken in a burette and titrated against 10 mL Fehling's solution to the brick red color end point, using methylene blue as an indicator; the same procedure was followed for calculation of the reducing sugars. Total sugar was calculated by the given formula:

Total sugar (%) = 
$$25 \times (X/Z)$$

where

X = mL of standard sugar solution reading used against 10 mL Fehling's solution.

Y = mL of sample aliquot used against 10 mL of Fehling's solution.

Non-reducing sugars were calculated with the formula given below:

Non-reducing sugars (%) = 
$$[TS (\%) - RS (\%)] \times 0.95$$

#### 2.3. Biochemical Leaf Analysis

Leaf chlorophyll a and b contents were determined [26]. Chlorophyll contents in leaves were determined by extracting an accurately weighted fresh plant leaf sample of 0.5 g in 15 mL acetone. The homogenized sample was centrifuged at 10,000 rpm for 15 min. The supernatant was separated, and 0.5 mL was mixed with 4.5 mL acetone. The solution mixture was analyzed with a spectrophotometer at wavelengths 663 nm and 645 nm (using a spectrophotometer, model: SP-3000 plus, Optima, Japan). The following formulas were used to calculate the chlorophyll a and b content in the leaves.

Chlorophyll a (C a) = 11.75 A663 - 2.350 A645 Chlorophyll b (C b) = 18.61 A645 - 3.960 A663*tag*2 Micronutrient content in the leaves was calculated through the dry ashing method [27]. One gram of powdered sample from each treated grapevine was placed in porcelain crucibles and transferred into a muffle furnace. The furnace was then gradually ignited up to 550 °C and heating was continued for a further 5–6 h after reaching the required temperature. After the designated time, the muffle furnace was switched off and crucibles with white ash were cooled. Cooled ash was dissolved in 5 mL of 2 N HCl and thoroughly mixed with a plastic rod. The total volume of this solution was made up to 50 mL using distilled water. The mixture was allowed to stand for 30 min. After filtering the mixture, the obtained aliquot was analyzed for concentrations of zinc and iron through atomic absorption spectrophotometry (SHIMADZU AA-6300). The obtained results were expressed in ppm.

To find out the concentration of boron in the leaf sample, absorbance was determined using a spectrophotometer at wavelength 420 nm with little modification of dry-ashing. One gram of powdered leaf sample was heated in the muffle furnace up to 550 °C for 6 h to ensure the formation of white ash. Crucibles with ash were taken out and cooled ash was mixed with 5 drops of distilled water as well as 10 mL of 0.36 N H<sub>2</sub>SO<sub>4</sub> solution. The solution was continuously stirred for a few intervals for 1 h. The mixture was filtered through Whatman No. 1 filter paper and the obtained aliquot was visualized under a spectrophotometer.

#### 2.4. Organoleptic Evaluation of Grapes

Organoleptic evaluation for texture, flavor and overall acceptability of the samples was performed by using the Hedonic scale [28].

A panel of five judges was selected on the basis of their consistency and reliability of judgment. This method involved presenting the judges with fruit samples, to assess organoleptic factors. Judges were also allowed to retaste a sample, if required. Judges were advised to score each sample by allotting numbers according to the following scale:

1 = Extremely disliked, 2 = Disliked very much, 3 = Moderately disliked, 4 = Slightly disliked, 5 = Neither liked nor disliked, 6 = Slightly liked, 7 = Moderately liked, 8 = Liked very much and 9 = Liked extremely.

## 2.5. Statistical Analysis

A randomized complete block design was used in the experiment, while a least significant difference test (LSD) at the 5% level of significance was used to compare the means obtained for the treatments used in the experiment [29].

## 3. Results

#### 3.1. Fruit Morphological Parameters

Foliar application of different micronutrients (Fe, Zn and B) on grapevines during two years of study (2018 and 2019) significantly increased almost all yield characteristics compared to the control (Tables 2 and 3). Yield attributes including bunch number per vine, bunch length, berry number per bunch, berry diameter and bunch weight increased significantly at higher concentrations of micronutrients. The highest number of bunches number per vine was observed in 200 ppm B (59), followed by 200 ppm Fe (55), 200 ppm Zn (54), 150 ppm B (54), 150 ppm Zn (52) and 100 ppm B (52) foliar treatment during 2018. Likewise, during 2019, the highest bunch number per vine was observed in 200 ppm Fe (47), followed by 200 ppm Zn and B (45), 150 ppm Fe (44) and 150 ppm Zn and B (43). At the same time, the lowest amount of bunches was produced in untreated grapevines (42 and 35 during the years 2018 and 2019, respectively). The highest bunch length (23.6) was observed in vines sprayed with 200 ppm Fe, followed by 200 ppm B (23.1), 200 ppm Zn and 150 ppm Fe (22.5) during 2018. Similarly, during 2019, foliar treatment of Fe at 200 ppm observed the highest bunch length (24.9), followed by 200 ppm B (24.4), 200 ppm Zn (24.3), and 150 ppm Zn and B (23.6). During both study years, i.e., 2018 and 2019, control grapevines produced smaller bunches (21.3 and 22.7, respectively). The highest berry number per bunch during 2018 was observed in grapevines that were foliarly sprayed with 200 ppm B (172), followed

by 200 ppm Fe (170), 200 ppm Zn, 150 ppm B (164), and 150 ppm Zn (162). However, the highest berry number per bunch during 2019 was observed in 200 ppm foliar treatment of Fe (207), followed by 200 ppm B (204), 200 ppm Zn (202), 150 ppm Zn (192), 150 ppm Fe (188) and 100 ppm Fe (182). With the decrease in nutrient concentration, berry number per bunch also decreased until the lowest amount of berries per bunch was observed in grapevines receiving no supplemental micronutrient doses, i.e., control. Foliar application of micronutrients significantly improved berry size as the highest berry diameter was noted in vines that received foliar treatment of 200 ppm B (14.12 mm) during 2018, followed by 200 ppm Fe (13.9 mm), 150 ppm B (13.71 mm), 150 ppm Fe (13.63 mm) and 200 ppm Zn (13.56). Likewise, during experimental year 2019, the highest berry diameter was observed in grapevines that received 200 ppm Fe (14.93 mm), followed by 200 ppm B (14.86 mm), 200 ppm Zn (14.81 mm), and 150 ppm of Fe and B (14.53 mm), whereas significantly smallest berry diameters were observed in unsprayed vines during 2018 and 2019: 12.92 and 13.83 mm, respectively. The heaviest bunches were produced by 200 ppm Fe foliarly treated grapevines (702 g), followed by 200 ppm Zn and B (683 g), respectively, during 2018. Similarly, observations were recorded during 2019, where 200 ppm foliar application of Fe resulted in the highest bunch weight (686 g), followed by 200 ppm Zn (684 g), 200 ppm B (683 g), 150 ppm Fe (666 g), 150 ppm Zn and B (663 g) and 100 ppm Zn (658 g). During both study years, controlled grapevines produced lighter bunches, with the lowest bunch weights being 628 g and 632 g, respectively. Almost all the parameters discussed above have the lowest results when grapevines received only foliar water treatment (control); however, with the increase in individual foliar treatment of micronutrients Fe, Zn and B at 50, 100, 150 and 200 ppm, these parameters increased gradually until significantly higher values were observed when grapevines were treated with 200 ppm of nutrients (Table 2). Similarly, bunch compactness, berry weight, and yield were significantly higher at higher concentrations of Fe, Zn and B. Grapevines sprayed with micronutrients exhibited compact bunches; the highest bunch compactness values recorded in 2018 were 7.43 and 7.41 at 200 ppm Fe and B, respectively, while comparable results were obtained in 2019 in 200 ppm Fe, Zn and B sprayed grapevines. Similar findings for berry weight were observed. The highest berry weight was observed in the highest doses (200 ppm) of Fe, Zn and B, respectively. During 2019, maximum berry weight was recorded in berries of vines treated with 200 Fe, Zn and B foliar application. Likewise, yield attribute varied significantly among all foliar treatments. Yield per vine increased significantly by application of micronutrients, but substantially higher yields were recorded at terminal concentrations of Fe, Zn and B (200 ppm) compared to small concentrations of micronutrients and control treatments during both years, i.e., 2018 and 2019 (Table 3).

## 3.2. Biochemical Fruit Quality Analysis

Application of foliar micronutrients, viz. Fe, Zn and B, significantly improved almost all fruit quality attributes as compared to the control (Tables 3 and 4). Soluble solids concentration (SSC) is one key factor to judge fruit quality. During two successive study years (2018 and 2019), the highest amounts of TSS, i.e., 15.1 and 15.1, 15.1 and 14.6, 15.1 and 14.6 Brix<sup>o</sup>, were observed in grapevines that received individual foliar treatments of Fe, Zn and B at 200 ppm, respectively. Increase in foliar dose of these micronutrients from control to 200 ppm increased TSS gradually until the highest results were observed in the 200 ppm nutrient treatment.

Trastmente	Bunch Number Per Vine		Bunch Length (cm)		Berry Number Per Bunch		Berry Dia	neter (mm)	Bunch Weight (g)	
ireatilients	2018	2019	2018	2019	2018	2019	2018	2019	2018	2019
Control	$42\pm2.02~e$	$35\pm2.6~{ m c}$	$21.3\pm0.43~\mathrm{c}$	$22.7\pm0.41~d$	$144\pm5.23~\mathrm{b}$	$165\pm6.80~\text{d}$	$12.92\pm0.14~d$	$13.83\pm0.29~\mathrm{c}$	$628\pm14.31~\mathrm{d}$	$632\pm0.11~{\rm c}$
Fe 50 ppm	$44\pm2.02~\mathrm{de}$	$40 \pm$ 3.21 abc	$21.7\pm0.57~\mathrm{abc}$	$23.2\pm0.43bcd$	$152\pm 8.66~\mathrm{ab}$	$171\pm7.85~{ m cd}$	$13.11 \pm 0.17 \text{ cd}$ $14.13 \pm 0.37 \text{ bc}$		$639\pm13.13~{ m cd}$	$641\pm0.13~{\rm c}$
Fe 100 ppm	$45 \pm$ 2.18 cde	$42\pm$ 2.88 abc	$21.9 \pm 0.52$ abc	$23.2\pm0.46$ bcd	$155\pm 8.66~\mathrm{ab}$	$182\pm5.51~\mathrm{bcd}$	$13.27 \pm 0.19$ bcd $14.41 \pm 0.4$ abc		$654\pm11.09~{ m bcd}$	$651\pm0.17~\mathrm{abc}$
Fe 150 ppm	$49~\pm$ 1.76 b–e	$44\pm2.3~ab$	$22.5\pm0.55~ab$	$23.5\pm0.43bcd$	$160\pm 8.66~\mathrm{ab}$	$188\pm 6.06$ a–d	$13.63\pm0.16~\mathrm{abc}$	$13.63 \pm 0.16 \text{ abc}$ $14.53 \pm 0.4 \text{ abc}$		$666 \pm 0.19$ abc
Fe 200 ppm	$55\pm2.3$ ab	$47\pm2.64$ a	$23.6\pm0.46~\mathrm{a}$	$24.9\pm0.69~\mathrm{a}$	$170\pm5.04~\mathrm{a}$	$207\pm6.48~\mathrm{a}$	$13.90\pm0.16~\mathrm{ab}$	$3.90 \pm 0.16 \text{ ab}$ 14.93 $\pm 0.38 \text{ a}$		$686\pm0.16$ a
Zn 50 ppm	$\begin{array}{c} 46 \pm \\ 3.01 \text{ cde} \end{array}$	$37\pm3.17~{ m bc}$	$21.3\pm0.49bc$	$23.1\pm0.44bcd$	$155\pm5.29~\mathrm{ab}$	$169\pm8.95\mathrm{cd}$	$13.21\pm0.24~cd$	$13.21 \pm 0.24 \text{ cd}$ $14.11 \pm 0.28 \text{ bc}$		$646\pm0.11\rm bc$
Zn 100 ppm	49 ± 2.33 b–e	$39 \pm 2.9$ abc	$21.7\pm0.46~\mathrm{abc}$	$23.5\pm0.42bcd$	$159\pm5.19~\mathrm{ab}$	$176\pm10.52~\rm cd$	$13.44\pm0.27bcd$	$3.44 \pm 0.27$ bcd $14.26 \pm 0.23$ abc		$658\pm0.13~\mathrm{abc}$
Zn 150 ppm	$52 \pm$ 2.96 abc	$43 \pm 2.3$ abc	$22.3\pm0.49~\mathrm{abc}$	$23.6\pm0.49~\text{a-d}$	$162\pm5.85~\mathrm{ab}$	$192 \pm 9.29$ abc	$13.43\pm0.26bcd$	$3.43 \pm 0.26$ bcd $14.43 \pm 0.29$ abc		$663\pm0.12~\mathrm{abc}$
Zn 200 ppm	$54 \pm 3.21$ ab	$45\pm2.64$ ab	$22.5\pm0.61~ab$	$24.3\pm0.41~\rm{abc}$	$169\pm6.48~\mathrm{a}$	$202\pm10.21~\mathrm{ab}$	$13.56\pm0.24~\mathrm{abc}$	$14.81\pm0.28~ab$	$683\pm13.29~\mathrm{ab}$	$684\pm0.18~ab$
B 50 ppm	$50 \pm$ 2.18 bcd	$37\pm2.02$ bc	$21.6\pm0.49~\mathrm{abc}$	$23.1\pm0.37~cd$	$153\pm7.21~\text{ab}$	$169\pm10.74~\rm cd$	$13.33\pm0.26bcd$	$14.11\pm0.32bc$	$641\pm11.86~{\rm cd}$	$639\pm0.12~\mathrm{c}$
B 100 ppm	$52 \pm 2.31$ bc	$41\pm$ 2.08 abc	$21.8\pm0.43~\mathrm{abc}$	$23.2\pm0.41bcd$	$158\pm7.23~\text{ab}$	$173\pm9.83~{ m cd}$	$13.47\pm0.25bcd$	$14.32\pm0.34~\mathrm{abc}$	$653 \pm 12.41$ bcd	$651\pm0.15~\mathrm{abc}$
B 150 ppm	$54\pm2.33~ab$	$43\pm2.02~ab$	$22.3\pm0.43~abc$	$23.6\pm0.51~\text{a-d}$	$164\pm8.08~\mathrm{ab}$	$181\pm8.76~{\rm bcd}$	$13.71\pm0.26~\mathrm{abc}$	$14.53\pm0.32~\text{abc}$	$665\pm12.03~\mathrm{bc}$	$663\pm0.16~\mathrm{abc}$
B 200 ppm	$59\pm2.61$ a	$45\pm20.8~ab$	$23.1\pm0.49~\mathrm{a}$	$24.4\pm0.41~\text{ab}$	$172\pm 6.69$ a	$204\pm 6.35~ab$	$14.12\pm0.28~\mathrm{a}$	$14.86\pm0.31~\mathrm{ab}$	$683\pm11.26~\mathrm{ab}$	$683\pm0.23~ab$
LSD	7	7	1.5	1.4	20	25	0.64	0.78	36	39

Table 2. Effect of foliar application of micronutrients on yield and physical characteristics of grapes cv. 'Flame Seedless' during the years 2018 and 2019.

Treatments	Bunch Compactness		Berry Weight (g)		Yield per	Yield per Vine (kg)		Soluble Solids (brix°)		Titratable Acidity (%)	
meatments	2018	2019	2018	2019	2018	2019	2018	2019	2018	2019	
Control	$6.39\pm0.14~d$	$7.11\pm0.36~{\rm f}$	$2.03\pm0.12~d$	$2.16\pm0.11~\mathrm{c}$	$27.2\pm1.04~d$	$20.9\pm1.44~\mathrm{e}$	$13.1\pm0.29~g$	$13.2\pm0.28~\mathrm{e}$	$1.41\pm0.09~\mathrm{a}$	$1.35\pm0.09~\mathrm{a}$	
Fe 50 ppm	$6.46\pm0.13~cd$	$7.39\pm0.21~def$	$2.11\pm0.11~cd$	$2.26\pm0.13~bc$	$30.1\pm1.14$ a–d	$23.2\pm1.61~\text{cde}$	$13.1\pm0.26~\text{fg}$	$13.4\pm0.27~\mathrm{e}$	$1.24\pm0.08~\text{ab}$	$1.26\pm0.08~ab$	
Fe 100 ppm	$6.68 \pm 0.24$ bcd	$\begin{array}{c} 7.81 \pm \\ 0.08 \ \mathrm{bcd} \end{array}$	$2.31\pm0.15\text{a-d}$	$2.48\pm0.17~\rm{abc}$	$30.6\pm1.47$ a–d	$24.5\pm1.51~\text{b-e}$	$13.8\pm0.25~\text{c-f}$	$13.5\pm0.22~\mathrm{de}$	$1.08\pm0.09 \text{bc}$	$1.15\pm0.09~\mathrm{abc}$	
Fe 150 ppm	6.78 ± 0.36 bcd	$8.02\pm$ 0.11 abc	$2.39\pm0.13~\text{a-d}$	$2.56\pm0.19~\text{abc}$	$31.4\pm1.79$ a–d	$25.3\pm1.27~bcd$	$14.4\pm0.28~{\rm bc}$	$14.2\pm0.28bcd$	$0.96\pm0.09~\rm{cd}$	$0.97\pm0.09~\text{cde}$	
Fe 200 ppm	$7.43\pm0.12~\text{a}$	$8.32\pm0.03~ab$	$2.61\pm0.12~\mathrm{a}$	$2.85\pm0.16~\text{a}$	$33.8\pm1.58~\mathrm{a}$	$28.1\pm1.1~\mathrm{ab}$	$\pm 1.1 \text{ ab}$ 15.1 $\pm 0.2 \text{ a}$ 15.1 $\pm 0.2$		$0.76\pm0.08~d$	$0.72\pm0.09~de$	
Zn 50 ppm	$6.62 \pm 0.18$ bcd	$6.96\pm0.21~\mathrm{f}$	$2.13\pm0.11~\text{cd}$	$2.27\pm0.11~bc$	$30.1\pm1.71$ a–d	$22.5\pm1.27~\mathrm{de}$	$13.5\pm0.28~\text{d-g}$	$13.3\pm0.25~\mathrm{e}$	$1.19\pm0.07~\mathrm{abc}$	$1.21\pm0.09~\mathrm{abc}$	
Zn 100 ppm	$6.74 \pm 0.17$ bcd	$7.22\pm0.18$ ef	$2.21\pm0.1~\text{bcd}$	$2.41\pm0.13~\mathrm{abc}$	$29.5 \pm 1.32$ a–d	$24.2\pm1.67be$	$13.8\pm0.25cde$	$13.4\pm0.25~\mathrm{e}$	$1.09\pm0.07\mathrm{bc}$	$1.09\pm0.09~\mathrm{abc}$	
Zn 150 ppm	$6.85\pm$ 0.19 a–d	$\begin{array}{c} \textbf{7.61} \pm \\ \textbf{0.15} \text{ cde} \end{array}$	$2.34\pm0.15\text{a-d}$	$2.54\pm0.12~\text{abc}$	$30.7\pm1.68$ a–d	$24.2\pm1.54~\text{b-e}$	$14.1\pm0.23~\text{cd}$	$14.2\pm0.26bc$	$0.95\pm0.08~\text{cd}$	$1.01\pm0.09~\rm{bc}$	
Zn 200 ppm	$7.21\pm0.2~ab$	$8.05 \pm 0.16$ abc	$2.54\pm0.17~ab$	$2.75\pm0.18~\mathrm{a}$	$32.9\pm1.39~\mathrm{ab}$	$28.5\pm1.52~\mathrm{ab}$	$15.1\pm0.21~\mathrm{ab}$	$14.6\pm0.28~\text{ab}$	$0.71\pm0.07~\mathrm{d}$	$0.69\pm0.09~\mathrm{e}$	
B 50 ppm	$6.71\pm0.2~bcd$	$7.03\pm0.19~\text{f}$	$2.12\pm0.13~cd$	$2.27\pm0.12bc$	$27.5\pm1.1~\text{d}$	$23.1\pm1.56~\text{cde}$	$13.2\pm0.24~efg$	$13.3\pm0.25~\mathrm{e}$	$1.21\pm0.09~abc$	$1.25\pm0.08~\text{abc}$	
B 100 ppm	$6.86\pm0.2$ a–d	$7.51\pm0.21~\text{c-f}$	$2.35\pm0.11~\text{a-d}$	$2.42\pm0.15~abc$	$28.4\pm1.3~cd$	$25.8\pm1.45bcd$	$13.7\pm0.29~defg$	$13.6\pm0.21~\text{cde}$	$1.09\pm0.08~bc$	$1.14\pm0.09~abc$	
B 150 ppm	$7.06 \pm 0.19  ext{ abc}$	$7.83 \pm 0.21$ bcd	$2.40\pm0.14~\mathrm{abc}$	$2.53\pm0.16~\mathrm{abc}$	$29.1\pm1.55~bcd$	$27.3\pm1.57~\mathrm{abc}$	$14.1\pm0.21~\rm cd$	$14.2\pm0.23bcd$	$0.96\pm0.09~{ m cd}$	$0.98 \pm 0.08 \text{cd}$	
B 200 ppm	$7.41\pm0.18$ a	$8.42 \pm 0.23$ a	$2.64\pm0.13~\mathrm{a}$	$2.71\pm0.23$ ab	$32.3\pm1.38~\text{abc}$	$30.9\pm1.64~\mathrm{a}$	$15.1\pm0.23~\mathrm{ab}$	$14.6\pm0.29~\mathrm{ab}$	$0.76\pm0.09~d$	$0.70 \pm 0.09$ de	
LSD	0.61	0.56	0.37	0.47	4.45	4.39	0.703	0.711	0.259	0.282	

Table 3. Effect of foliar application of micronutrients on different yield and quality characteristics of grapes cv. 'Flame Seedless' during the years 2018 and 2019.

Treatments	р	н	Ascorbic Acid (mg/100g)		Non-Reduci	Non-Reducing Sugar (%)		Reducing Sugar (%)		Total Sugar (%)	
incutificatio	2018	2019	2018	2019	2018	2019	2018	2019	2018	2019	
Control	$3.29\pm0.17~\mathrm{e}$	$3.43\pm0.19~d$	$3.89\pm0.31~\text{f}$	$3.58\pm0.14~g$	$1.14\pm0.08~g$	$1.19\pm0.17~g$	$8.2\pm0.17~g$	$8.1\pm0.21~\text{h}$	$9.4\pm0.12~\text{h}$	$9.3\pm0.37~g$	
Fe 50 ppm	$3.44\pm0.18~de$	$3.56\pm0.16~\text{cd}$	$4.42\pm0.26~\text{ef}$	$4.05\pm0.19~\text{fg}$	$1.38\pm0.07~\text{fg}$	$1.68\pm0.12$ ef	$9.2\pm0.22~\text{f}$	$8.5\pm0.19~\text{gh}$	$10.6\pm0.14~efg$	$10.3\pm0.32~\text{f}$	
Fe 100 ppm	$3.64 \pm 0.19$ cde	$3.86\pm0.21~\text{cd}$	$5.01\pm0.16~\rm cd$	$4.93\pm0.12~\mathrm{de}$	$1.55\pm0.04\text{ef}$	$1.94\pm0.03~{\rm cde}$	$9.8\pm0.13~de$	$9.1\pm0.26~def$	$11.4\pm0.08~{\rm cd}$	$11.9\pm0.29~bcd$	
Fe 150 ppm	3.99 ± 0.14 bcd	$4.02\pm$ 0.19 bcd	$5.59\pm0.14~\mathrm{ab}$	$5.63\pm0.21~\rm{abc}$	$1.82\pm0.1~\rm cd$	$2.08\pm0.08~\text{abc}$	$10.4\pm0.18~\mathrm{abcd}$	$9.5\pm0.21~\mathrm{cde}$	$12.3\pm0.29~\mathrm{b}$	$12.2\pm0.27~\mathrm{abc}$	
Fe 200 ppm	$4.48\pm0.19~\text{ab}$	$4.52\pm0.21~ab$	$6.05\pm0.15~\mathrm{a}$	$6.11\pm0.15~\mathrm{a}$	$2.23\pm0.06~ab$	$2.39\pm0.11~\mathrm{a}$	$10.8\pm0.17~\mathrm{a}$	$10.1\pm0.18~\mathrm{ab}$	$13.2\pm0.22~\mathrm{a}$	$12.7\pm0.21~\mathrm{ab}$	
Zn 50 ppm	$3.46\pm0.18~\mathrm{de}$	$3.63\pm0.19~cd$	$4.14\pm0.12~\text{ef}$	$4.12\pm0.12~\text{f}$	$1.34\pm0.11~\text{fg}$	$1.63\pm0.17~\mathrm{f}$	$8.8 \pm 0.19 \ \text{fg} \qquad \qquad 8.8 \pm 0.19 \ \text{fg}$		$10.2\pm0.29~\mathrm{fg}$	$11.1\pm0.33~\mathrm{de}$	
Zn 100 ppm	$3.68\pm$ 0.18 cde	$3.92\pm0.19~\text{cd}$	$4.62\pm0.15 de$	$4.92\pm0.16~\mathrm{de}$	$1.85\pm0.12~\rm cd$	$1.99\pm0.07~\rm{cd}$	$9.2\pm0.23~\text{ef}$	$9.2\pm0.23~def$	$11.2\pm0.34~\mathrm{de}$	$11.7\pm0.31~\rm cd$	
Zn 150 ppm	$3.92\pm0.17~\rm cd$	$3.99\pm$ 0.18 bcd	$5.22\pm0.18\mathrm{bc}$	$5.42 \pm 0.21$ bcd	$2.05\pm0.1bc$	$2.02 \pm 0.07$ bcd	$9.9\pm0.24cd$	$10.1\pm0.21~\mathrm{abc}$	$12.1\pm0.27\mathrm{bc}$	$12.6\pm0.28~\mathrm{ab}$	
Zn 200 ppm	$4.51\pm0.2~\text{ab}$	$4.63\pm0.21~\text{a}$	$5.95\pm0.18~\mathrm{a}$	$5.77\pm0.21~\mathrm{ab}$	$2.34\pm0.11~\text{a}$	$2.31\pm0.16~ab$	$10.6\pm0.25~ab$	$10.4\pm0.17~\mathrm{a}$	$13.1\pm0.37~\mathrm{a}$	$10.5\pm0.35$ ef	
B 50 ppm	$3.45\pm0.18~\mathrm{de}$	$3.57\pm0.18~\mathrm{cd}$	$4.13\pm0.12~\text{ef}$	$4.08\pm0.12~\text{fg}$	$1.42\pm0.06~\text{f}$	$1.74\pm0.06~{\rm def}$	$8.5\pm0.24~g$	$9.1\pm0.19~efg$	$10.1\pm0.29~gh$	$11.3\pm0.25~de$	
B 100 ppm	3.83 ± 0.19 cde	$3.88\pm0.21~\text{cd}$	$4.51\pm0.19~{\rm de}$	$4.52\pm0.21~\text{ef}$	$1.56\pm0.05\text{ef}$	$2.13\pm0.05~\mathrm{abc}$	$9.2\pm0.24~\mathrm{f}$	$9.6\pm0.14$ bcd	$10.8\pm0.22~def$	$12.1\pm0.14~\mathrm{abc}$	
B 150 ppm	$4.07 \pm 0.18  ext{ abc}$	$4.11\pm$ 0.21 abc	$5.01\pm0.19 \text{cd}$	$5.13\pm0.17~\rm{cd}$	$1.78\pm0.04~\mathrm{de}$	$2.19\pm0.06~\mathrm{abc}$	$10.2\pm0.23~bcd$	$9.9\pm0.12~\mathrm{abc}$	$12.1\pm0.26\mathrm{bc}$	$12.8\pm0.19~\mathrm{a}$	
B 200 ppm	$4.59\pm0.19~\mathrm{a}$	$4.\overline{67\pm0.19}$ a	$5.87\pm0.19a$	$5.95\pm0.11~\mathrm{a}$	$2.12\pm0.05~ab$	$2.34\pm0.05~a$	$10.5\pm0.22~\text{abc}$	$10.2\pm0.11~\mathrm{ab}$	$12.7\pm0.18~ab$	$1\overline{0.8\pm0.15}$ ef	
LSD	0.551	0.601	0.55	0.51	0.25	0.31	0.59	0.57	0.701	0.83	

Table 4. Effect of foliar application of micronutrients on different quality characteristics of grapes cv. 'Flame Seedless' during the years 2018 and 2019.

TA percentage of juice shows that the highest acidity (1.41% as well as 1.35% during 2018 and 2019, respectively) was observed in grapevines that received no supplemental micronutrients, whereas application of micronutrients significantly reduced acidity percentage, which was observed in 200 ppm of Fe-, Zn- and B-supplemented vines (Table 3).

Table 4 shows that all quality parameters including pH, ascorbic acid and sugars (reducing, non-reducing and total sugar) varied significantly with increase in concentration of foliar nutrients. pH indicates the amount of acid present in the juice, which determines their quality and flavor. More acidic berries (with lower pH) were observed in controlled grapevines, while lower doses of nutrients show little change in acidity as compared to the control but are still statistically insignificant with respect to control. However, application of Fe, Zn and B at 150 and 200 ppm shows a significantly higher level of pH as compared to the control.

Ascorbic acid (AA) concentration in the berries is given in Table 4. Control and 50 ppm foliar treatment of individual nutrient (Fe, Zn and B) concentrations resulted in the lowest values. In contrast, treatment of Fe, Zn and B significantly increased AA concentration with respect to control until the highest amount of AA (6.05/6.11%, 5.95/5.77% and 5.87/5.95% during the two study years 2018/2019, respectively) was observed in 200 ppm foliar treatment of Fe, Zn and B, respectively.

Sugar contents (%) in the berries of different grapevines are given in Table 4. It is obvious from the data that the concentration of micronutrients has a direct relationship with sugar (%), i.e., higher in the higher doses and vice versa. Non-reducing sugars in the berries shows that controlled grapevines have the lowest amount of non-reducing sugars (1.14/1.19% during 2018 and 2019, respectively). In contrast, the highest amounts of non-reducing sugars during the respective years of study 2018 and 2019 were 2.23/2.39% in the case of Fe, 2.34/2.31% in the case of Zn and 2.12/2.34% in the case of B foliar spray at 200 ppm. Further increases in nutrient concentration (Fe, Zn and B) significantly increased the level of non-reducing sugars. Similarly, reducing sugars also show a similar trend in response to foliar application of these nutrients. Controlled grapevines resulted the lowest amount of reducing sugars (8.2/8.1% during 2018 and 2019, respectively), whereas application of Fe, Zn and B at 50 ppm increased the concentration of reducing sugar; these are still statistically insignificant with respect to the control. Further increases in nutrients concentration increased the level of reducing sugars and the highest results (10.8/10.1% by Fe, 10.6/10.4% by Zn and 10.5/10.2% by B during 2018 and 2019, respectively) were observed in grapevines that received 200 ppm of individual nutrients. Total sugars of grapevines also show that application of nutrients significantly increased their concentration with respect to the control. The lowest values (9.4/9.3% during 2018 and 2019, respectively) were observed in the case of the control. However, application of individual nutrients Fe, Zn and B at the rate of 200 ppm observed the highest level of total sugars.

#### 3.3. Biochemical Leaf Analysis

Results of the biochemical leaf analysis are given in Table 5. Foliar application of Fe, Zn and B significantly increased representative mineral nutrient concentration in the leaves. For example, different foliar concentrations of Fe only increased Fe concentration in the leaves without significantly affecting Zn and B concentration in the leaves. From all treatments, the control has the lowest amount of Fe (38.6 and 39.6 ppm during 2018 and 2019, respectively) while application of Fe at 200 ppm resulted in the highest amount of mineral Fe (47.6 and 49.1 ppm during 2018 and 2019, respectively) in the leaves. However, application of Zn and B resulted in a small increase in leaf mineral Fe without any significant change.

Treatments	Iron (ppm)		Zinc (ppm)		Boron (ppm)		Chlorophy	yll a (mg/g)	Chlorophyll b (mg/g)	
ireatificities	2018	2019	2018	2019	2018	2019	2018	2019	2018	2019
Control	$38.6\pm2.33~\mathrm{c}$	$39.6\pm1.45~\mathrm{c}$	$15.3\pm1.45~\text{d}$	$11.1\pm1.15~\mathrm{d}$	$21.1\pm1.15~d$	$22.3\pm0.88~\mathrm{c}$	$2.47\pm0.23~\mathrm{e}$	$2.45\pm0.14~\mathrm{f}$	$1.32\pm0.18~d$	$1.03\pm0.06~\mathrm{e}$
Fe 50 ppm	$41.1\pm2.3~\text{bc}$	$41.1\pm1.15~\mathrm{c}$	$16.1\pm1.15~\mathrm{cd}$	$11.6\pm2.18~cd$	$22.2\pm0.57~d$	$23.3\pm1.45~\mathrm{c}$	$3.3 \pm 1.45 \text{ c}$ $2.64 \pm 0.21 \text{ de}$ $2.67 \pm 0.14 \text{ de}$		$1.53\pm0.19~bcd$	$1.25\pm0.11~\mathrm{de}$
Fe 100 ppm	$42.6\pm$ 2.33 bc	$\begin{array}{c} 43.3 \pm \\ 1.45  \mathrm{bc} \end{array}$	$16.6\pm1.76~{ m cd}$	$13.1\pm1.73~\mathrm{bcd}$	$22.6\pm0.88~\mathrm{d}$	0.88 d 24.6 $\pm$ 1.45 bc 2.91 $\pm$ 0.18 b–e 2.86 $\pm$		$2.86\pm0.14\text{a-f}$	$1.72\pm0.18$ a–d	$2.08\pm0.26~\mathrm{a}$
Fe 150 ppm	$44.3\pm$ 1.76 ab	$46.3 \pm 1.76  ext{ ab}$	$18.6\pm0.88bcd$	$13.6\pm1.76~\mathrm{bcd}$	$24.1\pm1.15~\text{cd}$	$25.3\pm2.18\mathrm{bc}$	$3.37\pm0.19$ ab	$2.93\pm0.12~\text{a-f}$	$1.84\pm0.18~\mathrm{abc}$	$1.85\pm0.11~\mathrm{abc}$
Fe 200 ppm	$47.6\pm2.02~\mathrm{a}$	$49.1\pm1.52~\mathrm{a}$	$19.3\pm1.45bcd$	$15.1\pm1.73~\mathrm{bcd}$	$24.6\pm0.88~\text{cd}$	$26.1\pm1.73\mathrm{bc}$	$3.61\pm0.2~\mathrm{a}$	$3.32\pm0.21~\mathrm{a}$	$2.16\pm0.1~\text{a}$	$1.99\pm0.18~\text{ab}$
Zn 50 ppm	$39.6\pm1.45~\mathrm{c}$	$40.1\pm2.08~\mathrm{c}$	$17.1\pm2.08~\mathrm{cd}$	$13.1\pm1.52~\mathrm{bcd}$	$22.1\pm1.15~d$	$22.6\pm1.45~\mathrm{c}$	$2.63\pm0.15 de$	$2.53\pm0.15~\text{ef}$	$1.43\pm0.14~\text{cd}$	$1.23\pm0.13~\mathrm{de}$
Zn 100 ppm	$40.3\pm1.45$ bc	$40.6\pm1.45~\mathrm{c}$	$20.3\pm1.76~\mathrm{bc}$	$16.2\pm1.52$ bc	$22.6\pm0.88~\mathrm{d}$	$24.1\pm1.73~\mathrm{c}$	$2.85\pm0.18~\text{b-e}$	$2.96\pm0.14~\mathrm{a\text{-}e}$	$1.84\pm0.12~\mathrm{abc}$	$1.43\pm0.14~{\rm cde}$
Zn 150 ppm	$40.6\pm1.2\mathrm{bc}$	$42.3\pm2.6bc$	$22.2\pm1.73~\mathrm{ab}$	$17.3 \pm 1.45$ ab	$23.3\pm1.85~cd$	$24.3\pm1.45~\mathrm{c}$	$3.17\pm0.17$ a–d	$3.09\pm0.16$ a–d	$1.98\pm0.18~\text{ab}$	$1.65\pm0.12$ a–d
Zn 200 ppm	$41.3\pm1.2~{\rm bc}$	$43.6\pm2.33$ bc	$26.6\pm2.6~\mathrm{a}$	$21.6\pm1.45~\mathrm{a}$	$24.1\pm0.57~cd$	$25.1\pm2.08~bc$	$3.23\pm0.2~\text{abc}$	$3.27\pm0.17~\mathrm{ab}$	$2.21\pm0.24~\mathrm{a}$	$1.81\pm0.11~\mathrm{abc}$
B 50 ppm	$39.3\pm2.02~\mathrm{c}$	$40.3\pm1.45~\mathrm{c}$	$15.6\pm1.45cd$	$11.3\pm1.45~cd$	$24.01\pm2.08~cd$	$26.3\pm1.76~\mathrm{bc}$	$2.74\pm0.16~\text{cde}$	$2.74\pm0.14~\text{c-f}$	$1.52\pm0.12~\mathrm{bcd}$	$1.26\pm0.13~\mathrm{de}$
B 100 ppm	$39.6\pm1.45~\mathrm{c}$	$41.1\pm1.73~\mathrm{c}$	$16.1\pm1.52~{ m cd}$	$12.3\pm1.45~cd$	$27.3\pm2.02bc$	$27.1\pm1.73~\mathrm{bc}$	$3.03\pm0.17bcd$	$2.79\pm0.15b\text{-f}$	$1.83\pm0.16~\mathrm{abc}$	$1.51\pm0.15~cd$
B 150 ppm	$\begin{array}{c} 40.3 \pm \\ 2.02 \text{ bc} \end{array}$	$42.6\pm 0.88$ bc	$16.6\pm1.45~\mathrm{cd}$	$13.6\pm1.85~\mathrm{bcd}$	$29.3\pm2.02~\mathrm{b}$	$30.1\pm2.31~\mathrm{ab}$	$3.12\pm0.13$ a–d	$3.09\pm0.14$ a–d	$1.94\pm0.11$ ab	$1.58\pm0.12$ bcd
B 200 ppm	$\begin{array}{c} 41.1 \pm \\ 1.73 \text{ bc} \end{array}$	$\begin{array}{c} 43.6 \pm \\ 1.45 \text{ bc} \end{array}$	$18.6 \pm 0.88$ bcd	$14.1 \pm 1.52$ bcd	$34.3 \pm 1.76$ a	33.3 ± 3.17 a	$3.31 \pm 0.13$ ab	$3.21 \pm 0.19$ abc	$2.07\pm0.14$ a	$1.78\pm0.17~\mathrm{abc}$
LSD	4.57	5.07	4.82	4.72	4.18	5.61	0.54	0.48	0.51	0.45

Table 5. Effect of foliar application of micronutrients on leaf mineral and chlorophyll concentration of grapes cv. 'Flame Seedless' during the years 2018 and 2019.

Foliar application of Zn similarly has no significant effect on Fe and B nutrient concentration in the leaves, whereas Zn mineral nutrient concentration significantly increased only in response to the increase in Zn foliar application with respect to the control. The highest amount of Zn in the leaves (26.6 and 21.6 ppm during 2018 and 2019, respectively) was recorded in 200 ppm Zn foliar treatment, while the lowest amount of Zn (15.3 and 11.1 ppm during 2018 and 2019, respectively) was observed in the case of the control.

Foliar application of B increased Fe and Zn nutrient concentration in the leaves but these are statistically insignificant with respect to the control. However, B mineral concentration changed with the change in the foliar concentration of B. The lowest amount of B leaf concentration (21.1 and 22.3 ppm during 2018 and 2019, respectively) was observed in the control, while the highest amount of B (34.3 ppm and 33.3 ppm during 2018 and 2019, respectively) resulted in 200 ppm B foliarly treated grapevines.

Concentration of chlorophyll (both a and b) in the leaves significantly increased with the increase in foliar concentration of the micronutrients (Table 5). Chlorophyll (a and b) concentration increased with the foliar treatment of Fe. The highest amount of chlorophyll a (3.61 and 3.32 mg/g) was observed as a result of 200 ppm Fe foliar spray, 3.23 and 3.27 mg/g as a result of 200 ppm Zn and 3.12 and 3.09 mg/g at 200 ppm B foliar spray during 2018 and 2019, respectively. Similarly, the highest concentration of chlorophyll b (2.16 and 1.99 mg/g) was observed as a result of 200 ppm Fe foliar spray, 2.21 and 1.81 mg/g as a result of 200 ppm Zn and 2.07 and 1.78 mg/g at 200 ppm B foliar spray. The lowest amounts of both chlorophyll a and b were observed in controlled grapevines. At starting doses of foliar micronutrient (Fe, Zn and B) spray, i.e., at 50, 100 and 150 ppm, both chlorophyll a and b concentration increased without any significant difference, but increasing the dose up to 200 ppm significantly increased chlorophyll concentration during the two study years (Table 5).

#### 3.4. Organoleptic Evaluation

Organoleptic evaluation of fruits of grapevines as a result of foliar treatment of different micronutrients Fe, Zn and B is given in Table 6. Flavor, texture, taste and acceptability of fruits were improved statistically, with respect to the control, when foliar treatments with different micronutrients were applied. The highest flavor grades during 2018 and 2019 were obtained in foliar treatment of Fe (8.3/8.3), Zn (8.6/8.6) and B (8.3/8.6) when each was separately applied at 200 ppm. Controlled grapevines have the lowest grades of flavor (5.3/6.1) during both study years. Texture values were also highest (8.6/8.6, 8.3/7.6 and 8.6/8.1) in the case of 200 ppm foliar treatment of Fe, Zn and B, respectively, during both study years. Controlled grapevines were lowest in texture grading (5.6/6.1) during both study years. During both study years, maximum values for taste, i.e., 8.3/8.6, 8.6/8.3 and 8.6/8.1, were recorded in grapevines foliarly treated with 200 ppm Fe, Zn and B, respectively. The lowest values for taste, i.e., 5.6/5.6, were obtained in fruits from controlled grapevines. Similarly, during the two respective study years, acceptability values were highest (8.3/8.3, 8/8.67 and 8/8) were observed in the case of Fe, Zn and B, respectively, whereas, again, controlled grapevines scored the lowest acceptability grades in all treatments.

Treatments	Fla	vor	Tex	ture	Ta	ste	Acceptability		
ireatinents	2018	2019	2018	2018	2019	2019	2018	2019	
Control	$5.3\pm0.33~\mathrm{e}$	$6.1\pm0.57~\mathrm{d}$	$5.6\pm0.57~\mathrm{e}$	$6.1\pm0.66~\mathrm{c}$	$5.6\pm0.33~\mathrm{f}$	$5.6\pm0.33~g$	$5.67\pm0.33~\mathrm{d}$	$5.33\pm0.67~\mathrm{f}$	
Fe 50 ppm	$6.6\pm0.66~\mathrm{cde}$	$6.1\pm0.57~\mathrm{d}$	$6.3\pm0.57~\mathrm{cde}$	$7.1\pm0.33~\mathrm{abc}$	$6.1\pm0.33~\mathrm{ef}$	$6.3\pm0.57~\mathrm{efg}$	$6.67\pm0.88~\mathrm{bcd}$	$5.67\pm0.33$ ef	
Fe 100 ppm	$7.1\pm0.57~ m bcd$	$6.6\pm0.66~{ m cd}$	$7.1\pm0.33~\text{b-e}$	$7.3\pm0.57~\mathrm{abc}$	$7.1\pm0.57~\mathrm{cde}$	$7.1\pm0.57$ c–f	$7\pm0.57$ a–d	$6.67\pm0.88~\mathrm{c-f}$	
Fe 150 ppm	$7.6\pm0.66$ a–d	$7.6\pm0.66~\mathrm{abc}$	$7.6\pm0.57~\mathrm{abc}$	$8.1\pm0.33~\mathrm{ab}$	$7.6\pm0.33$ a–d	$7.6\pm0.33$ a–d	$7.33\pm0.33$ abc	$7.33\pm0.67$ a–d	
Fe 200 ppm	$8.3\pm0.33~\mathrm{ab}$	$8.3\pm0.33$ ab	$8.6\pm0.33~\mathrm{a}$	$8.6\pm0.33~\mathrm{a}$	$8.3\pm0.33~\mathrm{ab}$	$8.6\pm0.33$ a	$8.33\pm0.33~\mathrm{a}$	$8.33\pm0.33~ab$	
Zn 50 ppm	$6.6\pm0.33~\mathrm{cde}$	$6.3\pm0.66~cd$	$6.1\pm0.88~{\rm de}$	$6.3\pm0.57~\mathrm{bc}$	$6.1\pm0.33~\mathrm{ef}$	$6.6\pm0.57$ d-g	$6.33\pm0.33~\mathrm{cd}$	$6.33\pm0.33~def$	
Zn 100 ppm	$7.1\pm0.57~ m bcd$	$7.1\pm0.57~\mathrm{bcd}$	$7.1\pm0.57$ b–e	$7.1\pm0.57~\mathrm{abc}$	$7.3\pm0.57~bcd$	$7.1\pm0.33$ c–f	$6.67\pm0.33$ bcd	$7\pm0.57$ b–e	
Zn 150 ppm	$7.6\pm0.33$ a–d	$7.6\pm0.33~\mathrm{abc}$	$7.6\pm0.88~\mathrm{abc}$	$7.6\pm0.33~\mathrm{abc}$	$8.1\pm0.57~\mathrm{abc}$	$8.1\pm0.57~\mathrm{abc}$	$7.33\pm0.33~abc$	$7.33\pm0.33~\text{a-d}$	
Zn 200 ppm	$8.6\pm0.33$ a	$8.6\pm0.33$ a	$8.3\pm0.33~\mathrm{ab}$	$7.6\pm0.33~\mathrm{abc}$	$8.6\pm0.33~\mathrm{a}$	$8.3\pm0.33$ ab	$8\pm0.57~ab$	$8.67\pm0.33~\mathrm{a}$	
B 50 ppm	$6.3\pm0.33~\mathrm{de}$	$6.3\pm0.33~\mathrm{cd}$	$6.3\pm0.33$ cde	$6.3\pm0.66~bc$	$6.6\pm0.57~\mathrm{def}$	$6.1\pm0.33~{ m fg}$	$5.67\pm0.33~\mathrm{d}$	$5.67\pm0.67$ ef	
B 100 ppm	$7.1\pm0.57~\mathrm{bcd}$	$7.1\pm0.57~bcd$	$7.1\pm0.57$ b–e	$7.1\pm0.57~\mathrm{abc}$	$7.3\pm0.33$ bcd	$7.3\pm0.67be$	$6\pm0.57~{ m cd}$	$6\pm0.57~def$	
B 150 ppm	$8.1\pm0.57~\mathrm{abc}$	$7.6\pm0.33~\mathrm{abc}$	$7.3\pm0.66$ a–d	$7.6\pm0.33~\mathrm{abc}$	$8.1\pm0.33~\mathrm{abc}$	$7.6\pm0.57$ a–d	$7\pm0.57$ a–d	$7.33\pm0.33$ a–d	
B 200 ppm	$8.3\pm0.33~\mathrm{ab}$	$8.6\pm0.33~\mathrm{a}$	$8.6\pm0.57~\mathrm{a}$	$8.1\pm0.33~\mathrm{ab}$	$8.6\pm0.57~\mathrm{a}$	$8.1\pm0.33~\mathrm{abc}$	$8\pm0.57~\mathrm{ab}$	$8\pm0.57~\mathrm{abc}$	
LSD	1.45	1.52	1.45	1.69	1.31	1.31	1.33	1.62	

Table 6. Effect of foliar application of micronutrients on different organoleptic qualities of grapes cv. 'Flame Seedless' during the year 2018 and 2019.

# 4. Discussion

All yield-related attributes significantly increased as a result of foliar application of micronutrients and the highest values were almost always observed in the highest doses of nutrients applied. Increases in bunch number per vine, bunch length, berry number per bunch, berry diameter, and bunch weight might be affected by the induction of flowers into fruits as a result of foliar treatment of nutrients, leading to increased grapevine yield (Tables 2 and 3). Such upturn in function of iron in fruit through different enzyme reactions and chlorophyll amalgamation might have increased photosynthesis. Improved berry diameter could be ascribed to increased chlorophyll content in the leaf, which is associated with a high production of photosynthate in a plant [30]. Zinc (Zn) increases vegetative growth (stem diameter) by synthesizing tryptophan and regulates growth and production of grapevines [31]. Our results can be correlated with previous findings [32–34], where foliar application of Fe, Zn and Br significantly improved for bunches per vine, bunch weight and the quality of grapevines. Increases in bunch weight of grapevines sprayed with B and Zn could be attributed to the increase in berry set and higher number of berries per bunch along with improved cell size [35]. Increases in yield attributes as a result of foliar application of B might be attributed to its synthetic role in different hormones and other metabolic reactions. Previously, foliar application of B increased fruit yield in naval orange [36] and comparative observations regarding B application were also reported in grapevines [31,37], whereas Fe regulated functions directly influencing fruit setting, fruit retention percentage, bunch number per vine, bunch length, berry number per bunch, berry diameter, bunch weight, bunch compactness, berry weight and yield per vine [38,39].

Fruit quality can also be assessed by parameters such as SSC, titratable acidity, firmness, size and color [40]. The gradual increase in TSS as a result of foliar application of B (Table 3) shows their direct relation with TSS, which might be due to B involvement in photosynthesis. Our findings are in agreement with previous experiments on grapevines [41,42], where foliar treatment of B significantly increased TSS level in grape berries. Zn being an essential micronutrient helps in the activation of enzymes (fructose-1 and 6-bis phosphatase) that play an important role in biochemical reactions accumulating sugars in the fruits [43,44]. The present results are supported by previous studies where foliar application of B, Fe, and Zn increased TSS of 'Perlette' grapes, mango, and strawberry fruit, respectively [31,45,46]. The inverse relationship of TA with foliar spray of Fe (Table 3) might be due to an increase in the metabolic rate that increases the conversion of organic acids into lower carbohydrates in the berry solution, resulting in a reduction in the acidity, whereas Zn helps in the translocation of carbohydrates from leaves to fruits that increases the berry quality and quantity with an improved amount of sugar content and reduced titratable acidity by their conversion into sugars. Similar results were reported [46,47] where application of B and other micronutrients (Zn, Fe) resulted in decreased TA in grape and strawberry. Meanwhile, ascorbic acid contents of grape berries were increased with increasing concentrations of micronutrients (Fe, Zn, B) and the maximum value was recorded in berries sprayed with 200 ppm B.

Ascorbic acid is an important dietary ingredient that works as a strong antioxidant as well as helping in the electron transport chain and it regulates enzymatic activities by performing their roles as co-enzymes. In the current study, foliar application of micronutrients (Fe, Zn, B) increased the ascorbic acid contents of grape berries during both growing years (Table 4). B as a micronutrient participates in a variety of biochemical processes and has been reported to increase ascorbic acid contents as it is a fundamental part of important cell structures [48]. Increases in ascorbic acid contents in the berries as a result of supplemental micronutrient treatments could be related to the regulation of various essential metabolic activities and similar findings have been reported in previous studies [49,50]. Our results showed that reducing, non-reducing and total sugar contents of grape berries were increased with increasing concentration of micronutrients (Table 4). Such an increase in sugar contents of grape berries could be related to the increased chlorophyll contents and the higher photosynthesis rate achieved through supplemental sprays of Fe, B and Zn [51]; similar findings were reported in apple and grapevines [52–54]. Higher sugar contents as a result of foliar sprays of micronutrients have also been reported in strawberry, pomegranate, and grapes [55–57].

Grapevines sprayed with supplemental sprays of Fe, Zn, and B exhibited significantly higher values of respective nutrients in the leaves, while each nutrient remained ineffective in increasing the contents of other nutrients in the leaves significantly (Table 5). Though insignificant, higher doses of each nutrient increased the concentration of other nutrients, which could be ascribed to the synergistic effect of these nutrients on each other [58–61].

The results of the present study depicted that grapevines sprayed with micronutrients exhibited higher chlorophyll contents in leaves (Table 5). This increase in chlorophyll content could be owed to the higher nutrient level of Mg, Fe, K and Ca as a result of the application of supplemental micronutrients. Previously, the application of micronutrients increased chlorophyll a and chlorophyll b contents of leaves of peas as well as peace lily [62,63]. Previously, foliar as well as soil application of Fe-EDTA increased chlorophyll a concentration in the foliage of wheat [64], whereas Zn has been proven to be helpful in improving photosynthetic efficiency in plants [65] by stabilizing the activity of carbonic anhydrase, which has a role in the accumulation of chloroplast and chlorophyll synthesis [66]. Likewise, foliar application of B increases the expression of auxin biosynthesis gene BnNIT1 [67], which is related to chlorophyll synthesis [68]. Similar observations were recorded where B application increased chlorophyll contents in olive, cashew, cucumber, and pepper leaves [69–72]. Sensory attributes of flame seedless grapes, including taste, texture, aroma, and acceptability, were improved with increasing concentrations of foliar sprays of micronutrients (Table 6). Zn is believed to be associated with auxin synthesis in plants and plays a vital role in enzymatic reactions that describe the final quality of the fruit. Zn helps in enzymatic reactions that lead to the transformation of carbohydrates, formation of cellulose and change in sugars [73]. Furthermore, Dutta and Dhua [73] observed an improvement in mango sensory attributes by application of micronutrients. Our results are comparable with the findings of Bhoyar and Ramdevputra [74], where the application of Zn, B and Fe increased the overall sensory quality of guava fruit. Similar findings were published [75], observing an improvement in sensory attributes of different pomegranate cultivars by the application of micronutrients.

## 5. Conclusions

The experiment was conducted to improve the quality of "Flame Seedless" table grapes by the foliar application of micronutrients, i.e., Fe, Zn and B, under the arid conditions of Pakistan. The results obtained from the present study proved that foliar application of micronutrients (Fe, Zn, and B) was the most effective treatment in improving the physical and chemical parameters of grape berries. To improve the yield and quality of "Flame Seedless" table grapes under the conditions in Pothwar, micronutrients (Fe, Zn and B) can be applied as foliar spray to the vineyards in order to enhance their efficiency and avoid losses. To our knowledge, this is the first report of micronutrient foliar sprays and fruit quality of grapes under the arid conditions of Pakistan. Thus, keeping in view the trends obtained from the data, further study is recommended with higher doses and combinations of the abovementioned micronutrients.

**Author Contributions:** I.A. conceived the idea. I.A. and M.J.T. conducted the experiment and collected the literature review. W.M.A., M.U.H. and M.S. provided technical expertise to strengthen the basic idea. X.W., A.Q., W.A. and S.F. helped in statistical analysis. S.F., W.A. and A.Q. proofread and provided intellectual guidance. All authors have read and agreed to the published version of the manuscript.

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