

ORIGINAL RESEARCH ARTICLE

Optimizing the Zn, Mn, and Fe mineral dose as tank mix foliar application for improvement of fruit yield, quality, and uptake of nutrients in the kinnow mandarin

Sarvpriya Singh¹, Jagdish Singh^{2,*}, Salwinder Singh Dhaliwal³

¹ Krishi Vigyan Kendra Bathinda, Punjab Agricultural University, Punjab 141027, India

² PAU-Regional Research Station, Gurdaspur 143524, India

³ Department of Soil Science, Punjab Agricultural University, Punjab 141027, India

* Corresponding author: Jagdish Singh, jagdishsingh@pau.edu

ABSTRACT

Kinnow production is hampered due to the lack of micronutrient applications such as zinc (Zn), iron (Fe), and manganese (Mn), which play a significant role in the metabolic activities of the plant, affecting yield and quality. The farmers of the region use mineral micronutrient fertilizers, but it leads to phytotoxicity due to unoptimized fertilizer application dose. In the present investigation, an attempt has been made to optimize the Zn, Mn, and Fe minerals dose as tank mix foliar application for improvement of fruit yield, quality, and uptake of nutrients. The twelve combinations of different doses of zinc sulphate, manganese sulphate, and ferrous sulphate fertilizers replicated three times were tested at kinnow orchards established at Krishi Vigyan Kendra, Bathinda, Punjab, India. The data revealed that the fruit drop was significantly low in the treatment F₁₂ (43.4%) (tank mix spray of 0.3% ZnSO₄ + 0.2% MnSO₄ + 0.1% FeSO₄) compared to control treatment. The fruit yield per tree was significantly higher in the treatment F₁₂ compared to untreated control. The juice percentage was also recorded higher in treatment F₁₂ as compared to control, and the juice percentage improved by 2.6%. The leaf nutrient analysis also revealed translocation of higher amount of nutrient from leaf to fruit under optimized supply of micronutrient. Thus, the application of tank mix spray of 0.3% ZnSO₄ + 0.2% MnSO₄ + 0.1% FeSO₄ may be used for better fruit yield and quality.

Keywords: tank mix; micronutrient; foliar application; kinnow quality; yield

ARTICLE INFO

Received: 20 November 2023

Accepted: 19 December 2023

Available online: 27 December 2023

COPYRIGHT

Copyright © 2023 by author(s).

Trends in Horticulture is published by

EnPress Publisher LLC. This work is licensed

under the Creative Commons Attribution-

NonCommercial 4.0 International License

(CC BY-NC 4.0).

<https://creativecommons.org/licenses/by-nc/4.0/>

1. Introduction

Kinnow mandarin (*Citrus deliciosa* Lour. × *Citrus nobilis* Tanaka) is a nutrient-exhaustive crop right from the beginning to the final stage of fruit harvest. The kinnow production was hampered due to the lack of proper management practices, including micronutrients like zinc (Zn), iron (Fe), and manganese (Mn), which contributed to various metabolic activities in the plant system. Zinc (Zn) plays a pivotal role in the synthesis of auxin, the synthesis of protein, energy generation, structural integrity of bio membrane; transformation and consumption of carbohydrates, formation of fruits, and maturity induction^[1]. Iron (Fe) is involved in the synthesis of chlorophyll content, photosynthesis, mitochondrial respiration, nitrogen assimilation, hormone biosynthesis (ethylene, gibberellin, and jasmonic acid), production and scavenging of reactive oxygen species (ROS), osmo-protection, pathogen defense, cell division, and growth^[2]. Manganese (Mn) contributes to the metabolism of organic acids, nitrogen metabolism, respiration, activator of enzymatic reactions such as oxidation/reduction and hydrolysis,

participation of the oxygen evolution complex in light reactions of photosynthesis, and is available in the form of Mn^{2+} to the plant^[3].

The deficiency of micronutrients alters plant growth and metabolic activity, resulting in the appearance of visual deficiency symptoms in most cases^[4]. The deficiency of zinc (Zn) causes irregular yellowing, rosetting/mottled leaves, and the terminal portion of the leaves becomes narrow and reduces the leaf area, which ultimately inhibits the metabolic and photosynthesis activity due to the various enzymes like alcohol dehydrogenase, glutamic dehydrogenase, and carbonic anhydrase through electron transport and auxin biosynthesis pathways^[3]. Iron (Fe) deficiency symptoms cause yellowing of terminal leaves and premature defoliation. The deficiency symptoms of manganese (Mn) cause leaf lamina to turn a light green colour with fine green veins, and under severe deficiency, light green turns a grey colour. The biochemical changes were also reported, i.e., a reduction in sugar and cellulose level, increased drought sensitivity, and reduced fertility by inhibition of respiration pathway^[5] due to micro-nutrient deficiencies. An adequate supply of micronutrients helps to protect the plant from adverse climatic conditions and biotic factors^[6].

An estimate by Sillanpää^[7] revealed that the world's agricultural soils were deficient in Zn (49%), Mn (10%), and Fe (3%), especially in regions with high pH, low mobility of nutrients, and free $CaCO_3$ in soil. In India, zinc deficiency is due to alkaline soil types; Zn solubility in soils also decreases with a rise in soil pH level, while high soil P content induces the deficiency of Zn^[8]. Iron (Fe) deficiency was also seen in similar regions where Zn deficiency occurred in calcareous soils. As noted above, Fe availability is also strongly influenced by soil pH, while Mn deficiency can occur in coarse textured alkaline soils where it can be leached out of the soils^[5]. There are several options to rectify the micronutrient deficiencies, viz., genetic modifications, application of nutrients directly to soil, and foliar application and mixing of multiple nutrients in single spray, but in fruit plants, the supplementation of micronutrients through foliar application is viable option for improving the yield and quality of the produce^[3]. The main advantage of foliar sprays is that they get instant results with more efficacy, save on input costs, use fewer fertilizers, reduce environmental load. Foliar feeding has been used as a means of supplying supplemental doses of minor and major nutrients, plant hormones, stimulants, and other beneficial substances. Observed effects of foliar fertilization have included yield increases, resistance to disease and insect pests, improved drought tolerance, and enhanced fruit quality^[9-11]. Application of some nutrients (not all) through foliage can be 10 to 20 times as efficient as soil application^[12]. However, this efficiency is not always achieved in actual practice due to weather extremes, application of the wrong spray mix, or application of the right mix at the wrong time^[13-16]. Foliar fertilizations are often timed to coincide with specific vegetative or fruiting stages of growth, and the fertilizer formula is adjusted accordingly. Therefore, judging what foliar materials to apply and at what plant stage to spray with soil applied organic and inorganic fertilizers are important principles to make the best use of this technique^[2]. A properly formulated foliar spray, particularly one amended with appetizers/bioactive materials/bio-stimulants and surfactants, increases the uptake of nutrients from the soil^[14] because foliar fertilization causes the plant to pump out more sugars and other exudates from its roots into the rhizosphere. The use of multi-nutrient plant growth regulator formulations amended with appetizers could be a new and innovative approach to developing a cost-effective foliar spray for improving crop yields^[7].

The kinnow growers are already using the foliar application of micro-nutrient mixtures; however, the dose of the micro-nutrient mixture has yet to be optimized for better yield and return without any effect on plants. Thus, the present investigation has been planned to assess the impact of foliar application of different micro-nutrient mixtures on the yield and quality of kinnow.

2. Material and methods

2.1. Experimental site and climatic condition

The experiment was initiated during 2020–2021 on twelve to thirteen year old kinnow orchard of Punjab Agricultural University, Krishi Vigyan Kendra, Bathinda (semi-arid) district of Punjab, India. The study location was far away from *shivallik hill* and closest to *Thar Desert* of Rajasthan, and is characterized by tropical *steppee*, semi-arid and hot climate during summers with scorching heat and cold winter^[17]. The south-west *monsoon* sets in second fortnight of June and towards end of September. The average annual rainfall was unevenly distribution in this region. The daily variation in maximum temperature (T_{max}), minimum temperature (T_{min}), and rainfall (mm) during the two year of study was illustrated in **Figure 1**.

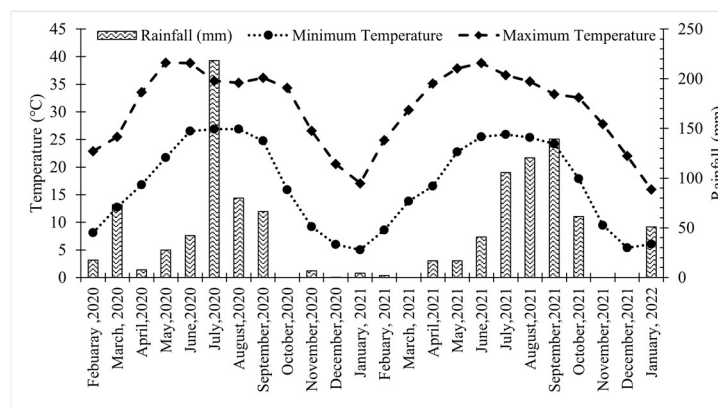


Figure 1. The data represents the maximum temperature (T_{max}), minimum temperature (T_{min}), and rainfall (mm).

2.2. Analysis of soil samples for basic properties

The texture of soil was sandy loam and having high pH level and due to high soil pH, the deficiency of micronutrients appeared in the kinnow orchard every year. The profile of the soil having physico-chemical properties was illustrated in **Table 1**. The physico-chemical characteristics of the soils were determined by adopting standard procedures. Mechanical analysis of soil was carried out by International Pipette Method^[18]. The pH and electrical conductivity were determined in 1:2 soil water suspensions by using Elico glass electrode pH meter and solubridge conductivity meter, respectively. Organic carbon was determined by rapid titration method described by Walkley and Black^[19]. Calcium carbonate was estimated by the methodology outlined by Puri^[20] using bromothymol blue and bromocresol green as indicators. Available phosphorus was extracted according to the method described by Olsen et al.^[21] whereas, available potassium was estimated by the method used by Jackson^[22] and Merwin and Peech^[23].

Table 1. Basic soil properties of experimental area from soil depth of 0–15 and 15–30 cm.

Soil properties	Soil depth (cm)		Reference
	0–15	15–30	
Soil texture	Sandy Loam	Sandy Loam	Jackson (1973)
pH	8.30	8.05	Jackson (1973)
EC ($dS\ m^{-1}$)	0.45	0.40	Jackson (1973)
OC	0.35	0.31	Walkley and Black (1934)
CaCO ₃ (%)	1.10	2.25	Puri (1949)
Total N (%)	0.30	0.25	Subbiah and Asija (1956)
Available P (%)	6.34	4.87	Olsen et al. (1954)
Available K (%)	157.4	121.7	Merwin and Peech (1950)
DTPA–Extractable Zn ($mg\ kg^{-1}$)	0.42	0.32	Lindsay and Norvell (1978)
DTPA–Extractable Fe ($mg\ kg^{-1}$)	1.61	0.91	Lindsay and Norvell (1978)
DTPA–Extractable Mn ($mg\ kg^{-1}$)	103.7	107.8	Lindsay and Norvell (1978)

The soil of the experimental site was sandy loam in texture and contained nitrogen at 220 kg ha⁻¹ available N^[24], phosphorus at 11.70 kg ha⁻¹ available P, potassium at 159.10 kg ha⁻¹ available K and iron at 5.60 mg kg⁻¹ available Fe.

2.3. Experiment details

The experiment was conducted to optimize the foliar application mix in different combinations. The various combinations of zinc sulphate (ZnSO₄.7H₂O), manganese sulphate (MnSO₄.5H₂O) and ferrous sulphate (FeSO₄.7H₂O) mineral fertilizers was applied through power sprayer during the month of April and August when the new flush of leaves was matured and achieved proper shape and size. The thirteen different treatments combination inclusive of one untreated of foliar application (control) were sprayed in ten fruit plants each experiment (**Table 2**). The units of treatment combination for spray application were expressed in percentage (%).

Table 2. Experimental treatment detail of tank-mix multi micro-nutrients mixture for application to kinnow tree.

Treatment	Tank mix spray combinations
F ₀	Control (No micronutrients application)
F ₁	0.4% ZnSO ₄ + 0.3% MnSO ₄
F ₂	0.4% ZnSO ₄ + 0.2% MnSO ₄
F ₃	0.3% ZnSO ₄ + 0.3% MnSO ₄
F ₄	0.3% ZnSO ₄ + 0.2% MnSO ₄
F ₅	0.4% ZnSO ₄ + 0.3% MnSO ₄ + 0.2% FeSO ₄
F ₆	0.4% ZnSO ₄ + 0.2% MnSO ₄ + 0.2% FeSO ₄
F ₇	0.3% ZnSO ₄ + 0.3% MnSO ₄ + 0.2% FeSO ₄
F ₈	0.3% ZnSO ₄ + 0.2% MnSO ₄ + 0.2% FeSO ₄
F ₉	0.4% ZnSO ₄ + 0.3% MnSO ₄ + 0.1% FeSO ₄
F ₁₀	0.4% ZnSO ₄ + 0.2% MnSO ₄ + 0.1% FeSO ₄
F ₁₁	0.3% ZnSO ₄ + 0.3% MnSO ₄ + 0.1% FeSO ₄
F ₁₂	0.3% ZnSO ₄ + 0.2% MnSO ₄ + 0.1% FeSO ₄

Zinc sulphate—ZnSO₄.7H₂O, Manganese sulphate—MnSO₄.5H₂O, Ferrous sulphate—FeSO₄.7H₂O

2.4. Orchard management

The application of 80 kg plant⁻¹ farmyard manure (FYM) was applied in the month of December and half dose of nitrogen (200–400 gm plant⁻¹), full dose phosphorous (1940 gm plant⁻¹), potassium (880 gm plant⁻¹) applied in the month of February and second dose of N (200–400 gm plant⁻¹) half in the month of April–May after fruit set and the irrigation was applied through drip irrigation as per recommendation of Punjab Agricultural University (PAU), Ludhiana, Punjab, India^[25].

2.5. Leaf sampling and analysis

The leaf sampling was done in the month October from the branches immediate behind the fruit. During time of leaf sampling 4–6 leaves were randomly selected from each direction (north, south, east and west) at 1–2 m height of the fruit plant. The analysis of leaf was done in the department of soil science; Punjab agricultural University, PAU, Ludhiana and Punjab for estimation the leaf nutrient content after two sprays of foliar application in different combination as mentioned above. The leaf samples were washed, decontaminated and then dried with hot air oven at 70 °C. The dried samples were grinded with stainless steel blades by passing through 40 mesh sieves. The leaf N content was estimated with micro-Kjeldahl method^[26], phosphorus by

vanadomolybdate-phosphoric yellow colour method^[27]. The leaf potassium (K), zinc (Zn), iron (Fe), and manganese (Mn) were estimated with atomic absorption spectrophotometer^[28].

2.6. Mandarin fruit and yield parameters

The fruit drop in kinnow crop was divided into three distinct stages which included first wave-post bloom drop, second wave-June drop and third wave-pre-harvest drop. The fruit drop (%) at various stages was calculated by counting the number of fruits dropped from the tagged fruit plants. For the purpose, the branches were tagged from four sides of fruit plants and estimated at different stages^[29] by using Equation (1).

$$\text{Fruit drop (\%)} = \frac{\text{Total fruit drop}}{\text{Total fruit set}} \times 100 \quad (1)$$

The twenty fruits were harvested from different treatments in the second fortnight of January to last harvest for measuring fruit length, diameter, peel thickness using digital vernier's caliper and expressed in mm. The fruit weight, peel weight, rag weight, juice weight and seed weight were measured with electronic weighing balance and expressed as g fruit⁻¹. The percentage of rag weight, juice weight, peel weight and seed weight were calculated as per Equations (2–5). Total fruit yield was estimated at the time of harvesting.

$$\text{Rag weight (\%)} = \frac{\text{Average total rag weight}}{\text{Average fruit weight}} \times 100 \quad (2)$$

$$\text{Juice content (\%)} = \frac{\text{Average juice weight}}{\text{Average fruit weight}} \times 100 \quad (3)$$

$$\text{Peel weight (\%)} = \frac{\text{Average total peel weight}}{\text{Average fruit weight}} \times 100 \quad (4)$$

$$\text{Seed weight (\%)} = \frac{\text{Average total seed weight}}{\text{Average fruit weight}} \times 100 \quad (5)$$

2.7. Determination of DTPA-available Zn, Mn and Fe in soil

The soil samples collected from the kinnow fruit (Mandrin) crop were air-dried, ground, passed through 2 mm sieve. DTPA-extractable Zn, Mn and Fe (mg kg⁻¹) was assessed by extracting 10 gm portion of soil sample with 20 mL of diethylene triamine penta acetic acid (DTPA) extractant (0.005 M DTPA + 0.01 M CaCl₂ + 0.1 M TEA buffer adjusted to pH 7.30) as described by Lindsay and Norvell^[30].

DTPA-Extractable Zn, Mn and Fe (mg kg⁻¹) in soil = Concentration of extractant on AAS × dilution factor of 2.

2.8. Determination of Zn, Mn and Fe content in plant parts

For estimation of Zn, Mn and Fe content, 0.5 g of leaf sample were digested using diacid mixture (HNO₃: HClO₄ = 4:1) as per method given by Miller and Keeney^[28] and their content in the digests after proper dilution were determined. After appropriate dilution, the concentration of Fe was measured on an atomic absorption spectrophotometer (AAS 240, FS-Varian Model).

Plant concentration of Fe (mg kg⁻¹) = Concentration of extractant on AAS × dilution factor

2.9. Statistical analysis

The Shapiro and Wilk test of normality^[31] was employed to test the homogeneity of data. The data was analysed for ANOVA by using complete randomized block design (CRBD) using IBM SPSS for Windows 21.0 (IBM SPSS 21.0, Inc., Chicago, U.S.A.). The mean comparison was made using Duncan Multiple Range Test where F test was found significant at $P < 0.05$.

3. Results and discussion

3.1. The effect of micro-nutrient application on the fruit drop of kinnow mandarin

The fruit drop is affected by a number of biotic and abiotic factors, such as soil nutrition, moisture regimes, weather, cultivar, etc. The data on fruit drop during first wave post bloom period revealed that the least fruit drop was recorded significantly low in the treatment F₁₂ (15.0%) among the other combinations of Zn, Mn and Fe and compared to control treatment (**Table 3**). The fruit drop during first wave in treatment F₁₂ was 41.4% less compared to control (F₁₃). During the second wave of fruit drop in the month of June, least fruit drop (12.5 %) was observed in the treatment F₁₂ (where 0.3% ZnSO₄ + 0.2% MnSO₄ + 0.1% FeSO₄) and significantly higher in the untreated control treatment F₀ (18.5 %). The fruit drop in the treatment F₁₂ was 32.4% lower than control (F₀), 18.8% lower than F₄ treatment. The third wave fruit drop, i.e., pre harvest fruit drop is generally affect by soil moisture condition, weather parameters, etc., where highest fruit drop was recorded in the treatment F₀ and significantly low in the treatment F₁₂. The fruit drop in F₁₂ was 43.4 %, 12.8%, 26.4% lower than the treatment F₀, F₁₁ and F₄, respectively.

The highest total fruit drop of 82.4% was observed in the untreated control, and least under the F₁₁ treatment. The fruit drop in treatment F₁₁ was 40.3, 10.2, 23.6% less than the treatment F₀, F₁₀ and F₄, respectively (**Table 3**). Though, all the treatments were able to reduce fruit drop significantly compared to untreated control, however, spray of 0.3% ZnSO₄ + 0.2% MnSO₄ + 0.1% FeSO₄ (F₁₁) was most efficient in reducing fruit drop. Zoremfluangi et al.^[32] also reported maximum fruit set and minimum fruit drop when treated with Zn (0.5%) + Cu (0.4%) + B (0.1%) in mandarin. The Zn might have influenced pollination through its effect on pollen tube generation^[33] and also reduced flower drop. Similar results were reported by Nawaz et al.^[34] and Akula et al.^[35], clarified on the significant role of Zn in the auxin synthesis, leading to better photosynthesis, more accumulation of starch in fruits and the auxin producing stability in the plant, which control the fruit drop and improves the number of fruits per tree. Goren et al.^[36] also reported rise in IAA synthesis in plant with the application of Zn that inhibits the ethylene synthesis in the abscission zone which prevent fruit drop. Liaquat et al.^[29] also attributed reduction in pre-harvest fruit drop with the application of zinc in kinnow mandarin crop.

Table 3. Effect of tank-mix multi micro-nutrients mixture application on of kinnow fruit drop. Different letters in each column of experimental factors show significant differences at $P < 0.05$ probability level.

Treatment	First wave-post-bloom drop (%)	Second wave-June drop (%)	Third wave-pre-harvest drop (%)	Total fruit drop (%)
F ₁	22.5 ^b	17.4 ^b	36.4 ^b	76.3 ^b
F ₂	21.4 ^c	16.9 ^{bc}	34.1 ^c	72.4 ^c
F ₃	20.7 ^d	15.8 ^d	30.4 ^d	66.9 ^d
F ₄	19.5 ^e	15.4 ^{de}	29.5 ^e	64.4 ^e
F ₅	21.4 ^c	16.5 ^c	33.8 ^c	71.7 ^c
F ₆	20.5 ^d	15.3 ^{de}	28.7 ^f	64.5 ^e
F ₇	19.0 ^{ef}	15.0 ^e	27.6 ^g	61.6 ^f
F ₈	18.6 ^f	13.9 ^f	26.5 ^h	59.0 ^g
F ₉	20.4 ^d	14.9 ^e	25.9 ^h	61.2 ^f
F ₁₀	17.5 ^g	13.2 ^g	24.1 ^j	54.8 ⁱ
F ₁₁	18.5 ^f	13.4 ^{fg}	24.9 ⁱ	56.8 ^h
F ₁₂	15.0 ^h	12.5 ^h	21.7 ^k	49.2 ^j
F ₀	25.5 ^a	18.5 ^a	38.4 ^a	82.4 ^a
CV	12.4	11.2	16.7	13.9

Notes: ^{a-j}: Each letter represents statistically similarity at $P < 0.05$.

3.2. Effect of micro-nutrient foliar application on fruit physical characteristics and yield at harvest

The fruit diameter is directly influenced by the cultivar, nutrient and moisture supply (**Table 4**). The analysis of the data on the fruit diameter revealed significantly higher fruit diameter (7.72 cm) was recorded in the treatment F₁₂ (where 0.3% ZnSO₄ + 0.2% MnSO₄ + 0.1% FeSO₄), statistically at par with F₁₁, F₁₀ and F₇, compared to control and all other treatments. However, fruit diameter was F₁₂, F₁₁, F₁₀ and F₇ was 24.5, 24.1, 23.7 and 24.0% higher compared to control. Zorentluangi et al.^[32] also reported maximum fruit weight with the treatment involving supply of Zn + B+ Mn. Similarly, Singh et al.^[37] reported improvement in fruit weight of sweet orange with application of Zn + Cu + B. Ilyas et al.^[38] attributed the improvement in fruit weight to the improved photosynthetic attributes of plant, with the foliar application of Zn, B and Cu.

The fruit yield per tree was significantly higher in the treatment F₁₂ compared to untreated control, followed by F₁₁ (**Table 4**). The fruit yield under treatment F₁₁ was statistically at par with F₁₀, F₉, F₈, and F₇ treatments. The fruit yield per tree in the treatment F₁₂ was 19.88%, 3.56%, 5.84%, 6.49%, 6.07% higher than F₁₃, F₁₀, F₉, F₈, and F₇ respectively. The treatments F₁₁, F₁₀, F₉, F₈, and F₇ were statistically similar, whereas F₁₀, F₉, F₈, F₇, F₆, and F₅ were statistically similar, but significantly lower than F₁₂. The higher fruit yield may be resulted from the improved supply of nutrients to plant through foliar application, which were deficient in the soil due to high soil pH, also reported by Papadakis et al.^[39] and Pestana et al.^[40]. However, Hasani et al.^[41] used 0.3% ZnSO₄ solution for foliar spray in pomegranate and reported 4–5% yield improvement compared to control. The precise application of Zn, Mn and Fe regulates the morphological, metabolic and cytological activities of citrus plants that resulted in higher fruit yields^[42].

Table 4. Effect of tank-mix multi micro-nutrients mixture application on the of kinnow fruit yield. Different letters in each column of experimental factors show significant differences at $P < 0.05$ probability level.

Treatment	Fruit length (cm)	Fruit diameter (cm)	Fruit weight (g)	Fruit yield (kg plant ⁻¹)
F ₁	5.70 ^e	6.90 ^e	152.30 ^f	83.15 ^{de}
F ₂	5.70 ^e	7.10 ^c	154.50 ^f	82.35 ^e
F ₃	5.60 ^f	6.90 ^e	161.20 ^e	80.10 ^f
F ₄	5.70 ^e	7.00 ^d	167.25 ^d	77.50 ^g
F ₅	5.73 ^e	6.57 ^f	151.30 ^f	84.65 ^{cd}
F ₆	6.04 ^d	7.02 ^d	164.00 ^{de}	83.95 ^{ede}
F ₇	6.58 ^a	7.69 ^a	179.20 ^c	85.00 ^{bcd}
F ₈	6.04 ^d	7.04 ^{cd}	165.35 ^d	86.14 ^{bc}
F ₉	6.22 ^c	7.40 ^b	181.50 ^{bc}	84.98 ^{bcd}
F ₁₀	6.37 ^b	7.67 ^a	183.80 ^{ab}	85.21 ^{bcd}
F ₁₁	6.51 ^a	7.70 ^a	185.43 ^a	87.25 ^b
F ₁₂	6.53 ^a	7.72 ^a	186.10 ^a	90.50 ^a
F ₀	5.60 ^f	6.20 ^g	121.50 ^g	72.50 ^h
CV	6.17	6.42	10.85	5.46

Notes: ^{a-h}: Each letter represents statistically similarity at $P < 0.05$.

3.3. The effect of micro-nutrient application on the fruit quality of kinnow mandarin

The fruit quality data presented in **Table 5**, revealed statistically similar peel thickness in the treatments F₁₂, F₁₁, F₁₀, F₉, and F₇, but significantly higher than the untreated control treatments. The thicker peel indicates adequate soil nutrient and moisture supply, and also protect the fruit from external damage resulted from biotic and abiotic factors. However, unusual thick and thin skin results indicate poor fruit quality. However, the peel

weight was significantly higher with treatment F₇ compared to control and other treatments, indicating thicker skin. The peel weight in treatment F₇ was 1.23% and 60.6% higher than F₁₂ and F₀ (control). The seed weight in kinnow fruit ranged from 2.21 g–3.40 g, where significantly higher seed weight was recorded in the treatment F₆ (where trees were sprayed with 0.4% ZnSO₄ + 0.2% MnSO₄ + 0.2% FeSO₄) compared to control and F₁₂ treatment. The seed weight in F₁₂ was significantly lower compared to control and other treatments. The seed weight in F₁₂ treatment was 18.1%, 16.6%, 43.4% less than control (F₀), F₁₁ and F₇ treatments (**Table 5**). The rag weight significantly higher in treatment F₁₀ and F₁₁ compared to control. The rag weight under treatment F₁₂ was 5.79 % and 0.85% less compared to F₁₁ and F₇, respectively. Dutta and Banik^[43] also recorded improved internal physiology with the exogenous application of ZnSO₄ results in better developing fruit for their suitable growth and development traits, resulting in reduction in rag weight and improvement in juice percentage. Barooah et al.^[12] reported that the foliar application of micro-nutrients has improved nutrient availability and absorption that help in improving yield and reducing rag weight.

Table 5. Effect of tank-mix multi micro-nutrients mixture application on the fruit quality of kinnow crop. Different letters in each column of experimental factors show significant differences at $P < 0.05$ probability level.

Treatment	Peel thickness (cm)	Peel weight (g)	Juice weight (g)	Rag weight (g)	Seed weight (g)	Peel weight (%)	Juice (%)	Rag weight (%)	Seed weight (%)
F ₁	2.89 ^c	41.14 ^c	72.4 ^{sh}	35.56 ^f	3.20 ^{cd}	27.01 ^{ab}	47.53 ^{ab}	23.35 ^f	2.10 ^b
F ₂	2.90 ^c	42.1 ^c	73.27 ^{fs}	35.96 ^f	3.17 ^d	27.25 ^a	47.43 ^{ab}	23.27 ^f	2.05 ^c
F ₃	3.25 ^d	42.14 ^c	75.17 ^{ef}	40.66 ^d	3.22 ^c	26.14 ^{cd}	46.6 ^{bc}	25.23 ^{de}	2.00 ^d
F ₄	3.54 ^c	44.57 ^b	78.14 ^{cd}	41.18 ^d	3.35 ^b	26.65 ^{abc}	46.72 ^{bc}	24.62 ^c	2.00 ^d
F ₅	2.80 ^f	39.88 ^d	69.84 ^h	38.39 ^e	3.19 ^{cd}	26.36 ^{de}	46.16 ^{cd}	25.37 ^{cd}	2.11 ^b
F ₆	3.59 ^{ab}	41.81 ^c	76.86 ^{de}	41.93 ^{cd}	3.40 ^a	25.49 ^{de}	46.85 ^{abc}	25.58 ^{cd}	2.07 ^{bc}
F ₇	3.62 ^a	46.85 ^a	80.27 ^c	48.9 ^b	3.17 ^d	26.14 ^{cd}	44.80 ^e	27.29 ^b	1.77 ^e
F ₈	3.55 ^{bc}	43.71 ^b	76.29 ^{de}	43.0 ^c	2.34 ^g	26.43 ^{bc}	46.14 ^{cd}	26.01 ^c	1.42 ^f
F ₉	3.61 ^a	45.9 ^a	83.98 ^b	49.0 ^b	2.61 ^e	25.29 ^e	46.27 ^{cd}	27.00 ^b	1.44 ^f
F ₁₀	3.62 ^a	46.06 ^a	83.20 ^b	52.3 ^a	2.25 ^h	25.06 ^e	45.27 ^{de}	28.45 ^a	1.22 ^h
F ₁₁	3.62 ^a	46.58 ^a	85.02 ^b	51.3 ^a	2.49 ^f	25.12 ^e	45.87 ^{cd}	27.69 ^b	1.34 ^g
F ₁₂	3.60 ^a	46.28 ^a	89.11 ^a	48.49 ^b	2.21 ⁱ	24.87 ^e	47.89 ^a	26.06 ^c	1.19 ⁱ
F ₀	2.76 ^g	29.19 ^e	56.69 ^j	33.0 ^g	2.61 ^e	24.03 ^f	46.67 ^{bc}	27.16 ^b	2.15 ^a
CV	10.54	10.78	10.54	14.56	15.20	3.74	2.12	6.10	20.83

Notes: ^{a-i}: Each letter represents statistically similarity at $P < 0.05$.

The significantly higher juice percentage was recorded in the treatment F₁₂ as compared to control (F₀), however juice percentage treatment F₁₂ was statistically at par with the treatment F₁, F₂ and F₆ treatments. The juice percentage with foliar spray of 0.3% ZnSO₄ + 0.2% MnSO₄ + 0.1% FeSO₄ (treatment F₁₂) has improved the juice percent by 5.46 %, 3.79% and 4.40% compared to treatment F₄, F₈ and F₁₁ treatments, respectively. The improved nutrient supply and absorption through foliar application of micro-nutrients has improved juice percentage in kinnow mandrin^[12].

3.4. The effect of micro-nutrient application on the leaf nutrient uptake of kinnow mandrin

The leaf nutrient concentration represents nutrient reserve available for synthesis and accumulation of photosynthates for fruit development and yield. The data presented in the **Table 6**, shows that the nitrogen content of leaf was significantly affected by the zinc application. The significantly high leaf N was observed in the treatment F₅ with the spray application of 0.4% ZnSO₄ + 0.3% MnSO₄ + 0.2% FeSO₄ compared to control, while statistically at par with the treatment F₁ and F₉. The nitrogen is associated with the enhance leaf area, chlorophyll content, and overall plant vigor, that contributed towards production and accumulation of photosynthates^[44]. However, low leaf N content as in treatment F₁₂ (0.3% ZnSO₄ + 0.2% MnSO₄ + 0.1% FeSO₄)

has been resulted from translocation of N and photosynthates from leaves towards fruits, whereas high Leaf N shows poor translocation.

The leaf phosphorus content was found highest in the treatment F₅ and least in the treatment F₁₁ (**Table 6**). The treatment F₁₁ has 4.1% less leaf P content compared to control, indicating more and efficient translocation of P towards mandarin fruit crop. However, leaf P content in F₁₂ treatment was 11.8 % and 15.4% higher P content compared to control and F₁₁ respectively. The leaf P content in F₁₁ treatment was statistically at par with the F₃, F₈, F₉, F₁₀ and F₁₂. The Zn deficiency appear on leaves due competition of nutrients with leaf P, Fe, Mn and Ca^[42,45], where balanced application of Zn, Mn and Fe regulates the morphological, metabolic and cytological activities of citrus plants^[42]. On the other hand, the potassium application is known to improve fruit quality attributes such as color, size and juice content. The leaf K content varied 0.92% to 1.30%, where highest leaf K content was observed in the leaves from the treatment F₅ and lowest under control plots. The reduction in leaf K content represents more translocation of K towards fruits^[42,46].

The zinc promotes pollen viability and enzymatic activity associated with carbohydrate metabolism in plant system leading to higher fruit set and fruit yield. The highest leaf Zn content was observed in the treatment F₂, followed by F₁ and F₁₂ (**Table 6**). The more leaf Zn may be resulted from higher Zn absorption through foliar Zn application and also resulted in higher yields. Nawaz et al.^[34] also reported significant role on Zn in the auxin synthesis and enhance vitamin C accumulation in ‘Kinnow’ mandarin, leading to better photosynthesis, more starch accumulation in fruits^[35] resulting in better fruit quality and yields. The Leaf Mn and Fe content also followed the similar trend where highest was observed in the treatments applied with foliar spray of zinc content. Kaur et al.^[45] applied different combinations of micronutrient in Kinnow mandarin reported significant increase in leaf iron content due to application of FeSO₄ nutrient to leaves.

Table 6. Effect tank-mix multi micro-nutrients mixture application on the leaf on the uptake macro and micronutrient in kinnow fruit crop. Different letters in each column of experimental factors show significant differences at $P < 0.05$ probability level.

Treatment	N	P	K	Zn	Mn	Fe
	%			mg kg ⁻¹		
F ₁	2.75 ^{abc}	0.130 ^{ab}	1.20 ^c	60.2 ^b	35.4 ^{ab}	63.2 ^{ij}
F ₂	2.65 ^{fg}	0.117 ^{bcd}	1.11 ^{de}	61.5 ^a	35.9 ^a	62.5 ^{ij}
F ₃	2.71 ^{cde}	0.110 ^{cde}	1.05 ^f	58.9 ^c	34.5 ^b	61.5 ^j
F ₄	2.59 ^h	0.100 ^{de}	0.95 ^g	56.2 ^d	36.1 ^a	64.5 ⁱ
F ₅	2.78 ^a	0.139 ^a	1.30 ^a	54.2 ^{ef}	33.2 ^c	70.3 ^h
F ₆	2.67 ^{ef}	0.130 ^{ab}	1.26 ^b	52.3 ^g	30.1 ^d	74.2 ^g
F ₇	2.73 ^{bcd}	0.120 ^{bc}	1.22 ^{bc}	55.2 ^{de}	28.4 ^e	77.8 ^f
F ₈	2.61 ^{gh}	0.110 ^{cde}	1.20 ^c	53.4 ^{fg}	29.4 ^{de}	81.4 ^e
F ₉	2.76 ^{ab}	0.110 ^{cde}	1.15 ^d	53.8 ^f	26.4 ^f	86.5 ^d
F ₁₀	2.65 ^{fg}	0.110 ^{cde}	1.10 ^e	56.1 ^d	32.5 ^c	90.2 ^c
F ₁₁	2.69 ^{def}	0.093 ^e	1.09 ^e	57.8 ^c	30.4 ^d	95.4 ^b
F ₁₂	2.62 ^{gh}	0.110 ^{cde}	1.15 ^d	60.1 ^b	35.2 ^{ab}	100.1 ^a
F ₀	2.45 ⁱ	0.097 ^e	0.92 ^g	18.2 ^h	19.6 ^g	61.3 ^j
CV	3.31	13.18	9.75	20.06	14.77	17.58

Notes: ^{a-j}: Each letter represents statistically similarity at $P < 0.05$.

4. Conclusions

The Zn, Mn, and Fe supply is imperative to the cultivation of kinnow mandarin due to their role in improving fruit set and reducing June and pre-harvest drops, thereby leading to higher fruit yield. These

nutrients have different roles within a plant; their combined application can enhance kinnow production more than that of individual spray. Among the treatments, spray of 0.3% ZnSO₄ + 0.2% MnSO₄ + 0.1% FeSO₄ in combination was the best, as it resulted in lower fruit drop, better fruit size, fruit weight and yield, in addition to more nutrients available for plant growth in the leaves. Hence, it is recommended to spray Zn, Mn, and Fe in combination to reduce the June drop and pre-harvest drop of fruit, which help in achieving a greater yield.

Author contributions

Conceptualization, SS and JS; methodology, SS and JS; investigation, SS; data curation, SS, JS, and SSD; writing—original draft preparation, SS, JS, and SSD; writing—review and editing, SS, JS, and SSD; supervision, SS. All authors have read and agreed to the published version of the manuscript.

Funding

For the conduct of this research investigation, the financial support was provided by the Punjab Agricultural University, Ludhiana, India.

Acknowledgment

The authors are thankful to the Punjab Agricultural University, Ludhiana, India for providing necessary facilities during the research work.

Conflict of interest

No potential conflict of interest was reported by the authors.

References

1. Radović A, Nikolić D, Milatović D, et al. The effect of plant hormones on pollen germination and pollen tube growth of almond cultivars. *Acta Horticulturae* 2016; 1139: 375–380. doi: 10.17660/actahortic.2016.1139.65
2. Graham RD, Webb MJ. Micronutrients and disease resistance and tolerance in plants. *Micronutrients in Agriculture* 1991; 4: 329–370. doi: 10.2136/sssabookser4.2ed.c10
3. Buchanan BB, Gruissen W, Jones RL. *Biochemistry and Molecular Biology of Plants*. American Society of Plant Physiologists; 2000.
4. Marschner P. *Marschner's Mineral Nutrition of Higher Plants*, 3rd ed. Elsevier Academic Press. 2011.
5. Reynolds MP, Braun HJ (editors). *Wheat Improvement*. Springer International Publishing; 2022. doi: 10.1007/978-3-030-90673-3
6. Dimkpa CO, Bindraban PS. Fortification of micronutrients for efficient agronomic production: A review. *Agronomy for Sustainable Development* 2016; 36(1). doi: 10.1007/s13593-015-0346-6
7. Sillanpää M. Micronutrients assessment at the country level: An international study. *FAO Soils Bulletin* 1990; 63: 208.
8. Rai S, Singh PK, Mankotia S, et al. Iron homeostasis in plants and its crosstalk with copper, zinc, and manganese. *Plant Stress* 2021; 1: 100008. doi: 10.1016/j.stress.2021.100008
9. Havlin JL. *Soil Fertility and Fertilizers: An Introduction to Nutrient Management*, 7th ed. Prentice Hall; 2004.
10. Hafez OM, El-Metwally IM. Efficiency of zinc and potassium sprays alone or in combination with some weed control treatments on weeds growth, yield and fruit quality of Washington navel orange orchards. *Journal of Applied Sciences Research* 2007; 3(7): 613–621.
11. Tariq M, Sharif M, Shah Z, et al. Effect of foliar application of micronutrients on the yield and quality of sweet orange (*Citrus sinensis* L.). *Pakistan Journal of Biological Sciences* 2007; 10(11): 1823–1828. doi: 10.3923/pjbs.2007.1823.1828
12. Barooah A, Chakravarty M, Hazarika B. Response of nutrient management on growth, yield and quality of citrus crop: A review. *Journal of Pharmacognosy and Phytochemistry* 2022; 11(5): 291–297.
13. Sajida P. Effect of foliar application of zinc, manganese and boron in combination with urea on the yield of sweet orange. *Pakistan Journal of Agricultural Research* 2000; 16(2): 135–141.
14. Yaseen M, Nadeem M, Hussain S. Investigating the effectiveness of micropower foliar spray on growth and yield of different crops. *Pakistan Journal of Life and Social Sciences* 2004; 2(2): 156–158.
15. Alva AK, Paramasivam S, Obreza TA, et al. Nitrogen best management practice for citrus trees. *Scientia Horticulturae* 2006; 107(3): 233–244. doi: 10.1016/j.scienta.2005.05.017

16. Qamar-uz-Zaman, Schumann AW. Nutrient management zones for citrus based on variation in soil properties and tree performance. *Precision Agriculture* 2006; 7(1): 45–63. doi: 10.1007/s11119-005-6789-z
17. Central Ground Water Board. Play on artificial recharger to ground water and water conservation in overexploration blocks of Bathinda District, Punjab. Available online: http://cgwb.gov.in/District_Profile/Punjab/Bathinda.pdf (assessed on 21 December 2023).
18. Piper CS. *Soil and Plant Analysis*. Pp. 368. Hans Publishers; 1996.
19. Walkley A, Black IA. An examination of the Degtjareff method for determining soil organic matter, and a proposed modification of the chromic acid titration method. *Soil Science* 1934; 37(1): 29–38. doi: 10.1097/00010694-193401000-00003
20. Puri AH. *Soil, Their Physics and Chemistry*. Reinhold Publications Corporation; 1950.
21. Olsen SR, Cole CV, Watanabe FS, Dean LA. Estimation of available phosphorus by extraction with sodium biocarbonate. Available online: <https://ia903207.us.archive.org/21/items/estimationofavai939olse/estimationofavai939olse.pdf> (accessed on 21 December 2023).
22. Jackson ML. *Soil Chemical Analysis*. Prentice Hall of India Pvt. Ltd.; 1973. 498p.
23. Merwin HD, Peech M. Exchangeability of soil potassium in the sand, silt, and clay fractions as influenced by the nature of the complementary exchangeable cation. *Soil Science Society of America Journal* 1951; 15: 125–128. doi: 10.2136/sssaj1951.036159950015000c0026x
24. Subbaiah BV. A rapid procedure for estimation of available nitrogen in soil. *Current Science* 1956; 25: 259–260.
25. Thind SK. Package of practices for cultivation of fruits. Available online: https://www.pau.edu/content/ccil/pf/pp_fruits.pdf (accessed on 21 December 2023).
26. Isaac RA, Johnson WC. Determination of total nitrogen in plant tissue, using a block digester. *Journal of the Association of Official Analytical Chemists* 1976; 59(1): 98–100. doi: 10.1093/jaoac/59.1.98
27. Chapman HD, Pratt PF. Methods of analysis for soils, plants and waters. *Soil Science* 1962; 93(1): 68. doi: 10.1097/00010694-196201000-00015
28. Miller RH, Keeney DR. *Methods of Soil Analysis. Part 2. Chemical and Microbiological Properties*, 2nd ed. American Society of Agronomy (ASA) and Soil Science Society of America; 1982.
29. Liaquat M, Ali I, Ahmad S, et al. Efficiency of exogenous zinc sulfate application reduced fruit drop and improved antioxidant activity of ‘kinnow’ mandarin fruit. *Brazilian Journal of Biology* 2023; 83. doi: 10.1590/1519-6984.244593
30. Lindsay WL, Norvell WA. Development of a DTPA soil test for zinc, iron, manganese, and copper. *Soil Science Society of America Journal* 1978; 42(3): 421–428. doi: 10.2136/sssaj1978.03615995004200030009x
31. Shapiro SS, Wilk MB. An analysis of variance test for normality (complete samples). *Biometrika* 1965; 52(3–4): 591–611. doi: 10.1093/biomet/52.3-4.591
32. Zoremthuangi J, Saipari E, Mandal D. Influence of foliar micronutrients on growth, yield and quality of Khasi mandarin (*Citrus reticulata* Blanco) in Mizoram. *Research on Crops* 2019; 20(2): 322–327. doi: 10.31830/2348-7542.2019.047
33. Marschner H. *Mineral Nutrition of Higher Plant*. Academic Press; 1995. pp. 330–355.
34. Nawaz MA, Ahmad W, Ahmad S, Khan MM. Role of growth regulators on preharvest fruit drop, yield and quality in Kinnow mandarin. *Pakistan Journal of Botany* 2008; 40(5): 1971–1981.
35. Akula V, Delvadia DV, Sharma LK, et al. Effect of micronutrient application on flowering, fruiting and yield of acid lime (*Citrus aurantifolia* L.). cv. Kagzi lime. *International Journal of Tropical Agriculture* 2014; 32(3/4): 331–334.
36. Goren R, Huberman M, Zhong GY. Basics aspects of hormonal control of abscission. *Proceedings International Society of Citriculture* 2000; 9: 591–594.
37. Singh Y, Thakur N, Meena NK. Studies on the effect of foliar spray of Zn, Cu and B on growth, yield and fruit quality of sweet Orange (*Citrus sinensis* L.) cv. Mosambi. *International Journal of Chemical Studies* 2018; 6(5): 3260–3264.
38. Ilyas A, Ashraf MY, Hussain M, et al. Effect of micronutrients (Zn, Cu and B) on photosynthetic and fruit yield attributes of *Citrus reticulata* Blanco var. Kinnow. *Pakistan Journal of Botany* 2015; 47(4): 1241–1247.
39. Papadakis IE, Protopapadakis E, Therios IN, et al. Foliar treatment of Mn deficient ‘Washington navel’ orange trees with two Mn sources. *Scientia Horticulturae* 2005; 106(1): 70–75. doi: 10.1016/j.scienta.2005.02.015
40. Pestana M, Correia PJ, de Varennes A, et al. Effectiveness of different foliar iron applications to control iron chlorosis in orange trees grown on a calcareous soil. *Journal of Plant Nutrition* 2001; 24(4–5): 613–622. doi: 10.1081/pln-100103656
41. Hasani M, Zamani Z, Savaghebi G, et al. Effects of zinc and manganese as foliar spray on pomegranate yield, fruit quality and leaf minerals. *Journal of Soil Science and Plant Nutrition* 2012. doi: 10.4067/s0718-95162012005000009
42. Srivastava AK, Singh S. Citrus decline: Soil fertility and plant nutrition. *Journal of Plant Nutrition* 2009; 32(2): 197–245. doi: 10.1080/01904160802592706

43. Dutta P, Banik AK. Effect of foliar feeding of nutrients and plant growth regulators on physico-chemical quality of sardar guava grown in red and lateritic tract of west Bengal. *Acta Horticulturae* 2007; 735: 407–411. doi: 10.17660/actahortic.2007.735.57
44. Razzaq K, Khan AS, Malik AU, et al. Foliar application of zinc influences the leaf mineral status, vegetative and reproductive growth, yield and fruit quality of 'kinnow' mandarin. *Journal of Plant Nutrition* 2013; 36(10): 1479–1495. doi: 10.1080/01904167.2013.785567
45. Kaur N, Monga PK, Arora PK, et al. Effect of micronutrients on leaf composition, fruit quality and yield of kinnow mandarin. *Journal of Applied and Natural Science* 2015; 7(2): 639–643. doi: 10.31018/jans.v7i2.658
46. Singh J, Singh NP, Sangwan AK, et al. Standardization of litchi leaf nutrient composition for tissue analysis in sub tropics of India. *Communications in Soil Science and Plant Analysis* 2019; 50(22): 2889–2898. doi: 10.1080/00103624.2019.1689253