

EFFECTS OF FOLIAR APPLICATION OF MICRONUTRIENTS ON GROWTH, YIELD AND QUALITY OF SWEET ORANGE (*CITRUS SINENSIS (L.) OSBECK*)

KUMARI NANDITA, MANOJ KUNDU*, RAJIV RAKSHIT¹
AND SAREETA NAHAKPAM²

Department of Horticulture (Fruit & Fruit Technology), Bihar Agricultural University,
Sabour, Bhagalpur, Bihar, India-813210

Keywords: Calcareous soil, Foliar spray, Micronutrients, Mosambi, Non-Traditional track, Fruit quality, Yield

Abstract

Calcareous and alkaline nature of soil hinders smooth up take of micronutrient from soil. Hence, the impact of foliar feeding of micronutrients (Zn, Fe, B, Cu) in different combinations on growth, yield and quality of sweet orange cv. Mosambi in a non-conventional citrus growing area was evaluated. Results revealed that treatment combination of Zn @ 0.5% + Fe @ 0.2% + B @ 0.3% + Cu @ 0.1% (T₉) followed by B @ 0.3% + Fe @ 0.2% (T₆) and Zn @ 0.5% + B @ 0.3% (T₇) was most effective for improving growth of Mosambi plants in terms of shoot length, leaf number, leaf area as well as photosynthetic efficiency of the plant. The yield per plant was also obtained maximum in T₉ (14.94 kg). Further, fruit quality attributes in terms of TSS, total sugar, flavonoids, ascorbic acid content as well as antioxidant capacity of the fruit were recorded maximum in T₉ with at par result in T₆. Therefore, three foliar spray of Zn @ 0.5% + Fe @ 0.2% + B @ 0.3% + Cu @ 0.1% from May-July can be recommended to get maximum yield with better fruit quality from mosambi orchard under non-conventional track having calcareous and alkaline nature of soil.

Introduction

Citrus, one of the most economically important fruits grown worldwide is a rich source of vitamin C along with phenolics compounds, protein, minerals, pigments, volatile compounds (present in the essential oil), lipids, sugars, acids and fibre. These components collectively increases the nutritional as well as antioxidant properties of the fruit and makes them very useful for human health. With increasing awareness about the nutritional security and faster development of processing industries throughout the globe, the demand of citrus has increased tremendously which ultimately forced to increase the area and production of the crop in India during last two decades. But the productivity of the crop in the country is still quite low. Even, it is to be given further attention under unfavourable soil condition as calcareous in nature and alkaline in reaction where optimum uptake of micronutrients from the soil is not as would ordinarily be expected (Zekri and Obreza 2003). Among different low cost technologies, micronutrient management is appeared as one of the key technologies to enhance the productivity of different perennial crops (Kazi *et al.* 2012, Sikarwar and Tomar 2018, Nandita *et al.* 2020). Apart from activating various enzymes, these micronutrients are involved in the synthesis of many compounds essential for plant growth and development which ultimately boost up the crop yield and quality of different fruit crops.

*Author for correspondence: <manojhorti18@gmail.com>. ¹Department of Soil Science and Agricultural Chemistry, Bihar Agricultural University, Sabour, Bhagalpur, Bihar, India-813210. ²Department of Biochemistry & Crop Physiology, Bihar Agricultural University, Sabour, Bhagalpur, Bihar, India-813210.

Therefore, foliar application of micro-nutrients under this unconventional mosambi growing part of the country was carried out to investigate its effect on yield and quality attributes of sweet orange cv. Mosambi.

Materials and Methods

Ten years old sweet orange (*Citrus sinensis* (L.) Osbeck) cv. Mosambi plants with uniform growth and free from any injuries, pest and disease infestation were selected at Horticulture Garden, Bihar Agricultural College, Sabour, Bhagalpur, Bihar, India during 2018-19. The trial was continued for two consecutive growing seasons with the following treatment variables under foliar spray- T_1 = Control (distilled water spray), T_2 = Zn @ 0.5%, T_3 = Cu @ 0.1%, T_4 = B @ 0.3%, T_5 = Cu @ 0.1% + Fe @ 0.2%, T_6 = B @ 0.3% + Fe @ 0.2%, T_7 = Zn @ 0.5% + B @ 0.3%, T_8 = Cu @ 0.1% + B @ 0.3%, and T_9 = Zn @ 0.5%+ Fe @ 0.2% + B @ 0.3% + Cu @ 0.1%. Zn-EDTA (chelated), Cu-EDTA (chelated), Fe-EDTA (chelated) and solubor were used as the source of Zn, Cu, Fe and B, respectively. Working solutions were sprayed though foot sprayer to the entire canopy of the selected mosambi plants during the morning hours. Three foliar sprays at one month interval were done on each experimental plant starting from the month of May. Among the selected micronutrient, application of zinc solution was done fifteen days before the application of other micronutrients at each interval to avoid any antagonistic effect among these micronutrients. The experiment was laid in randomized complete block design (RCBD) with three replications.

Leaf area, leaf area index and chlorophyll content present in the leaf were measured at vegetative as well as reproductive stage of the plant. Leaf area were measured by leaf area meter (CI-203 CA) while leaf area index (LAI) and leaf area duration was calculated to know the photosynthetic efficiency of the plant following the method given by Watson (1947). Chlorophyll content (chlorophyll a, chlorophyll b and total chlorophyll) of the leaves was measured by the method described by Barnes *et al.* (1992). Total number of fruits, fruit size as well as yield was measured manually.

In respect of quality, fruit juice was collected and TSS was estimated using a digital refractometer (Atago, Tokyo, Japan) while titratable acidity was determined by using titration method (Rangana 2010). Total sugar content was determined by Lane and Eynone (1923) method. Ascorbic acid and flavonoid content of the fruit was estimated by 2, 6-dichlorophenol indophenol dye method (Jones and Hughes 1983) and aluminum chloride method (Zhishen *et al.* 1999), respectively while antioxidant capacity was estimated using the method described by Apak *et al.* (2008).

Data of two consecutive years were pooled to prepare average data for each and every parameters. Data were analysed using statistical analysis software (SAS 9.2; SAS Institute, Cary, NC, USA) and the means were compared using DMRT.

Results and Discussion

The growth parameters of Mosambi plants varied significantly with treatment differences over control (Table 1). The T_9 treatment with optimum combination of micronutrients (Zn @ 0.5% + Fe @ 0.2% + B @ 0.3% + Cu @ 0.1%) showed maximum shoot length, leaves shoot⁻¹, leaf area index at vegetative and fruiting stages and leaf area duration as 72.74 cm, 82.67, 0.89, 4.13 and 150.37, respectively. On the other hand, control (T_1) showed significantly lower values by 27.03, 71.05, 154.28, 175.33 and 170.74%, respectively with that of T_9 for each parameter (Table 1). This enhanced vegetative growth by optimum application of Zn, Fe, B and Cu is mainly due to the positive impact of these micronutrients on plants. Zn helps in tryptophane formation which ultimately accelerates auxin biosynthesis resulting improved vegetative growth (Mallick and Muthukrishnan 1979). Application of Fe helps in chlorophyll formation and activation of several

enzymes including those involved in the oxidation or reduction processes of photosynthesis and respiration; B enhance photosynthesis and Cu helps in the starch and nitrogen metabolism thus vegetative growth increased in citrus (Stenico *et al.* 2009).

Table 1. Effect of foliar spray with combination of micronutrients on vegetative growth of sweet orange cv. Mosambi.

Treatment	Shoot length (cm)	Leaf /shoot (No.)	Leaf area index		Leaf area duration (days)
			Vegetative stage	Fruiting stage	
T ₁ = Control	57.26i	48.33d	0.35e	1.50e	55.54f
T ₂ = Zn @ 0.5%	59.58h	52.83d	0.49cde	1.67e	65.03f
T ₃ = Cu @ 0.1%	61.13g	50.67d	0.43de	1.55e	59.52f
T ₄ = B @ 0.3%	65.45e	63.83c	0.58bcd	2.36d	88.07e
T ₅ = Cu @ 0.1% + Fe @ 0.2%	62.92f	64.67c	0.60bcd	2.50d	93.11de
T ₆ = B @ 0.3% + Fe @ 0.2%	71.22b	71.67b	0.67bc	3.06c	111.85c
T ₇ = Zn @ 0.5% + B @ 0.3%	69.45c	74.00b	0.77ab	3.54b	129.17b
T ₈ = Cu @ 0.1% + B @ 0.3%	67.48d	70.50bc	0.62bcd	2.81cd	103.30cd
T ₉ = Zn @ 0.5%+ (Fe @ 0.2% + B @ 0.3% + Cu @ 0.1%)	72.74a	82.67a	0.89a	4.13a	150.37a
SEm (\pm)	0.61	0.78	0.08	0.14	1.01

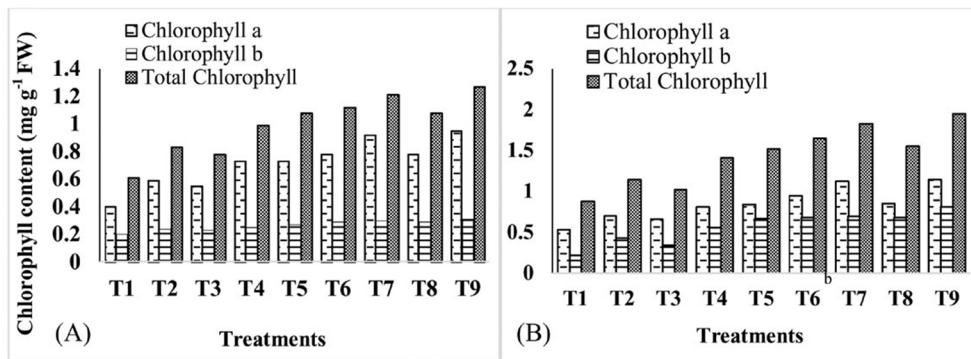
Value indicates mean of three replicates. Different letters in the same column indicate significant differences at $P \leq 0.05$ (DMRT).

Leaf area index (LAI) measured during vegetative stage was recorded maximum in T₉ treatment with par result in T₇ (1.54 and 1.20 times higher than control, respectively) (Table 1). Same trend was also observed in LAI at fruiting stage; although, the LAI at fruiting stage was measured much higher than vegetative stage. Further, photosynthetic efficiency as measured in terms of leaf area duration (LAD), was recorded maximum in T₉ followed by T₇ and T₆ treatment (1.71, 1.33 and 1.01 times higher than control, respectively) (Table 1).

Chlorophyll content of mosambi plants at vegetative stage with respect to chlorophyll a, b and total chlorophyll content were also varied significantly with treatment differences over control (Fig. 1). The T₉ treatment with optimum combination of micronutrients (Zn @ 0.5% + Fe @ 0.2% + B @ 0.3% + Cu @ 0.1%) had maximum chlorophyll a, b and total chlorophyll content as 0.95, 0.31 and 1.27 mg g⁻¹ FW respectively and at par with T₇ (0.92, 0.30 and 1.21 mg g⁻¹ FW respectively). Besides, T₆, T₈, T₅, T₄, T₂ and T₃ also increased the chlorophyll a, b and total chlorophyll content in the leaf significantly at vegetative stage. Further, during fruiting stage, chlorophyll content (chlorophyll a, b and total chlorophyll) was increased as compared to vegetative stage. Total leaf chlorophyll as well as chlorophyll a and b content at fruiting stage was recorded maximum in T₉ (1.22, 1.15 and 2.68 times higher than control, respectively) followed by T₇ (1.08, 1.11 and 2.09 times higher than control, respectively).

Foliar application of micronutrients increased all the photosynthetic compounds significantly within the plant system resulting improved physiological growth with reduced leaf drop. Zn accelerate metabolites translocation and increases photosynthesis by increasing the activity of carbonic anhydrase (Qiao *et al.* 2009). Similarly, boron indirectly increased photosynthesis by involving in the carbohydrate metabolism while Cu regulates different metabolic pathways

(including ATP synthesis) as cofactor for various enzymes. Hence, combined application of all these four micronutrients (Fe, Zn, Cu and B) ultimately enhanced the physiological activities significantly. Further, the increase of chlorophyll a, b and total chlorophyll by combined spray of Fe, Zn, Cu and B is mainly due to enhancement of secondary metabolites (Shitole and Dhumal 2012).



T₁ = Control, T₂ = Zn @ 0.5%, T₃ = Cu @ 0.1%, T₄ = B @ 0.3%, T₅ = Cu @ 0.1% + Fe @ 0.2%, T₆ = B @ 0.3% + Fe @ 0.2%, T₇ = Zn @ 0.5% + B @ 0.3%, T₈ = Cu @ 0.1% + B @ 0.3%, T₉ = Zn @ 0.5% + (Fe @ 0.2% + B @ 0.3% + Cu @ 0.1%).

Fig. 1. Leaf chlorophyll a, b and total chlorophyll content at vegetative (A) and fruiting stage (B) of sweet orange cv. Mosambi as influenced by different combination of micronutrients spray. The SEM (\pm) for chlorophyll a, b and total chlorophyll content at vegetative stage are 0.07, 0.04 and 0.12 respectively while SEM (\pm) for chlorophyll a, b and total chlorophyll content at fruiting stage are 0.08, 0.06 and 0.17, respectively.

Number of fruits shoot⁻¹, fruits plant⁻¹ and yield plant⁻¹ varied significantly with treatment differences over control (Table 2). The T₉ treatment with optimum combination of micronutrients (Zn @ 0.5% + Fe @ 0.2% + B @ 0.3% + Cu @ 0.1%) showed maximum number of fruits shoot⁻¹, fruits plant⁻¹ and yield plant⁻¹ as 13.67 and at par (12.67) with T₇, 93.33 and 14.94 kg and at par (12.46 and 14.25 kg) with T₇ and T₆ (Table 2). On the other hand, control (T₁) showed significantly lower values as 5.67 and 67.33 number of fruits and 8.30 kg, respectively for these three parameters. However, maximum fruit length was recorded in T₆ followed by T₇, T₉ and T₈ treatment (6.61-12.68%, higher than the control); while, fruit width (68.62 mm) was recorded maximum in T₉. This enhancement of yield attributing characters particularly in T₉ and T₇ is mainly due to the positive influence of Zn and B on reproductive growth of the plants. Boron improves the pollen tube growth resulting better fruit setting (Singh *et al.* 2003). Further, it regulates carbohydrate metabolism and accelerate the carbohydrate supply to reproductive buds resulting improved flower and fruit setting with reduced fruit abscission (Smit and Combrink 2005). While Zn enhanced the photosynthates translocation in developing fruits and reduces fruit abscission by increasing IAA synthesis (Shnain *et al.* 2014).

Fruit peel and juice weight of Mosambi were found to vary significantly with treatment differences over control (Fig. 2); however, rag weight was not influenced significantly by foliar feeding of different micronutrient combinations. The T₇ treatment with Zn @ 0.5% + B @ 0.3% had maximum (77.44 g) juice content in the ripened mosambi fruit and at par (75.45 g) with T₆. However, control (T₁) showed significantly lower values by 38.02% for juice content (Fig. 2). On the other hand, peel weight was measured maximum in T₆ followed by T₇, T₉ and T₈ treatments.

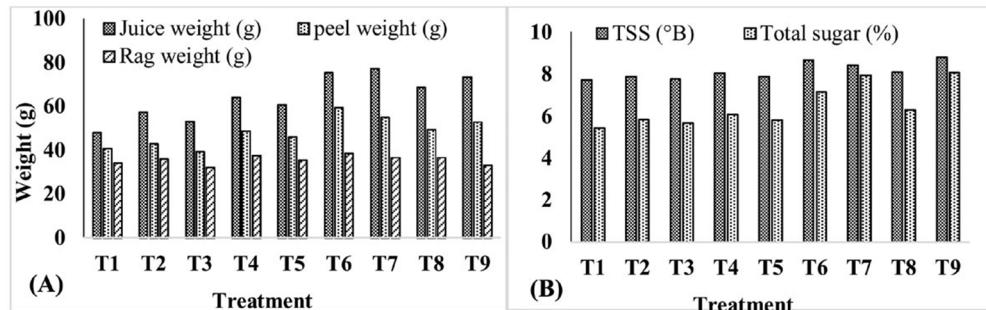
The biochemical parameters with respect to total soluble solid content, total sugar content, ascorbic acid and flavonoid content as well as total antioxidant capacity were found to vary significantly with treatment differences over control in different micronutrient treated plants (Fig. 2 and Table 2). The T₉ treatment with optimum combination of micronutrients showed maximum total soluble solid content, total sugar content, ascorbic acid and flavonoid content as well as total antioxidant capacity as 8.83 °B, 8.09%, 50.83 mg 100g⁻¹ FW, 13.99 mg g⁻¹ FW and 196.37 µmol. Trolox equiv. 100g⁻¹ FW, respectively and at par with T₇. On the other hand, control (T₁) showed significantly lower values by 12.57, 32.63, 36.06, 42.03 and 15.66%, respectively with that of T₉ for each parameter (Fig. 2 and Table 2). However, titratable acidity of mosambi fruits decreased significantly in all the micronutrient treatment with minimum acidity (0.194%) in T₉ (Table 2).

Table 2 Effect of foliar spray with combination of micronutrients on yield and fruit quality attributes of sweet orange cv. Mosambi.

Treatment	Yield attributes			Fruit size			Fruit quality attributes			
	Fruits/ shoot (No.)	Fruits/ plant (No.)	Yield/ plant (kg)	Fruit length (mm)	Fruit width (mm)	Acidity (%)	Ascorbic acid (mg/ 100 g FW)	Flavonoids (mg/g FW)	Antioxidant capacity (µmol. Trolox equiv./ 100 g FW)	
T ₁ = Control	5.67 ^e	67.33 ^d	8.30 ^e	60.64 ^d	60.25 ^d	0.313 ^a	32.50 ^f	8.11 ^d	165.61 ^c	
T ₂ = Zn @ 0.5%	7.67 ^d	71.67 ^d	9.80 ^d	62.10 ^d	61.42 ^{cd}	0.260 ^b	42.50 ^{cd}	9.54 ^{cd}	171.13 ^c	
T ₃ = Cu @ 0.1%	6.33 ^e	67.00 ^d	8.36 ^e	60.76 ^d	60.77 ^{cd}	0.273 ^b	36.50 ^{ef}	8.30 ^d	166.79 ^c	
T ₄ = B @ 0.3%	7.67 ^d	76.67 ^c	11.57 ^c	63.07 ^{bcd}	64.34 ^{bc}	0.243 ^{bc}	40.73 ^{de}	10.11 ^{cd}	181.16 ^b	
T ₅ = Cu @ 0.1% + Fe @ 0.2%	8.33 ^d	81.67 ^b	11.63 ^c	62.29 ^{cd}	62.93 ^{bcd}	0.228 ^{cd}	45.33 ^{bcd}	10.12 ^{cd}	179.66 ^b	
T ₆ = B @ 0.3% + Fe @ 0.2%	12.33 ^b	82.00 ^b	14.25 ^a	68.33 ^a	68.62 ^a	0.195 ^e	45.00 ^{bcd}	10.62 ^{bc}	185.03 ^b	
T ₇ = Zn @ 0.5% + B @ 0.3%	12.67 ^{ab}	85.33 ^b	14.46 ^a	65.08 ^b	64.52 ^{bc}	0.200 ^{de}	49.17 ^{ab}	12.20 ^{ab}	194.10 ^a	
T ₈ = Cu @ 0.1% + B @ 0.3%	10.33 ^c	83.00 ^b	12.88 ^b	64.65 ^{bc}	64.75 ^{bc}	0.199 ^{de}	45.83 ^{bc}	11.45 ^{bc}	192.79 ^a	
T ₉ = Zn @ 0.5% + (Fe @ 0.2% + B @ 0.3% + Cu @ 0.1%)	13.67 ^a	93.33 ^a	14.94 ^a	64.92 ^b	65.68 ^{ab}	0.194 ^e	50.83 ^a	13.99 ^a	196.37 ^a	
SEm (±)	0.11	0.63	0.16	0.59	0.48	0.007	0.54	0.17	1.06	

Value indicates mean of three replicates. Different letters in the same column indicate significant differences at P ≤ 0.05 (DMRT).

Improvement of fruit quality particularly under the treatment of T₉ and T₇ is mainly associated with the role of Zn and B. Zn specifically accelerate the activity of aldolase enzyme which in turn helps in higher accumulation of sugar in the fruits. While, boron helps to increase sugar translocation from source to sink by forming sugar-boron complex (furanosecis-diol structure). Fe helps in the synthesis of carbohydrate and act as a strong sink resulting enhancement of sugar content and TSS in ripened mosambi fruits (Ram and Bose 2000). In addition, copper has positive impact on improving fruit quality particularly TSS and sugar content in ripe fruits (Khurshid *et al.* 2008). Hence the combined application of Zn, Fe, Cu and B together as well as Zn and B together ultimately improved the overall fruit quality attributes significantly as compared to other treatments.



T₁ = Control, T₂ = Zn @ 0.5%, T₃ = Cu @ 0.1%, T₄ = B @ 0.3%, T₅ = Cu @ 0.1% + Fe @ 0.2%, T₆ = B @ 0.3% + Fe @ 0.2%, T₇ = Zn @ 0.5% + B @ 0.3%, T₈ = Cu @ 0.1% + B @ 0.3%, T₉ = Zn @ 0.5% + (Fe @ 0.2% + B @ 0.3% + Cu @ 0.1%).

Fig. 2. Juice, peel and rag weight (A) TSS and sugar content (B) of ripped sweet orange cv. Mosambi fruit as influenced by different combinations of micronutrients spray. The SEM (\pm) for juice, peel, rag weight, TSS and Total sugar content are 0.87, 0.62, 0.41, 0.13 and 0.09, respectively.

Results of the present investigation showed that the physiological growth of sweet orange (*Citrus sinensis* (L.) Osbeck) cv. Mosambi has improved significantly at T₉ in non-traditional citrus growing part of the country. This improved physiological growth ultimately helps to increase fruit yield with improved fruit quality. Hence, three foliar application of Zn @ 0.5% + (Fe @ 0.2% + B @ 0.3% + Cu @ 0.1%) (T₉) during the month of May, June and July may be recommended for getting maximum profit from mosambi orchard particularly under non-conventional citrus growing track having calcareous and alkaline nature of soil.

Reference

- Apak R, Guclu K, Ozyurek M and Celik SE 2008. Mechanism of antioxidant capacity assays and the cupric (cupric ion reducing antioxidant capacity) assay. *Microchi. Acta* **160**: 413-419.
- Barnes JD, Balaguer L, Manrique E, Elvira S and Davison AW 1992. A reappraisal of the use of DMSO for the extraction and determination of chlorophylls a and b in lichens and higher plants. *Environ. Exp. Bot.* **32**: 85-100.
- Jones E and Hughes RE 1983. Foliar ascorbic acid in some angiosperms. *Phytochem.* **22**: 2493-2499.
- Kazi SS, Ismail S and Joshi KG 2012. Effect of multi-micronutrient on yield and quality attributes of sweet orange. *African J. Agricul. Res.* **7**: 4118-4123.
- Khurshid F, Khattak RA and Sarwar S 2008. Effect of foliar (Zn, Fe, Cu and Mn) in citrus production. *Sci. Technol. Develop.* **27**: 34-42.
- Lane JH and Eynone L 1923. Determination of reducing sugars by means of Fehling solution with methylene blue indicator as an internal indicator. *J. Indian Chem. Soc.* **42**: 32.
- Mallick MFR and Muthukrishnan CR 1979. Effect of micro nutrients on tomato (*Lycopersicon esculentum* Mill.), Effect on flowering, fruit-set and yield. *South Indian Horticul.* **28**: 14-20.
- Nandita K, Kundu M, Rani R, Khatoon F and Kumar D 2020. Foliar feeding of micronutrients: an essential tool to improve growth, yield and fruit quality of sweet orange (*Citrus sinensis* (L.) Osbeck) cv. Mosambi under non-traditional citrus growing track. *International Journal of Curr. Microbiol. Appl. Sci.* **9**(3): 473-483.
- Qiao X, He Y, Wang Z, Li X, Zhang K and Zeng H 2009. Effect of foliar spray of β carbonic anhydrase expression and enzyme activity in rice (*Oryza sativa* L.) leaves. *Acta Physiologae Planta.* **36**: 263-272.
- Ram RA and Bose TK 2000. Effect of foliar application of manganesium and micronutrients on growth, yield and fruit quality of mandarin orange. *Indian J. Horticul.* **57**: 215-220.
- Rangana S 2010. *Handbook of Analysis and quality control for fruit and vegetable products*, Tata Mc Graw-Hill Ltd., New Delhi.

- Shitole SM and Dhumal KN 2012. Influence of foliar applications of micronutrients on photosynthetic pigments and organic constituents of medicinal plant *Cassia angustifolia* Vahl. Ann. Biol. Res. **3**: 520-526.
- Shnain RS, Prasad VM and Saravanan S 2014. Effect of zinc and boron on growth, yield and quality of tomato (*Lycopersicon esculentum* Mill) cv. Heem Sohna, under protected cultivation. European Acad. Res. **3**: 4572-4597.
- Sikarwar PS and Tomar KS 2018. Nutrient management study in sweet orange (*Citrus sinensis* L) cv. Mosambi. J. Pharmaco. Phytochem. **7**(2): 2217-2219.
- Singh M, Batra VK, Bhatia AK, Singh V and Arora SK 2003. Response of foliar application of micronutrients on tomato variety HisarArun. Veget. Sci. **30**(2): 182-184.
- Smit JN and Combrink NJJ 2005. Pollination and yield of winter-grown greenhouse tomatoes as affected by boron nutrition, cluster vibration and relative humidity. South African J. Plant and Soil **22**: 110-115.
- Stenico SME, Pacheco FTH, Pereira- Filho ER, Rodrigues JLM, Souza AN, Etchegaray A, Gomes JE Tsai SM 2009. Nutritional deficiency in citrus with symptoms of citrus variegated chlorosis disease. Brazilian J. Biol. **69**: 1-7.
- Watson DJ 1947. Comparative physiological studies on the growth of field crops I. Variation in net assimilation rate and leaf area between species and varieties and within and within and between years. Annals of Botany-London **11**: 41-76.
- Zekri M and Obereza TA 2003. Micronutrient deficiencies in citrus: Iron, zinc and manganese. Institute of Food and agricultural Services, University of Florida, USA. <http://edis.ifas.ufl.edu>.
- Zhishen J, Mengcheng T and Jianming W 1999. The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. Food Chem. **64**: 555-559.

(Manuscript received on 16 June, 2020; revised on 29 November, 2021)