www.ThePharmaJournal.com

The Pharma Innovation

**ISSN (E): 2277- 7695**
**ISSN (P): 2349-8242**
**NAAS Rating: 5.23**
TPI 2021; 10(4): 671-674

www.thepharmajournal.com
Received: 16-02-2021
Accepted: 18-03-2021

**Dr. Manoj Bhanukar**
Ph.D. Scholar Department of Horticulture, CCS HAU, Hisar, Haryana, India

**Dr. GS Rana**
Principal Scientist, Department of Horticulture, CCS HAU, Hisar, Haryana, India

**Preeti**
Post Graduate from Department of Horticulture, CCS HAU, Hisar, Haryana, India

**Corresponding Author:**
**Preeti**
Post Graduate from Department of Horticulture, CCS HAU, Hisar, Haryana, India

# Effect of foliar application of micronutrients and soil application of zinc sulphate on total chlorophyll and carotenoid content in sweet orange (*Citrus sinensis*) cv. blood red

**Dr. Manoj Bhanukar, Dr. GS Rana and Preeti**

**DOI:** https://doi.org/10.22271/tpi.2021.v10.i4j.6034

**Abstract**
The present investigation was carried out for two consecutive years of 2016 and 2017 at experimental orchard of Department of Horticulture, CCS HAU, Hisar to assess the performance of sweet orange cv. Blood Red to foliar application of micronutrients and soil application of zinc sulphate. In experiment I, the foliar application of micronutrients was done two times i.e. Ist spray was done on first week of april and 2nd spray on first week of july. The experiment comprises of thirteen treatments, *viz*., Zinc sulphate ($ZnSO_4$) @ 0.25%, 0.50% &1.00%; Iron sulphate ($FeSO_4$) @ 1.5%, 2.0% & 2.5%; Manganese sulphate ($MnSO_4$) @ 0.25%, 0.50% & 1.00%; Boric acid ($H_3BO_3$) @ 0.1%, 0.2% & 0.3% and Control (No micronutrients). In experiment II, the soil application of zinc sulphate ($ZnSO_4$) was done two times i.e. half dose in second fortnight of february and half dose in second fortnight of May. The experiment comprises of six treatments, *viz*., Zinc sulphate ($ZnSO_4$) @ 50 g/plant, 100 g/plant, 150 g/plant, 200 g/plant, 250 g/plant and Control (No Zn application). Results indicated that foliar application of micronutrients and soil application of zinc sulphate affected significantly the chlorophyll and carotenoid content.

**Keywords:** *Citrus sinensis*, foliar application, soil application, micronutrients, sweet orange

## Introduction
The genus *Citrus* is economically very important and is known for its juice and pulp throughout the world. Citrus is one of the choicest fruit with high consumer preference both as fresh fruit as well as for its refreshing processed juice. It is extensively grown in tropical and sub-tropical regions. Citrus is a commercially important fruit crop of India and grown across its length and breadth with a production of 13200 thousand MT from an area of 1034 thousand hectares during year of 2018-19 (Anonymous, 2018-19) [1]. Sweet orange (*Citrus sinensis* (L.) Osbeck) has been reported to be originated in Southern China and it was introduced to India during thirteenth century (Swingle, 1943; Webber, 1948) [9, 11]. It is the second largest citrus fruit, cultivated in tropical and subtropical regions of the country. Total sweet orange production in india is 3401 thousand MT with an area of 190 thousand hectares during year of 2018-19 (Anonymous, 2018-19) [1]. The productivity of sweet orange mainly depends on adequate supply of plant nutrients seems to be a very important factor in regulating cropping and influencing the quality of sweet orange. The micronutrients are required in small amounts, but play a great role in plant metabolism (Kazi *et al*., 2012) [4]. Despite some shortcomings, foliar application is regarded as the best method under certain conditions (Marschner and Marschner, 2012) [6]. Foliar or soil application of Zn increases the biosynthesis of chlorophyll and carotenoid synthesis that are important for proper performance of photosynthetic process (Mousavi, 2011) [7]. The beneficial effects of amelioration of zinc, iron, manganese and boron deficiencies by foliar application have been documented by previous studies (Hippler *et al*., 2015) [3]. The present investigation has been undertaken to study the effect of foliar application of micronutrients and soil application of zinc sulphate on chlorophyll and carotenoid content of fruit sweet orange.

## Material and Methods
The present investigation was carried out on fifteen years old sweet orange cv. Blood Red trees planted at a spacing of 6m X 6m in experimental orchard of Department of Horticulture, CCS

HAU, Hisar situated at 215.2 m above mean sea level with coordinates of 29º10′ N latitude and 75º46′ E longitudes, during the year 2016-17 and 2017-18. In Experiment I, the foliar application of micronutrients was done two times i.e. Ist spray was done on first week of april and 2nd spray on first week of july. The experiment comprises of thirteen treatments, *viz*., Zinc sulphate ($ZnSO_4$) @ 0.25%, 0.50% &1.00%; Iron sulphate ($FeSO_4$) @ 1.5%, 2.0% & 2.5%; Manganese sulphate ($MnSO_4$) @ 0.25%, 0.50% & 1.00%; Boric acid ($H_3BO_3$) @ 0.1%, 0.2% & 0.3% and Control (No micronutrients). In experiment II, the soil application of zinc sulphate ($ZnSO_4$) was done two times i.e. half dose in second fortnight of february and half dose in second fortnight of May. The experiment comprises of six treatments, *viz*., Zinc sulphate ($ZnSO_4$) @ 50 g/plant, 100 g/plant, 150 g/plant, 200 g/plant, 250 g/plant and Control (No Zn application). Treatments were allocated in randomized block design (RBD) with three replications in each treatment. The recommended standard package of practises and plant protection measures were adopted to keep the plants in good health. Methods used to measure the chlorophyll and carotenoid content are as follows:-

**Total chlorophyll and carotenoid Content:** Chlorophyll pigments namely chl. a, chl. b, total chlorophyll and carotenoids were estimated in the fruits taken from different sampling stages by the method given by Wellburn (1994) [12] and expressed in terms of µ moles/g fresh weight of peel tissue.

**Extraction of pigments:** Two discs (0.28 $cm^2$ area) of the fruit peel were suspended in a test tube containing 10 ml of dimethyl sulphoxide (DMSO). The weight of the discs was recorded before putting them into test tubes. The test tubes were then placed in an oven at 60ºC for four hours to facilitate the extraction of pigments.

**Estimation:** After the incubation period for extraction, the test tubes were cooled to room temperature and the optical density was read at 454, 645 and 665 nm on a spectrophotometer (Spectronic-20D). DMSO was used as blank.

The chlorophyll pigments were calculated by using the following equations:-

Chlorophyll - a (µg/ml) = 12.19 $A_{665}$ – 3.45 $A_{645}$

Chlorophyll - b (µg/ml) = 21.99 $A_{645}$ – 5.32 $A_{665}$

Total Chlorophyll (µg/ml) = Chlorophyll a + Chlorophyll b

$$\text{Carotenoids (µg/ml)} = \frac{(1000\ A_{454} - 2.86\ \text{chl. a} - 129.2\ \text{chl. b})}{221}$$

The quantity of the above pigments were calculated in mg $g^{-1}$ tissue fresh weight and expressed in µ moles/g tissue fresh weight by using the following relationship:-

µ moles of Chlorophyll – a = µg chl. a X 1.119

µ moles of Chlorophyll – b = µg chl. b X 1.102

µ moles of Carotenoids = µg carotenoids X 1.809

## Results and Discussion

**Table 1:** Effect of foliar application of micronutrients on total chlorophyll content (µ moles/g fr. wt.) of sweet orange cv. Blood Red

| Treatments | Total Chlorophyll content 2016 | Total Chlorophyll content 2017 |
|---|---|---|
| $ZnSO_4$ 0.25% | 180.86 | 182.10 |
| $ZnSO_4$ 0.50% | 182.03 | 183.71 |
| $ZnSO_4$ 1.00% | 183.92 | 185.22 |
| $FeSO_4$ 1.5% | 185.44 | 186.89 |
| $FeSO_4$ 2.00% | 187.24 | 188.14 |
| $FeSO_4$ 2.5% | 189.91 | 189.80 |
| $MnSO_4$ 0.25% | 176.35 | 177.78 |
| $MnSO_4$ 0.50% | 177.78 | 179.12 |
| $MnSO_4$ 1.00% | 179.21 | 180.56 |
| $H_3BO_3$ 0.1% | 172.36 | 174.18 |
| $H_3BO_3$ 0.2% | 173.39 | 175.01 |
| $H_3BO_3$ 0.3% | 174.98 | 176.37 |
| Control | 171.12 | 172.81 |
| C.D. at 5% | 1.16 | 1.12 |

The data concerning total chlorophyll content is presented in Table 1, which indicates that different concentrations of $ZnSO_4$, $FeSO_4$, $MnSO_4$ and $H_3BO_3$ affected total chlorophyll content significantly during both the years. In year 2016, maximum (189.91) total chlorophyll content was observed with $FeSO_4$ 2.5% followed by $FeSO_4$ 2.00% (187.24) and minimum (171.12) in control. Similar trend was observed in the year 2017, showing maximum (189.80) total chlorophyll content was observed with $FeSO_4$ 2.5% followed by $FeSO_4$ 2.00% (188.14) and minimum (172.81) in control, respectively. The results on total chlorophyll were found significant with foliar application of micronutrients. The highest total chlorophyll was observed with $FeSO_4$ 2.5%. An increase in total chlorophyll content with foliar application of micronutrients might be due to delayed chlorophyll degradation. The results are in conformation with Shahin *et al.* (2010) [8] in apple and Eiada and Al-Hadethi (2013) [2] in pomegranate. This might also be due to delayed senescent pigment changes and oppose the ethylene induced loss of chlorophyll (Trebitsh *et al.*, 1993) [10].

**Table 2:** Effect of foliar application of micronutrients on carotenoid content (µ moles/g fr. wt.) of sweet orange cv. Blood Red

| Treatments | Carotenoid content 2016 | Carotenoid content 2017 |
|---|---|---|
| $ZnSO_4$ 0.25% | 246.55 | 249.67 |
| $ZnSO_4$ 0.50% | 245.01 | 247.09 |
| $ZnSO_4$ 1.00% | 242.89 | 245.80 |
| $FeSO_4$ 1.5% | 241.08 | 243.19 |
| $FeSO_4$ 2.00% | 238.89 | 241.98 |
| $FeSO_4$ 2.5% | 235.78 | 238.87 |
| $MnSO_4$ 0.25% | 251.17 | 253.19 |
| $MnSO_4$ 0.50% | 249.61 | 251.65 |
| $MnSO_4$ 1.00% | 248.02 | 250.12 |
| $H_3BO_3$ 0.1% | 255.50 | 257.45 |
| $H_3BO_3$ 0.2% | 254.26 | 256.56 |
| $H_3BO_3$ 0.3% | 252.60 | 254.62 |
| Control | 257.01 | 259.01 |
| C.D. at 5% | 1.41 | 1.20 |

The data presented in Table 2 indicate that different concentrations of $ZnSO_4$, $FeSO_4$, $MnSO_4$ and $H_3BO_3$ significantly affected carotenoid content of sweet orange cv. Blood Red fruits during both the years. In the year 2016, the highest carotenoid content (257.01) was found in control followed by $H_3BO_3$ 0.1% (255.50) and lowest (235.78) in $FeSO_4$ 2.5%. During the year 2017, highest carotenoid content (259.01) was found in control followed by $H_3BO_3$ 0.1% (257.45) and lowest (238.87) in $FeSO_4$ 2.5%. The results on content were found significant with foliar application of micronutrients. The highest carotenoid content was found in control whereas lowest in $FeSO_4$ 2.5%. A decrease in carotenoid content with foliar application of micronutrients might be due to inhibition of carotenoid beta-cyptoxanthin biosynthesis and its accumulation, which inhibit the development of citrus fruit colour and luster. The results are in conformation with Shahin *et al.* (2010) [8] in apple and Eiada and Al-Hadethi (2013) [2] in pomegranate.

**Table 3:** Effect of soil application of zinc sulphate on total chlorophyll content (µ moles/g fr. wt.) of sweet orange cv. Blood Red

| Treatments | Total chlorophyll content 2016 | Total chlorophyll content 2017 |
|---|---|---|
| $ZnSO_4$ 50 g/plant | 176.44 | 176.92 |
| $ZnSO_4$ 100 g/plant | 177.98 | 178.02 |
| $ZnSO_4$ 150 g/plant | 179.76 | 180.23 |
| $ZnSO_4$ 200 g/plant | 181.48 | 182.91 |
| $ZnSO_4$ 250 g/plant | 184.67 | 185.01 |
| Control | 172.78 | 173.66 |
| C.D. at 5% | 1.54 | 1.24 |

Data presented in Table 3 revealed that total chlorophyll content was significantly affected by various $ZnSO_4$ concentrations during both the years. In year 2016, the maximum total chlorophyll content (184.67) was observed with $ZnSO_4$ 250g/plant followed by $ZnSO_4$ 200g/plant (181.48) and minimum (172.78) in control. In year 2017, the maximum total chlorophyll content (185.01) was observed with $ZnSO_4$ 250g/plant followed by $ZnSO_4$ 200g/plant (182.91) and minimum (173.66) in control.

The highest total chlorophyll was observed with $ZnSO_4$ 250g/plant. This might be because zinc delayed chlorophyll degradation. Zinc application enhances the rate of photochemical reductions and chlorophyll content in cucumber (Kazemi, 2013) [5]. The soil application of Zn increases the biosynthesis of chlorophyll that is important for proper performance of photosynthetic process (Mousavi, 2011) [7].

**Table 4:** Effect of soil application of zinc sulphate on carotenoid content (µ moles/g fr. wt.) of sweet orange cv. Blood Red

| Treatments | Carotenoid content 2016 | Carotenoid content 2017 |
|---|---|---|
| $ZnSO_4$ 50 g/plant | 252.14 | 254.23 |
| $ZnSO_4$ 100 g/plant | 250.54 | 252.14 |
| $ZnSO_4$ 150 g/plant | 248.89 | 250.53 |
| $ZnSO_4$ 200 g/plant | 245.12 | 247.91 |
| $ZnSO_4$ 250 g/plant | 243.33 | 245.03 |
| Control | 254.48 | 256.11 |
| C.D. at 5% | 1.12 | 1.78 |

Data given in Table 4 indicated that carotenoid content was significantly affected by various $ZnSO_4$ concentrations during both the years. In year 2016, the carotenoid content was found maximum (254.48) in control followed by $ZnSO_4$ 50g/plant (252.14) and minimum (243.33) with $ZnSO_4$ 250g/plant .In the next year 2017, the carotenoid content was recorded maximum (256.11) in control followed by $ZnSO_4$ 50g/plant (254.23) and minimum (245.03) with $ZnSO_4$ 250g/plant .

The results carotenoid content were found significant with soil application of zinc sulphate. The highest carotenoid content was found in control. An decrease in carotenoid content might be because zinc inhibit carotenoid beta-cyptoxanthin biosynthesis and its accumulation, which inhibit the development of citrus fruit colour and luster. The results are in conformity with Shahin *et al.* (2010) [8] in apple and Eiada *et al.* (2013) [3] in pomegranate.

## References


1. Anonymous. 3rd Advance Estimate. 2018-19; http://www.nhb.org.in
2. Eiada AO, Al-Hadethi MEA. Effect of foliar application with manganese and zinc on pomegranate growth, yield and fruit quality. Journal of Horticultural Science and Ornamental Plants 2013;5(1):41-45.
3. Hippler FWR, Boaretto RM, Quaggio JA, Azevedo RA, Mattor DJ. Towards soil management with Zn and Mn: estimates of fertilization efficiacy of citrus trees. Annals of Applied Biology 2015;166(3):484-495.
4. Kazi SS, Sayed I, Joshi KG. Effect of multi-micronutrient on yield and quality attributes of sweet orange. African Journal of Agricultural Research 2012;7(29):4118-23.

5. Kazemi M. Application of iron and zinc on growth and productivity of cucumber. Bull. Environ. Pharmacol. Life Sci 2013;2:11-14.
6. Marschner H, Marschner P. Marschner's mineral nutrition of higher plants. Third edition, Elsevier, Academic Press, Amsterdam 2012, 651.
7. Mousavi SR. Zinc in crop production and interaction with phosphorus. *Australian* Journal of Basic and Applied Sciences 2011;5:1503-1509.
8. Shahin MFM, Fawzi MIF, Kandil EA. Influence of foliar application of some nutrient (Fertifol Misr) and gibberellic acid on fruit set, yield, fruit quality and leaf composition of "Anna" apple trees grown in sandy soil. Journal of American Science 2010;6(12): 202-208.
9. Swingle WT. The botany of citrus and its wild relatives in the orange subfamily. In: Citrus Industry, Webber H. S. Batchelor, D. L. (eds). University of California, Berkeley 1943;1:128-427.
10. Trebitsh T, Goldschmidt EE, Riov J. Ethylene induces de nova synthesis of chlorophyllase, a fruit peel. Proceeding Nath, Academy of Science, USA. 1993;90:9441-9445.
11. Webber HJ. Plant characteristic and climatology. Chapter II In Citrus Industry. Vol. I. Ed. By Batcelor, L. D. and Webber, H. J., Univ. Calif. Press Berkeley and Los Angeles 1948.
12. Wellburn AR. The spectral determination of chlorophylls a and b, as well as total carotenoids, using various solvents with spectrophotometers of different resolution. Journal of Plant Physiology 1994;144(3):307-313.