Journal of Pharmacognosy and Phytochemistry
Available online at www.phytojournal.com

E-ISSN: 2278-4136
P-ISSN: 2349-8234
JPP 2018; 7(6): 1606-1610
Received: 25-09-2018
Accepted: 27-10-2018

Yogendra Singh
Department of Fruit Science, VCSG UUH & F Bharsar, Uttarakhand, India

Prerak Bhatnagar
Department of Fruit Science, CH&F, Jhalawar, Rajasthan, India

Nirmal Kumar Meena
Department of Fruit Science, CH&F, Jhalawar, Rajasthan, India

Suresh Chandra Gurjar
Department of Horticulture, Rajasthan College of Agriculture, MPUAT, Udaipur, Rajasthan India

Correspondence
Yogendra Singh
Department of Fruit Science, VCSG UUH & F Bharsar, Uttarakhand, India

The effect of foliar spray of Zn, Cu and B on physico-chemical parameters of sweet orange (*Citrus sinensis* L.) cv. Mosambi

Yogendra Singh, Prerak Bhatnagar, Nirmal Kumar Meena and Suresh Chandra Gurjar

Abstract
The present investigation was conducted at the farm of Department of Fruit Science, VCSG Uttarakhand University of Horticulture and Forestry Bharsar, Pauri Garhwal Uttarakhand during 2016-17. The experiment was laid out in Randomized Complete Block Design comprising ten treatments and three replications. Based on the result obtained from the present investigations, it was observed that the highest juice percentage (28.16%), total soluble solids (10.49^{0}Brix), ascorbic acid (38.52 mg/100 ml), reducing sugar (4.95%), non-reducing sugar (1.95%), total sugar (6.90and minimum titratable acidity (0.89%), number of seeds (8.10%) and peel thickness (4.20%) were observed with T_7 treatment [Zn (0.5%) + B (0.3%) + Cu (0.7%)]. The above mentioned treatment of micronutrients was found most effective in terms of enhancement of fruit quality attributes of sweet orange and could be recommended to the stakeholders and farmers.

Keywords: Foliar spray, Mosambi, Micro-nutrients, Ascorbic acid, Quality

Introduction
Sweet orange is the important fruit crop of the world in terms of nutritional and antioxidant value. The genus Citrus includes more than 162 species belonging to the order Geraniales, family Rutaceae and sub family Aurantoideae having predominance of schizolysigenic oil glands. Citrus fruits are a fair source of vitamin C and their daily consumption protects mankind from scurvy, a disease commonly associated with inadequate availability of vitamin C in the dietary foods. Citrus fruits are being cultivated in India in four different zones i.e. Central India (Madhya Pradesh, Maharashtra and Gujarat), Southern India (Andhra Pradesh and Karnataka), North-Western India (Punjab, Rajasthan, Haryana and Western UP) and North-Eastern India (Meghalaya, Assam and Sikkim). These zones have different leading cultivar that occupies a place of prominence in the respective area (Singh and Singh 2006) [42]. Among the various citrus species, mandarin, sweet orange and lime are the common major citrus fruits having 50, 21 and 15 per cent of total area under them, respectively (NHB. Database, 2017-18). Foliar application of mineral nutrients is a method for quick supply of the elements for the higher plants. This technique allows the plants to consume nutrients much faster than their uptake from soil by their roots. Despite some shortcomings, it is regarded as the best method under certain conditions (Marschner and Marschner, 2012) [24]. The micronutrients are required in small amounts, but play a great role in plant metabolism (Katyal, 2004; Kazi *et al*., 2012) [18, 19]. These are involved in the synthesis of many compounds essential for plant growth and productivity and act as activators for various enzymes. Manganese is required in the process of photosynthesis (Mengel and Kirkby, 1987) [25] and ferrous plays a key role in several enzyme-systems, in which haeme or haemin is the prosthetic group (Khurshid *et al*., 2008) [20]. Fully grown up tree bears 125 kg fruits. The total area of sweet orange in India is 1.85 lakh hectares and production 2982 ('000 MT) with the productivity of 10.4 MT/ha (NHB Database, 2017-18). The micro nutrients play an important role in the development and growth of new cells in plant meristem as citrus needs exhaustive nutrition has an impact in terms of macro and micro nutrients. Zn and B foliar sprays either alone or in combination on the different measured parameters of “sweet orange cv. Mosambi. These findings go in line with previously conducted investigations (Ashraf *et al.* 2013 and Razzaq *et al.* 2013) [3, 34] working on “Kinnow” mandarin. They found that the Zn foliar sprays enhanced fruit productivity with better fruit quality. They also noted that the Zn spray had a significant increased fruit ascorbic acid content and pH because it plays an active role in auxins biosynthesis. Similarly, Tariq *et al.* (2007) [46] and Ashraf *et al.* (2012) [4] reported that micronutrients foliar spray increased

the fruit numbers/tree, juice volume and ascorbic acid contents of sweet orange and Kinnow mandarin fruits. B foliar spray increases fruit set per cent and yield in several fruit trees species, such as almond, prune, olive and sour cherry (Slavko *et al*. 2001) [43]. This increment of tree yield might be due to the beneficial roles of B in pollination process (Lee and Kim 1991) [23]. Shivanandam *et al*. (2007) [38] reported that the Zn has been identified as a component of approximately 60 enzymes involved in the growth hormone promoter synthesis (auxin), which is directly linked with the improvement of the fruits quality of sweet orange. Likewise the effect of boron foliar spray on increased fruit quality might be due to its role in cell division, cell elongation, sugar metabolism and accumulation of carbohydrates (Sourour 2000 and Dutta 2004) [45, 12]. It is concerned with oxidation-reduction equilibrium in cells. Boron is also a heavy metal micronutrient. It is absorbed by plants in the form of boric acid (H_3BO_3). The role of micro- nutrients is a necessity in sweet orange as it is shy bearer and more prone to granulation under nutrients and moisture stress conditions.

Materials and Methods

The present investigations were conducted at the fruit orchard, Department of Fruit Science, College of Horticulture, VCSG Uttarakhand University of Horticulture and Forestry, Bharsar, Pauri Garhwal, Uttarakhand during the year 2016-17. The experiment was conducted to find out the most suitable combination of micronutrients for sweet orange cv. Mosambi in terms enriched fruit quality. This experiment was laid out in Randomized Block Design (RBD) with ten treatments and three replications. The treatment combinations consisted of three different micronutrients namely copper sulphate (0.3, 0.5, and 0.7%), zinc sulphate (0.5%) and boric acid (0.3, 0.5, and 0.7%) with three concentrations of each. The treatments were, T_1 (control)- Water spray,T_2- Zn (0.5%) +B (0.5%) + Cu (0.5%),T_3 - Zn (0.5%) +B (0.5%) + Cu (0.3%),T_4 - Zn (0.5%) +B (0.5%) + Cu (0.7%),T_5 - Zn (0.5%) +B (0.3%) + Cu (0.5%),T_6 - Zn (0.5%) +B (0.3%) + Cu (0.3%) T_7 - Zn (0.5%) +B (0.3%) + Cu (0.7%),T_8 - Zn (0.5%) +B (0.7%) + Cu (0.5%),T_9 - Zn (0.5%) +B (0.7%) + Cu (0.3%) and T_{10} - Zn (0.5%) +B (0.7%) + Cu (0.7%). The zinc was neutralized by lime for correction of pH. The micronutrients were applied as a foliar spray thrice at monthly interval from March to September 2016 and spray was given in the evening hours between 3.00-5.00 pm by using a knapsack sprayer**.** The required quantities of micronutrients were dissolved in water separately and then pH of these nutrient solutions was adjusted by lime and sprayed in four stages viz vegetative, before flowering, after fruit set stages (when it becomes berry sized) and at 40 days interval of second spray. (When fruit was plum size). The simple water spray was done on the tree under control treatment. In each spray treatment, Teepol was added as sticking agent in prepared solution for proper nutrient ‘absorption. The following observations as listed below were recorded.

The observations were recorded on different aspects viz. physical and chemical characteristics of fruits. Physical and chemical composition of sweet orange fruits with respect to peel thickness, the peel thickness was measured with the help of digital vernier caliper in millimeter and average was worked out for juice percentage. The juice was weight with the help electronic balance and the percentage of juice was worked out on the basis of total weight of fruit and weight of juice. Numbers of seeds per fruit were measured visually by counting the number of seeds and TSS content of juice samples under different treatments were measure by ATAGO hand refracto meter, however titratable acidity was measured using phenolphthalein indicator as per method of A.O.AC. (2007), however, reducing sugars non-reducing sugars, total sugars were determined by the methods as suggested by (Ranganna, 2006) [32].

Statistical Analysis: All obtained data were tested as one way analysis by the general linear model (GLM) and analysis of variance (ANOVA) technique. Treatments means were separated and compared using the least significant differences (LSD) at 0.05 level of significance according to Snedecor and Cochran (1989) [44]. The statistical analysis was performed using statistical analysis system (SAS 1988) [36].

Results and Discussion

Physico-chemical characteristics of fruit

Effect on peel thickness (mm): The perusal of the data in Table 4.1 revealed that among different treatments, maximum peel thickness (5.50 mm) was recorded with the treatment T_1 (control) while minimum peel thickness (4.20 mm.) was recorded with T_{10}. Patil *et al.* (2014) [30] observed the minimum peel thickness (0.48 mm) in Kinnow mandarin. The physical quality of citrus fruit indicate that the peel percentage in sweet oranges was significantly reduced by B and Cu application @ 0.7% and Zn treatments compared with the control treatment. The size of fruit was however significantly increased by all micronutrient treatments. The interactions between micronutrients with respect to size were found statistically significant. The overall results regarding physical quality of mosambi fruit indicate that B significantly decreased the peel and rag percentage and improved the size of mosambi fruit.

Effect on juice percentage: The data presented in Table 1 revealed that juice percentage was influenced significantly by application of different treatments, the application of T_7 treatment resulted in maximum juice percentage (28.16%), while minimum juice percentage (26.50%) was recorded with T_1 (control). Sajid *et al.* (2012) [35] reported that foliar application of zinc and boron significantly enhanced fruit juice content of sweet orange fruit. Application of micronutrients might have improved tree health by better sugar metabolism and translocation of assimilates and this could be the main reason for increase in juice content. It has been reported by various researchers that balanced nutrition considerably increased the juice content in the fruit of healthy trees. The result obtain in present investigations confirm the findings of many earlier researchers (Ghosh and Basra, 2000; Kulkarni 2004) [16, 21] in sweet orange. Likewise, Devi *et al*. (1997) [11] also found that juice content of sweet orange fruits was significantly increased when the plants were supplied with soil application at $ZnSO_4$, $FeSO_4$ at 50 g/tree and combined foliar spray of the micronutrients at 0.5% concentration.

Effect on number of seeds per fruit: The perusal of the data in Table 1 revealed that the numbers of seeds/fruit were influenced significantly by different treatments. The application of T_1 (control) recorded maximum number of seeds per fruit (11.33) while minimum numbers of seeds per fruit (8.10) were recorded with T_7. The results of present findings are supported by result of Kumar *et al*. (2013) who reported that foliar spray of $ZnSO_4$ 0.8% + borax 0.4% + NAA 50 ppm + GA_3 100 ppm was found best to reduce the seed per cent and seed pulp ratio which ultimately increased the yield per tree in guava cv. Chittidar.

Effect on ascorbic acid (mg/100 ml): The data presented in Table 1 revealed that ascorbic acid was significantly affected by different treatments application. The treatment T_7 resulted in maximum ascorbic acid (38.52 mg/100 ml) while minimum ascorbic acid (38.52 mg/100 ml) was recorded with T_1 (control). Sajid *et al.* (2012) [35] reported that foliar application of zinc and boron significantly enhanced fruit juice content and ascorbic acid of sweet orange fruit. Highest increase in the level of ascorbic acid contents was recorded in the fruits harvested from the trees which were sprayed with 0.3% boric acid + 0.5% zinc sulphate at sweet orange fruit. Zn plays an active role in the production of auxin in plant species (Alloway, 2008) [2] and the production of auxin increases ascorbic acid content and increase of ascorbic acid in Kinnow mandarin as reported by Nawaz *et al.* (2008) [27]. Similarly, foliar application of Zn sprays have also been reported to increase ascorbic acid contents in Balady mandarin trees (El-Menshawi *et al*., 1997) [14].

Effect on total soluble solids (^{0}Brix) The data presented in Table 1 revealed that total soluble solids (^{0}Brix) were influenced significantly by different treatments application. The treatment T_7 resulted in maximum total soluble solids (10.49 ^{0}Brix) while minimum total soluble solids (8.24^{0}Brix) was recorded with T_1 (control). The present investigation are supported by finding of Kumar *et al.* (2014). [31] who reported that foliar application of zinc and boron had significant effect on fruit quality of fruits in terms of TSS (12.99°B) which was obtained with highest doses of zinc and boron in combinations of guava fruits. The foliar spray of micronutrients (0.5%) +B (0.3%)+ Cu (0.7%) resulted in improvement of quality of fruit in terms of TSS content and it might be contributed to the fact that micronutrients directly play an important role in plant metabolism as zinc is needed in enzymatic reaction like hexokinase, formation of carbohydrate and protein synthesis (Pamila *et al*., 1992) [28]. Further boron facilitated sugar transport through boron-sugar complex and it also increase hydrolysis of saccharides into simple sugar (Shanmugavelu *et al*., 1973) [37] and copper play an important role in enhancement of photosynthetic efficiency that cause higher photosynthetic rate. The main end substrate of photosynthesis is sugar, so upsurge in the photosynthesis by combined action of Zn, B and Cu might lead to increase in the sugar compounds and causes more accumulation of total soluble solids in fruit juice. Thus, fruit quality in term of TSS content was improved by zinc, boron and copper. The findings of present study are in accordance with those of Babu and Yadav (2005) [5] in Khasi mandarin, Rawat *et al*., (2010) [33] in guava, Shukla *et al*., (2011) [39] in aonla, Sajid *et al*., (2012) [35] in sweet orange, Bhatt *et al*., (2012) [7] in mango, Eiada and Mustafa (2013) [13] in pomegranate, Singh *et al*., (2013) [40] in mango, Sohrab *et al*., (2013) in pomegranate, and Bakshi *et al*., (2013) [6] in strawberry. The higher total soluble solids could also be attributed to the efficient translocation of photosynthesis to the fruit by regulation of copper, boron and zinc as investigated by Ullah *et al*. (2012) [47].

Table 1: Effect of foliar spray of Zn, Cu and B on peel thickness, juice percentage, number of seeds per fruit, ascorbic acid and total soluble solids of sweet orange cv. Mosambi

Treatments	Peel thickness (mm)	Juice percentage (%)	Number of seeds/fruit	Ascorbic acid (mg/100 ml)	Total soluble solids (^{0}Brix)
T_1 (control) Water spray	5.50	26.11	11.33	35.86	8.24
T_2 Zn (0.5%) +B (0.5%) + Cu (0.5%)	4.60	27.31	8.85	36.65	9.61
T_3 Zn (0.5%) +B (0.5%) + Cu (0.3%)	4.67	26.31	9.06	36.12	8.78
T_4 Zn (0.5%) +B (0.5%) + Cu (0.7%)	4.72	28.01	9.46	38.06	10.02
T_5 Zn (0.5%) +B (0.3%) + Cu (0.5%)	4.75	27.12	9.55	36.41	9.19
T_6 Zn (0.5%) +B (0.3%) + Cu (0.3%)	4.45	27.13	9.17	36.48	9.28
T_7 Zn (0.5%) +B (0.3%) + Cu (0.7%)	4.20	28.16	8.10	38.52	10.49
T_8 Zn (0.5%) +B (0.7%) + Cu (0.5%)	5.15	27.96	10.84	37.26	9.85
T_9 Zn (0.5%) +B (0.7%) + Cu (0.3%)	4.94	26.54	10.56	36.24	9.00
T_{10} Zn (0.5%) +B (0.7%) + Cu (0.7%)	4.39	27.44	8.93	37.25	9.71
SE (d)	0.34	0.50	0.62	0.61	0.42
C.D. (0.05)	0.72	1.06	1.32	1.30	0.89

Zn - Zinc, B – Boron, Cu – Copper, * Significant

Effect on titratable acidity (%): The perusal of the data in Table 1 revealed that titratable acidity was influenced significantly by application of different treatments. Titratable acidity ranged from minimum (0.89%) in T_7 treatment to maximum (0.98%) in control (T_1 treatment). The present results are supported by finding of Shukla *et al.* (2011) [39] who reported that among borax concentrations (0.4%) proved most effective in increasing the TSS (10.75°Brix), total sugar (8.75%) and reduced titratable acidity (2.14%) in aonla fruits. Ullah *et al.* (2012) [47] revealed that acidity percentage of mandarin fruit might have been reduced due to higher synthesis of nucleic acids, on account of maximum availability of plant metabolism. The decrease in acidity content as result of micronutrient application in fruit juice samples might be due to their utilization in respiration and rapid metabolic transformation of organic acids in to sugars (Brahamachari *et al*, 1997) [8]. Similar results were obtained by Singh *et al*. (2003) [41] in pomegranate and Patil and Hiwarale (2004) [29] in acid lime.

Effect on reducing sugars (%), non-reducing sugars (%) and total sugars (%): The data presented in (Table 2) clearly indicates that the foliar spray of copper, boric acid and zinc sulphate had significantly increased the sugar content (reducing, non-reducing and total sugar) of Sweet orange fruits. In the present investigation of Sweet orange, the highest reducing sugar (4.95%), non-reducing (1.95%) and total sugar content (6.90%) were recorded with T_7 treatment. Whereas, the minimum reducing sugar (4.45%), non-reducing sugar (1.51%) and total sugar (5.96%) were recorded at control. The increase in sugars component by the foliar feeding of zinc and boron might be due to their active involvement in photosynthesis of metabolites and rapid translocation of sugars from other part of the plants to developing fruits. These results are in conformity with the findings of El-Rahman (2003) [15] in Naval Orange, and Kumar *et al*. (2014). [31]. In Guava. Bhatt *et al*. (2012) [7] reported that the trees sprayed with 0.5% borax showed maximum reducing sugar, non-reducing sugar and total sugar content in mango.

Table 2: Effect of foliar spray of Zn, Cu and B on titratable acidity, reducing sugars, non-reducing sugars and total sugars of sweet orange cv. Mosambi

Treatments	Titratable acidity (%)	Reducing sugars (%)	Non-reducing sugars (%)	Total sugars (%)
T_1 (control) Water spray	0.98	4.45	1.51	5.96
T_2 Zn (0.5%) +B (0.5%) + Cu (0.5%)	0.93	4.66	1.65	6.31
T_3 Zn (0.5%) +B (0.5%) + Cu (0.3%)	0.90	4.50	1.56	6.06
T_4 Zn (0.5%) +B (0.5%) + Cu (0.7%)	0.92	4.80	1.80	6.60
T_5 Zn (0.5%) +B (0.3%) + Cu (0.5%)	0.94	4.55	1.58	6.13
T_6 Zn (0.5%) +B (0.3%) + Cu (0.3%)	0.93	4.62	1.58	6.20
T_7 Zn (0.5%) +B (0.3%) + Cu (0.7%)	0.89	4.95	1.95	6.90
T_8 Zn (0.5%) +B (0.7%) + Cu (0.5%)	0.91	4.74	1.74	6.48
T_9 Zn (0.5%) +B (0.7%) + Cu (0.3%)	0.95	4.51	1.68	6.19
T_{10} Zn (0.5%) +B (0.7%) + Cu (0.7%)	0.92	4.71	1.72	6.43
SE (d)	0.01	0.09	0.09	0.15
C.D. $_{(0.05)}$	0.04	0.20	0.20	0.33

Zn - Zinc, B – Boron, Cu – Copper

Conclusion

The present investigations of foliar spray of Zn, Cu and B on Mosambi cultivar it revealed that the highest juice percentage, total soluble solids, reducing sugars, non-reducing sugars, total sugars, ascorbic acid and minimum titratable acidity, number of seeds, peel thickness were observed with the treatment T_7 [Zn (0.5%) + B (0.3%)+Cu (0.7%)]. Hence, it can be inferred that the application of micronutrients like Zn, Cu and B are beneficial in increasing the fruit quality of sweet orange cv. Mosambi. The treatment T_7 [Zn (0.5%) + B (0.3%) +Cu (0.7%)] was the best in terms of overall enhancement of fruit quality attributes of sweet orange.

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