

Pre-harvest Treatment of Zn & B Affects the Fruit Quality and Storability of Sweet Orange

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Received: December 24, 2011 / Published: December 20, 2012.

Abstract: Physico-chemical composition of sweet orange (*Citrus sinensis* L.) cv. blood red was determined in relation to different storage conditions and micronutrients application at department of horticulture, Agricultural University Peshawar, Pakistan during 2006-2007 and 2007-2008. The post-harvest quality of sweet orange was evaluated for 60 days storage with 20 days intervals. Fruit were harvested after the foliar application of zinc and boron in two consecutive seasons. The harvested fruits were stored at an ambient temperature (ATS) of 25 ± 2 °C and at low temperature storage (LTS) of 15 ± 2 °C with 60%-70% relative humidity (RH) for 60 days. Sweet oranges stored at LTS maintained better fruit quality than ATS. The foliar application of zinc and boron significantly enhanced fruit juice content, total soluble solids (TSS), ascorbic acid (AA) and non-reducing sugar (NRS) of fruit. However, fruit juice content, TSS and AA were observed significantly higher, when the fruit was treated with high zinc (1%) and low boron (0.02%). The percent of weight loss, disease incidence, TSS and reducing sugar (RS) increased with increasing the storage durations. A reduction was noted in fruit juice, AA and NRS with increasing the storage durations.

Key words: Zinc, boron, storage conditions, fruit quality, ascorbic acid, total soluble solids, sweet orange.

1. Introduction

Citrus is an important fruit crop in more than 135 countries. The world over production is about 102.64 million tones and the major fruit of Pakistan. Among all fruits, citrus holds number one position on the basis of area and production in the country. Pakistan is among the top 15 citrus producing countries of the world [1]. The annual production of citrus fruit in Pakistan was 2,294.47 thousand tons from an area of 199.369 thousand hectares in year 2007-2008 [2], yet, the availability of good quality fruit in domestic markets for extended duration limited due to improper pre- and post-harvest management. According to National Commission of Pakistan on Agriculture, the

post-harvest losses in fruits and vegetables range from 20% to 40% [3] or even greater [4]. The post-harvest losses in citrus fruits are estimated to about 9%-25% [5] and could be as high as 35% [6]. Citrus is the most important exportable fruit of Pakistan. Total export of citrus during 2003-2004 amounted to Rs.1771.34 million [7]. The success of citrus fruit export as an export commodity depends on the quality of produce and phyto-sanitary conditions set by the importing country and WTO and proper post-harvest management so that the produce can reach destination market without quality deterioration and pathogenic incidence [8]. Therefore, both pre- and post-harvest management technologies need to be optimized for quality fruit production and retention of quality during storage [6]. The adoption of poor production practices such as low yielding genotypes [9], deficiency of

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nutrients specially the micronutrients [10] and post-harvest problems such as poor sorting and grading [11], moisture condensation resulting in pathogen infestation, improper transportation and storage [5], and distant and time consuming marketing are the major causes of losses in quality and quantity [12]. In Pakistan, citrus are grown in micronutrients deficient soil, i.e., semi-arid or calcareous soils. The deficiency of micronutrients not only decreases the life span of the plants, production, but also influences the quality of sweet orange fruit [13]. The sweet orange fruit quality can be enhanced by the application of micronutrients (Zn & B), which increased the juice content [10] and other quality attributes such as total soluble solids (TSS), total sugars (TS), reducing sugar (RS), non-reducing sugar (NRS), ascorbic acid (AA) and TSS/acid ratio of papaya fruit [14]. The present study resulted in substantial increase in fruit yield and quality attributes of sweet orange with the foliar application of micronutrients (Zn & B). Thus, it is required that the storage performance of the fruit as well as storage conditions is optimized for maximum increase in shelf life of citrus fruits. While considerable research is conducted on the influence of calcium, Mg [15], sodium [16] and micronutrients [17] on fruit quality, very few studies have investigated the influence of zinc and boron on the storability of sweet orange fruits and the results have been quite non-persistent [18]. The present experiment was conducted to find out the main instrumental parameters affecting the acceptability of sweet orange and also to investigate the influence of zinc and boron on the storability of sweet orange fruits.

2. Materials and Methods

This work was carried out at Department of Horticulture, Agricultural University Peshawar, Pakistan during 2006-2007 and 2007-2008. The sweet orange cv. blood oranges were collected from orchid at Dargai, Malakand Agency, Pakistan and stored at

post-harvest laboratory, Department of Horticulture. The fruits were divided into two major groups and stored one at low temperature storage (LTS) (15 ± 2 °C) and other at ambient temperature (ATS) (25 ± 2 °C) with 60%-70% relative humidity for 60 days during two consecutive seasons (2007 & 2008).

2.1 Experimental

The experiment was laid out in randomized complete block design (RCBD) with factorial arrangement having three replications and nine treatments during 2007 and 2008. All the recorded data was from five randomly selected fruit taken periodically at 20 days intervals.

2.1.1 Treatments of Foliar Spray (F)

Treatment 1: Zn0 (0.00%) + B0 (0.00%);
 Treatment 2: Zn1 (0.05%) + B0 (0.00%);
 Treatment 3: Zn2 (1.00%) + B0 (0.00%);
 Treatment 4: Zn0 (0.00%) + B1 (0.02%);
 Treatment 5: Zn0 (0.00%) + B2 (0.04%);
 Treatment 6: Zn1 (0.05%) + B1 (0.02%);
 Treatment 7: Zn2 (1.00%) + B1 (0.02%);
 Treatment 8: Zn1 (0.05%) + B2 (0.04%);
 Treatment 9: Zn2 (1.00%) + B2 (0.04%).

2.1.2 Weight Loss (%)

The initial weight of each fruit was noted with the help of electronic balance. The average loss of weight was calculated as:

$$\text{Wt loss (\%)} = \frac{W_1 - W_2}{W_1} \times 100 \quad (1)$$

W_1 = weight of fresh fruits;

W_2 = weight of fruits after interval.

2.1.3 Percent of Juice Content (%)

Juice was extracted from each treatment with the help of juice extracting machine, weighed and the percentage was computed as:

$$\text{Juice content (\%)} = \frac{\text{Wt of Juice fruit}^{-1}}{\text{Average Wt of fruits}} \times 100 \quad (2)$$

2.1.4 TSS (Brix°)

TSS was measured with a brix refractometer. The juice from sample fruits was thoroughly mixed to make the contents homogeneous. A drop of juice was

placed on the slab of refractrometer. The rotation was observed through the eyepiece of the equipment.

2.1.5 AA (mg/mL), RS (%) and NRS (%)

AA (mg/mL), RS (%) and NRS (%) were determined by the standard method as reported in AOAC [19].

2.1.6 Percent of Disease Incidence (%)

Percent disease incidence in each replication of treatments was visually examined and counted. The disease percentage of fruits was calculated as:

$$\text{Disease incidence (\%)} = \frac{\text{Number of diseased fruits}}{\text{Total number of fruits}} \times 100 \quad (3)$$

2.2 Data Analysis

The data from different parameters were analyzed by analysis of variance (ANOVA) to observe the differences between the treatment and their interactions. In cases where the differences were significant, the means were further assessed for differences through least significant difference (LSD) test. Mstatc (Michigan State University, USA) software was used for ANOVA and LSD.

3. Results and Discussion

The results obtained on chemical composition of sweet orange fruit treated with the foliar application of zinc and boron, stored at different temperature conditions, are given in Tables 1 and 2.

3.1 Weight Loss (%)

Analysis of the data (Tables 1 and 2) revealed that the foliar application of zinc and boron, storage conditions (ATS & LTS) and storage durations significantly affected weight loss of sweet orange. Year as source of variation also had significant effect on weight loss of sweet orange. All interactions were non-significant for weight loss. The fruit weight loss consistently increased with increasing the storage durations. The maximum weight loss (5.87%) was

noted in fruits stored for 60 days followed by 40 and 20 days (Table 1). The LTS significantly decreased the weight loss (3.08%) compared to ATS (3.89%). Minimum weight loss (3.46%) was recorded in fruits when treated with high zinc (1%) and low boron (0.02%) in combination. Thus, the foliar application of high zinc and low boron in combination reduced the fruit weight loss. There was also significant decrease in weight loss from 3.64% in 2007 to 3.33% in the year 2008. Higher weight loss was recorded during 1st year (2007) as compared to 2nd year (2008) of the experiment (Table 1). High weight losses in citrus fruit are due to loss of moisture by evapotranspiration [20] during storage [21, 22]. The maximum weight loss at ATS may be due to high temperature, which results in higher rates of respiration and evapotranspiration [23] or internal breakdown and senescence during storage [24].

3.2 Fruit Juice Contents (%)

The data in Tables 1 and 2 showed that there was a significant effect of foliar spray, year effect, storage condition and storage durations on percent juice contents of sweet orange. The interaction of storage duration \times condition (D \times T) had significant influence on percent juice contents of sweet orange fruit, but rest of the interactions were non-significant. It was noted that the percent juice contents significantly decreased from 52.24% in fresh fruit to 35.78% in fruit stored for 60 days. The rate of decrease was only 1.97%, but increased to 7.91% and 6.58% between 20 to 40 and 40 to 60 days storage. High losses in juice contents might be due to damage to natural surface coating of sweet orange fruit during storage [25]. The losses in juice contents were significantly lower at LTS compared to ATS. The foliar spray significantly increased the percent juice contents of sweet orange fruit particularly when applied in combination. The maximum percent of juice contents (46.61%) was found in with high concentration of zinc and low concentration of boron. The interaction of foliar spray

Table 1 Effect of zinc and boron on physico-chemical changes of sweet orange during storage.

Treatments	Weight loss (%)	Juice content (%)	TSS (Brix ^o)	Reducing sugars (%)
Control	3.51 a	42.49 e	10.18 bc	10.22 ab
Zn1 @ 0.5%	3.49 ab	43.84 d	10.21 ab	10.16 b
Zn2 @ 1.0%	3.48 ab	45.17 bc	10.12 c	10.19 ab
B1 @ 0.02%	3.47 ab	44.72 cd	10.20 ab	10.24 a
B2 @ 0.04 %	3.51 a	45.73 abc	10.24 ab	10.22 ab
Zn1B1 (0.5% + 0.02%)	3.48 ab	45.24 bc	10.23 ab	10.17 b
Zn1B2 (0.5% + 0.04%)	3.50 ab	46.57 a	10.23 ab	10.20 ab
Zn2B1 (1.0% + 0.02%)	3.46 b	46.61 a	10.26 a	10.20 ab
Zn2B2 (1.0% + 0.04%)	3.47 ab	46.08 ab	10.22 ab	10.19 ab
LSD	0.04703	1.105	0.06371	0.06731
Storage duration (Days)	Weight loss (%)	Juice content (%)	TSS (Brix ^o)	Reducing sugars (%)
0	0.00 d	52.24 a	8.97 d	6.55 d
20	3.24 c	50.27 b	9.52 c	8.64 c
40	4.83 b	42.36 c	10.14 b	11.33 b
60	5.87 a	35.78 d	12.20 a	14.26 a
LSD	0.1177	0.7368	0.0424	1.3621
Storage duration (Days)	Non-reducing	Ascorbic acid (mg/mL)		
0	3.20 a	53.31 a		
20	2.77 a	46.63 b		
40	1.65 b	37.04 c		
60	0.62 c	26.08 d		
LSD	0.012	4.307		
Interactions of foliar spray and storage durations (F × D)				
	Juice content (%)	TSS	A. Acid (mg/mL)	
F × D	*Fig. 1	*Fig. 3	----	
T × D	*Fig. 2	----	*Fig. 4	

Mean followed by similar letter(s) in column do not differ significantly from one another;

NS = non Significant and * = Significant at 5% level of probability;

ATS = ambient temperature storage; LTS = Low temperature storage; F × D = interaction of foliar spray and storage duration; A.

Acid = Ascorbic acid; T × D = interaction of storage temperature and duration; Foliar application of Zn and B had non-significant effect on non-reducing sugar.

Table 2 Effect of storage temperatures (LTS & ATS) on the physico-chemical attributes of sweet orange during 2007 and 2008.

Storage conditions	Weight loss (%)	Juice content (%)	TSS (Brix ^o)	Reducing sugars (%)	Non-reducing (%)	Ascorbic acid (mg mL ⁻¹)	Disease incidence (%)
ATS (25 ± 2 °C)	3.89 a	43.48 b	10.21	10.21a	2.06	39.97 b	4.38 a
LTS (15 ± 2 °C)	3.08 b	46.84 a	10.20	10.19 b	2.06	41.56 a	1.77 b
Sig. level	*	*	NS	*	NS	*	*
Years	Weight loss (%)	Juice content (%)	TSS (Brix ^o)	Reducing sugars (%)	Non-reducing (%)	Ascorbic Acid (mg/mL)	Disease incidence (%)
2006-2007	3.64 a	42.78 b	10.15 b	10.20	2.37 b	40.04	3.15 a
2007-2008	3.33 b	47.54 a	10.27 a	10.20	2.74 a	41.49	3.00 b
Sig. level	*	*	*	NS	*	NS	*

and storage durations indicates that the fruit juice contents decreased with increasing storage durations irrespective of foliar spray (Fig. 1). The maximum decline was noted in juice contents in fruit stored at

ambient conditions with increasing the storage durations (Fig. 2). The increase in fruit juice contents of plant sprayed with micronutrients might be due to improved vegetative features such as low dieback

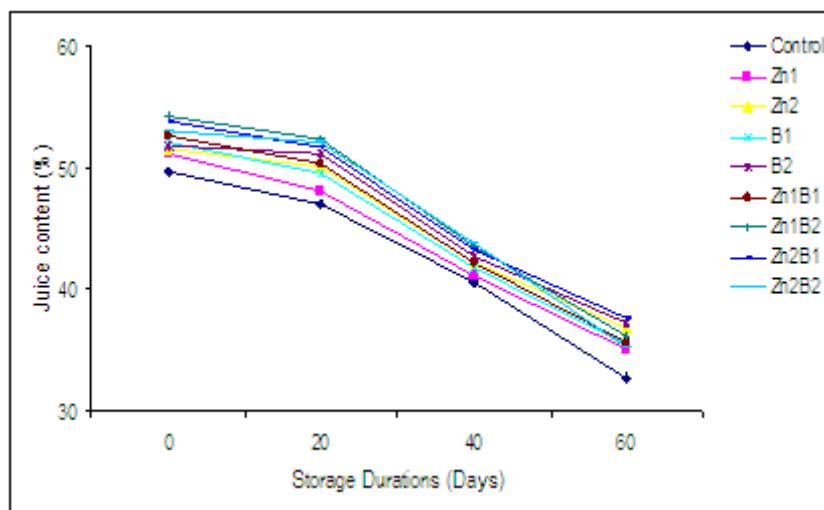


Fig. 1 Foliar spray (Zn & B) and storage duration affect the percent of juice content of sweet orange.

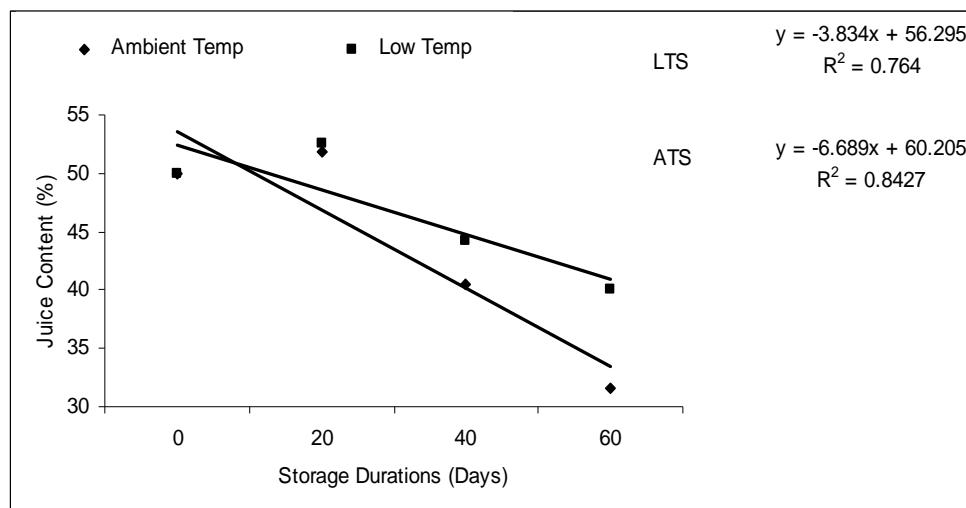


Fig. 2 Storage temperatures (LTS & ATS) and storage duration affects the juice content of sweet orange.

incidence [26], decreased rosette formation [27] and chlorosis [28]. The higher juice contents of fruits at LTS might be due to lower rate of evapotranspiration [29].

3.3 Total Soluble Solids (TSS)

Data concerning TSS of fruit juice are given in Tables 1 and 2. Zinc and boron as foliar application and storage duration had significant effect on TSS of the fruit. The effect of storage temperature/conditions was non-significant. The interactions of foliar spray \times storage duration ($F \times D$) had also a significant effect on TSS (Fig. 3) while other interactions had

non-significant response on TSS. A significant increase in TSS (12.20%) was recorded with 60 days storage as compared to fresh fruit (8.97%). The foliar application of zinc and boron mostly increased TSS of fruit juice. The TSS content of sweet orange fruits increased from 10.15% in 2007 to 10.27% in the year 2008. Thus, the year wise increase in TSS was 1.20% (Table 2). The increase in TSS of citrus fruit with increasing storage duration [22, 30] could be attributed to the conversion of complex sugars to simple sugars during storage [31] and moisture loss. The increase in TSS with the application of zinc or boron was possibly due to more efficient translocation

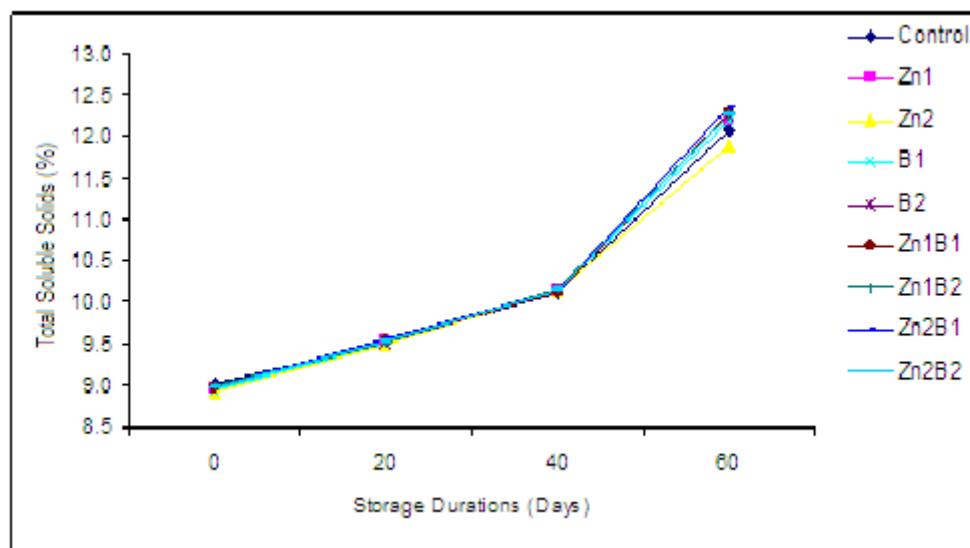


Fig. 3 Foliar spray (Zn & B) and storage duration affect TSS of sweet orange.

of photosynthate to fruit [32]. Boron also plays an important role in certain metabolic activities such as sugar transport and carbohydrate metabolism [33] and rapid translocation of photosynthetic products and minerals from other plant parts into developing fruits [34]. The ripening in citrus fruits is characterized by considerable physiological and biochemical changes in the fruit and the cell wall is subject to considerable alterations [35] leading to changes in carbohydrates compositions [36].

3.4 Percent of Reducing Sugars (RS)

The effect zinc and boron as foliar spray, year as a source of variation, storage durations and condition (temperature) were significantly effected the RS of sweet orange fruit (Tables 1 and 2). The interaction of storage durations and storage temperatures ($D \times T$) had a significant difference for RS, whereas, others interactions had no significant affect. A gradual increase in RS was observed with increasing storage duration. The maximum RS (14.26%) was recorded in fruits stored for 60 days as compared to fresh harvested fruits (6.55%). Different storage temperatures also significantly affected the RS. However, the maximum RS (10.21%) was recorded in fruits at ATS as compared to LTS (10.19%). The

interaction of storage durations and temperatures ($D \times T$) indicated that the RS significantly increased with increasing the storage duration irrespective of storage temperatures. The maximum RS (14.28%) was recorded when fruits stored for 60 days at ambient/room temperature followed by the fruits (14.24%) stored for 60 days under low temperature. This increase might be due to conversion of non-reducing sugar to reducing sugar, break down of cell wall (cellulose, hemicellulose and pectin) and loss of moisture during storage [35-37]. The response of the plants to zinc and boron as a foliar spray was significant but inconsistent in relation to RS of the fruit. However, maximum RS (10.24%) was noted in fruit treated with low concentration of boron (0.02%) alone. The plants sprayed with low concentration of zinc alone had less RS (10.16%). The difference in reducing sugars during 2007 and 2008 was also non-significant. These results are in line with Tariq et al. [10] and findings of Singh et al. [14] who found that Zn and B as foliar spray significantly increased the RS in papaya fruit.

3.5 Percent of Non-reducing Sugars (NRS)

Year as a source of variation, zinc and boron as foliar spray and storage durations significantly affect

the percent of NRS of sweet orange fruit (Tables 1 and 2). The interaction of foliar spray, storage duration and storage condition had non-significant effect on NRS of sweet orange. A gradual decrease was observed with increasing the storage durations from 0-60 days. The NRS decreased 3.20% in fresh fruits to 0.62% in fruits stored for 60 days. Since the increased storage of sweet orange fruits may result in hydrolysis of the cell wall and juice vesicles [35], it is likely to observe the decrease in NRS with increasing storage duration [37]. Zinc and boron as foliar application generally decreased NRS of the juice. The mean data revealed that the maximum NRS (2.08%) was recorded in fruits treated with high concentration of zinc (1%) alone and in control treatment. The minimum NRS (2.03%) was recorded in fruits treated with low concentration of boron (0.02%). The interaction of storage durations and temperatures ($D \times T$) showed an inconsistent response to NRS of the juice. The maximum NRS (3.20%) was recorded in fresh fruit stored at both ATS and LTS. The minimum NRS (0.59%) was noted in fruits stored for 60 days at ATS. Similarly, maximum NRS (2.74%) was observed in fruit from 2008 as compared to 2007 (2.37%).

3.6 Ascorbic Acid (AA)

The foliar application of zinc and boron, storage durations and temperatures significantly influenced the AA of sweet orange fruit (Tables 1 and 2). The interaction between storage duration and storage temperatures ($T \times D$) for AA was significant (Fig. 4). All other interactions and year as source of variation were non-significant. There was a consistent decrease in ascorbic acid of the fruit with increasing storage durations and temperature. The maximum ascorbic acid (53.31 mg mL^{-1}) was recorded in fresh fruit, which gradually decreased to the minimum of 26.08 mg mL^{-1} after 60 days storage. Also, the AA content was higher (41.56 mg mL^{-1}) in fruits at LTS as compared to ATS (39.97 mg mL^{-1}). The application of zinc and boron as foliar spray improved the AA

content of sweet orange. The maximum AA (41.29 mg mL^{-1}) was recorded in fruit when treated with high concentration of zinc (1%) in combination with low concentration of boron (0.02%), closely followed by the fruit (41.02 mg mL^{-1}) which received low concentration of zinc (0.5%) with low concentration of boron (0.02%) in combinations. The minimum AA (40.40 mg mL^{-1}) was noted in fruit of control treatment. The interaction of foliar spray and storage durations indicated that the foliar application of zinc and boron showed inconsistent response to AA. The interaction of foliar application of zinc and boron \times storage temperatures indicated that the maximum AA (42.51 mg mL^{-1}) was retained in fruit when treated with high zinc (1%) and low boron (0.02%), followed by fruits (41.82 mg mL^{-1}) which received low zinc (0.5%) and low boron (0.02%) in combination stored at LTS. The minimum AA (39.63 mg mL^{-1}) was recorded in fruits treated with only zinc in high concentration (1%) stored at ATS. AA is the most sensitive to destruction when the commodity is subjected to adverse handling and storage temperatures. Most of the fruits show reduction in AA during post-harvest ripening [38]. Losses in AA are enhanced by extended storage, high temperature and low relative humidity [39]. Ascorbate oxidase has been proposed to be the major enzyme responsible for the enzymatic degradation of AA [40]. The fruits stored at low temperature decrease enzymatic activity and delay the ripening, which in turn protect the degradation of AA [41]. The interaction of foliar spray and storage durations ($F \times D$) indicated that there was decrease in AA with increase in storage durations without any response of foliar spray of zinc and boron in both seasons. The AA is one of the most labile vitamin in fruits and vegetables that tend to decline during storage [42] and temperature during storage [43, 44].

3.7 Disease Incidence (%)

Data concerning disease incidence are shown in Tables 1 and 2. The statistical analysis of the data

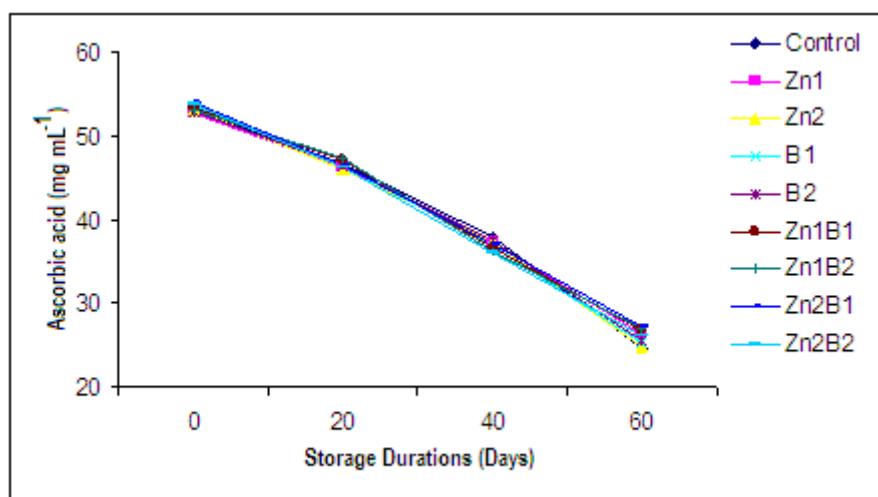


Fig. 4 Foliar spray and storage duration affect the ascorbic acid (mg mL^{-1}) of sweet orange.

exhibited that the storage duration and temperatures significantly affected the disease incidence of sweet orange. The foliar application of zinc and boron had non-significant effect on disease incidence. Year as a source of variation showed significant effect on disease incidence. Their interaction had non-significant response to disease incidence of sweet orange during storage. Disease incidence increased significantly with increasing the storage durations. The minimum disease incidence on 0 day storage increased to 1.73% and 3.88% with 20, 40 days storage and finally increased to the maximum of 6.70% when storage duration was extended to 60 days. The low temperature was found superior as compared to ambient/room storage in reducing the disease incidence of sweet orange. Since the development of disease is influenced by temperature and is generally low at low temperature, it is likely to observe less disease incidence at low temperature storage than room temperature [28, 44]. The application of zinc and boron does not influence the disease incidence in sweet orange fruits during storage. But the year-wise data indicated that disease incidence was significantly higher in the year 2007 (3.15%) which decreased to 3.00% in the year 2008. The decreased disease incidence can not be attributed to high zinc or boron concentration in the plants rather to relatively wet

harvesting season during 2007.

4. Conclusions and Recommendations

From this research, it is concluded that sweet orange fruit should be stored at the lowest possible temperature to retain the quality of sweet orange fruit. Since the sweet orange fruits are chilling sensitive, it is desirable to determine the lowest safe temperature for its storage so that the storage life is enhanced without the detrimental effects of chilling injury. The foliar application of zinc and boron in combination (1.00% Zn and 0.02% B) are recommended for the enhancement of fruit quality of sweet orange but not as a technique to extend the storage life. The decrease in different quality attributes during storage was not significantly influenced by the application of zinc and boron. The sweet orange fruit should be stored at low temperature ($15 \pm 2^\circ\text{C}$) to retain the quality attributes for longer period of time as compared to ambient storage condition ($25 \pm 2^\circ\text{C}$).

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