



Full Length Article

Changes in Fruit Quality Attributes of Ambient and Low Temperature Stored “Kinnow Mandarin” with Respect to Geographic Locality

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Abstract

Two separate studies were conducted to compare and evaluate the storability/shipping potential and fruit quality of “Kinnow” mandarin fruit, harvested from four districts [Sargodha (SGD), Khanewal (KHW) Toba Tek Singh (TTS) and Vehari (VHR)] of Punjab province, Pakistan. Fruits were harvested at commercial maturity and evaluated for physico-chemical quality under ambient storage conditions (25±1°C; 60-65% relative humidity/RH) for seven days and cold storage conditions [4±1°C; 85-90% RH] for thirty and sixty days. Under ambient shelf storage, peel color was improved while decrease in peel smoothness and fruit weight was observed in all districts after shelf storage. Moreover, fruit harvested from SGD had higher fruit firmness and juice contents while weight loss was significantly higher in fruits harvested from KHW as compared to other three districts. Total soluble solids/TSS, TSS/TA (titratable acidity) ratio and vitamin C increased in fruits harvested from VHR and TTS after shelf storage. Under cold storage conditions, fruits harvested from TTS and VHR exhibited higher TSS, TSS/TA ratio, acidity and vitamin C after 2nd removals (60 days of cold storage). However, fruit samples of SGD showed higher juice contents after 1st and 2nd removal while fruit firmness was significantly higher in fruits from VHR after 2nd removal. No symptoms of chilling injury (CI) were observed after 1st removal; however, it was found in samples only from KHW and VHR after 2nd removal. Total phenolic content (TCP) were improved with increase in storage period while antioxidants activity showed non-significant response. In conclusion, fruits harvested from VHR and TTS observed better fruit quality during ambient, and cold storage conditions; and therefore fruits harvested from these two districts showed good potential for export purpose beside SGD. © 2019 Friends Science Publishers

Key words: Antioxidant activity; Geographical locations; ‘Kinnow’ fruit; Rainfall; Storage life; Quality

Introduction

Pakistan produces several important citrus fruits in handsome quantity and “Kinnow” mandarin (*Citrus nobilis* Lour × *Citrus deliciosa* Tenora) among them is a valuable citrus cultivar. According to numerous estimates, Pakistan holds 95% share of globally produced “Kinnow” mandarin at current time (Tahir, 2014). This mandarin variety is appreciated nationally and around the globe due to its good blend of total soluble solids (TSS) to acid ratio, natural antioxidants, physicochemical properties and sensory attributes (Chopra *et al.*, 2004). Pakistan generated revenue of worth 173 U.S. million dollars by exporting 393 thousand tons of “Kinnow” mandarin, during 2014-15 (A.M.I.S., 2015). Among four provinces of the country,

Punjab province occupies dominant position for citrus production (95%) and about half (48%) of this production is produced by a single district *i.e.*, Sargodha (SGD). More than 90% of the Punjab production is comprised of single cultivar *i.e.*, “Kinnow” mandarin (Usman *et al.*, 2018). ‘Kinnow’ is also being grown in other districts of Punjab prominently Toba Tek Singh (TTS), Khanewal (KHW) and Vehari (VHR) *etc.* (Khalid *et al.*, 2018).

Same citrus cultivars harvested from different geographical regions differ in qualitative as well as in quantitative attributes. “Kinnow” fruit harvested from different regions and at different times varies in fruit quality and storage/shipping behavior in India (Meena and Yadav, 2001) and prone to number of postharvest disorders and diseases during storage (Sonkar *et al.*, 1999).

Different geographical regions have different agro-climatic conditions and have significant impact on fruit quality and this impact is more important than any other factor (*i.e.*, soil, genetic factors and cultural practices *etc.*) (Ladaniya, 2008). Regions characterized with high temperatures (*i.e.*, tropical regions), the internal fruit quality of mandarins is generally appeared to be very poor, with low acidity and intermediate sugars contents (Albrigo, 2004). Degradation in green pigments is required to obtain natural yellow colour; and this process is suitable in warmer climate of tropics, and thus grapefruit and lemons are most suited in these regions (Albrigo, 1990). Absence of chilling interval coupled with soil and air temperatures below the threshold, which prompts dormancy, might be one of the preventive factors in obtaining good yield of citrus (Poerwanto and Inoue, 1990).

Postharvest losses in fruits and vegetables are estimated between 35-40% in Pakistan mainly due to poor postharvest practices. These losses can be minimized in citrus by adopting wax coating, low temperature storage, modified atmosphere (MA) storage, controlled atmosphere (CA) storage *etc.* (Benítez *et al.*, 2015). Low temperature storage is still a standard practice for the storage of 'Kinnow' mandarin in Pakistan. To extend the storage life of mandarins, storage at 4-8°C coupled with 90-95% relative humidity (RH) is highly desired (Kader and Arpaia, 2002) and is highly helpful for preserving the fruit quality (Arnon *et al.*, 2015).

Almost 90% 'Kinnow' exports come from district SGD that are routed through sea. It is believed that (with no scientific base), 'Kinnow' from this district SGD got good postharvest storage life. This perception has narrowed down the opportunity of sourcing of fruit, establishment of 'Kinnow' processing industry and exports from other 'Kinnow' growing district (KHW, VHR and TTS *etc.*). We hypothesized that other 'Kinnow' growing districts also have potential for good postharvest storage life besides district SGD. To the best of our knowledge, no scientific work is reported previously in 'Kinnow' mandarin for the comparison of postharvest storage life among different 'Kinnow' growing districts of Punjab, Pakistan. Hence, this study was designed to determine the postharvest ambient and cold storage quality profile of 'Kinnow' fruit being grown in different 'Kinnow' growing districts of Punjab to widen the 'Kinnow' export base for the welfares of 'Kinnow' industry and nation.

Materials and Methods

Two separate experiments were conducted to compare storage/shipping potential and fruit quality of 'Kinnow' fruit harvested from four districts of Punjab, Pakistan *i.e.*, Sargodha (SGD) (32° 5' 1.468" N 72° 40' 18.695" E), Khanewal (KHW) (30° 18' 14.162" N 71° 55' 47.564" E), Toba Tek Singh (TTS) (30° 58' 21.612" N 72° 29' 5.95" E) and Vehari (VHR) (30° 2' 38.954" N 72° 20' 38.648" E). The cultural and environmental profile of selected

districts were also obtained from Khalid *et al.* (2018) and expressed in Fig. 6, 7 and 8. In first experiment, storage potential and quality of fruits was evaluated at ambient storage conditions (25 ± 1°C) while cold storage (4 ± 1°C; 85-90% RH) potential and fruit quality was assessed in second experiment.

Orchard Selection in Districts

Six commercial orchards (replications) were selected from each district (treatments) and this selection was done following the procedure outlined by Khalid *et al.* (2018). Fruit harvested from each district were washed with tap water, air dried at room temperature and shifted to Postharvest Research and Training Center (P.R.T.C.), Institute of Horticultural Sciences (I.H.S.), University of Agriculture, Faisalabad (U.A.F.), Pakistan after appropriate packaging.

Experiment 1: Storage Potential of 'Kinnow' Fruit at Ambient/Shelf Conditions (25 ± 1°C)

The 540 uniform fruits, free from insect pest and disease attack were harvested from each districts at commercial fruit maturity (total number of fruits from all four districts = 540 fruit per district × 4 districts = 2160 fruits). The harvested fruits from each district were divided into two equal lots *i.e.*, each lot contains 270 fruits; 6 replications (No. of orchards in each district) each replication contains 45 fruits. One lot was used for fruit physico-chemical analysis immediate after harvest while other lot was stored at ambient/shelf conditions *i.e.*, (25 ± 1°C) for seven days preceding fruit physico-chemical analysis. Shelf life and fruit quality at pre and post storage stages was also compared among four districts.

Experiment 2: Storage Potential of 'Kinnow' Fruit at Cold Storage Conditions (4 ± 1°C; 85-90% RH)

This experiment was conducted to evaluate the cold storage/shipping potential and fruit quality at cold storage conditions (4 ± 1°C; 85-90% RH) among selected four districts. The 540 uniform fruits, free from insect pest and disease attack were harvested at commercial fruit maturity from each district (total number of fruits from all four districts = 540 fruit per district × 4 districts = 2160 fruits). The harvested fruits from each district were divided into two equal lots *i.e.*, each lot contains 270 fruits; 6 replications (No. of orchards in each district) each replication contains 45 fruits). These lots were stored under cold storage conditions. One lot was analyzed for fruit physico-chemical characteristics after 30 days (1st removal) while second lot was analyzed after 60 days (2nd removal) of cold storage followed by seven days of shelf storage respectively. Comparison of shelf life and fruit quality at harvest, 30 and 60 days after cold storage was compared among four districts. Data for quality attributes at harvest was used of experiment 1.

Fruits External Quality Attributes

Peel smoothness, rind color, fruit diameter and fruit firmness was measured by adopting the procedure outlined earlier by Khalid *et al.* (2018).

Fruits Internal Quality Attributes

Total soluble solids (TSS) were measured by using digital refractometer (Model Atago-PAL-1 Tokyo, Japan) and expressed as °Brix. Titratable Acidity (TA) was determined by following the method outlined by Khalid *et al.* (2018) and presented as % citric acid. The ratio of TSS:TA was assessed by dividing TSS to TA. Ascorbic acid (vitamin C) content of fruit juice was determined by following the procedure of Ruck (1969) and total sugars in the fruit juice were analyzed by the following the method depicted by Khan *et al.* (2009) and expressed as percentage.

Statistical Analysis

Collected data of both experiments were analyzed following two-way ANOVA (districts and storage time) to judge the overall significance of data using statistix v® 8.1 software, while least significant difference (LSD) test ($P \leq 0.05$) was used to compare the differences among treatments means.

Results

Experiment 1: Storage Potential of 'Kinnow' Fruit at Ambient/Shelf Conditions ($25 \pm 1^\circ\text{C}$)

Physical characters: Peel color of "Kinnow" fruits on the first day of harvest did not show significant differences among all districts; meanwhile, after ambient storage "Kinnow" fruits harvested from TTS exhibited significantly higher peel color score than other districts (Fig. 1-A). Peel smoothness was decreased at shelf storage; while, differences for peel smoothness were also found non-significant among all districts (Fig. 1-B). Fruit firmness was decreased significantly over the time during shelf storage; whereas, significantly higher fruit firmness was observed in fruits harvested from district VHR at the day of harvest (2.32 score) and after shelf storage (2.19 score) (Fig. 1-C). Similar trend was observed for fruit diameter and for fruit juice percentage where maximum diameter and juice percentage was recorded at harvest; however, statistical analysis of the data did not show significant differences at harvest and after shelf life evaluation (Fig. 1-D; 1-E). Moreover, higher fruit weight loss was observed in fruits from district KHW (5.58%) among all districts (Fig. 1-F).

Bio-chemical characters: Increasing trend was recorded in total soluble solids (TSS) in all districts during shelf storage; however, significantly higher TSS was observed in district TTS (11.78 °Brix) and VHR (11.68 °Brix) after shelf storage (Fig. 2-A). Titratable acidity (TA) did not vary significantly

among all districts before and after shelf storage (Fig. 2-B). TSS/TA ratio varied considerably at harvest and after shelf storage in all four districts. TSS/TA ratio was increased after shelf storage. Maximum TSS/TA ratio was observed in district TTS (12.02) followed by district SGD (11.54) and VHR (11.0) after shelf storage (Fig. 2-C). Likewise, "Kinnow" fruit harvested from different districts showed significant variations in vitamin C contents with maximum vitamin C being noted in SGD and VHR districts while lowest vitamin C was recorded in "Kinnow" fruits harvested from TTS district (Fig. 2-D). Total sugars of "Kinnow" fruit also varied significantly among all four districts; whereas, decreasing trend was obvious from total sugar percentage at harvest to total sugar percentage after 7 days of shelf storage. Moreover, higher total sugars were observed in fruits collected from SGD (7.01%) followed by TTS and VHR (6.33% and 6.25% respectively) after shelf storage (Fig. 2-E).

Experiment No. 2: Storage Potential of 'Kinnow' Fruit at Cold Storage Conditions ($4 \pm 1^\circ\text{C}$; 85-90% RH)

Physical characters: Fruit firmness was continuously decreased in "Kinnow" fruits under cold storage regardless of harvest districts; however, significantly higher fruit firmness was observed in "Kinnow" fruits from district VHR at 1st and 2nd removal obtaining 2.31 score and 2.06 score, respectively (Fig. 3-A). Juice percentage of "Kinnow" fruit was decreased substantially from harvest till the end of cold storage; while, all the districts showed significant variances with highest juice percentage observed in fruits from district SGD after 30 and 60 days of cold storage (42.45% and 38.96% respectively) followed by district VHR (Fig. 3-B). No symptoms of chilling injury were observed at first removal (30 days of cold storage) however, significantly higher percentage of chilling injury symptoms were recorded in "Kinnow" fruits harvested from district KHW and VHR (44.44% each) at second removal *i.e.*, after 60 days of cold storage (Fig. 4).

Biochemical characters: Increasing trend was recorded in TSS of "Kinnow" fruit with the increase in storage time in all districts; however, significantly higher TSS was recorded in fruits from district TTS (12.25 °Brix) followed by VHR (11.95 °Brix) after 60 days of cold storage (Fig. 5-A). While, TA of "Kinnow" fruits were significantly decreased in all four districts with the increase in storage time; whereas, significantly higher acidity was observed in "Kinnow" fruits harvested from district VHR (0.92%) after 60 days of cold storage (Fig. 5-B). TSS/TA ratio of "Kinnow" fruit juice was increased substantially from the day of harvest to 60 days of cold storage. "Kinnow" fruits harvested from district SGD showed significantly higher value of TSS/TA ratio (15.17) followed by TTS and VHR after 60 days of cold storage (Fig. 5-C). Vitamin C being a key nutritive constituent of "Kinnow" fruit was maximum at the day of harvest that was

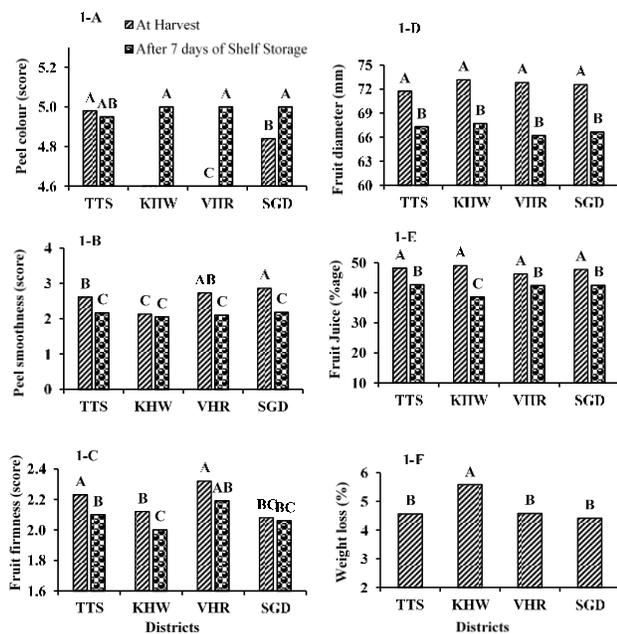


Fig. 1: External quality attributes and physiological weight loss in ‘Kinnow’ fruit from different growing districts under ambient conditions

Here TTS = Toba Tek Singh; KHW = Khanewal; VHR = Vehari; SGD = Sargodha

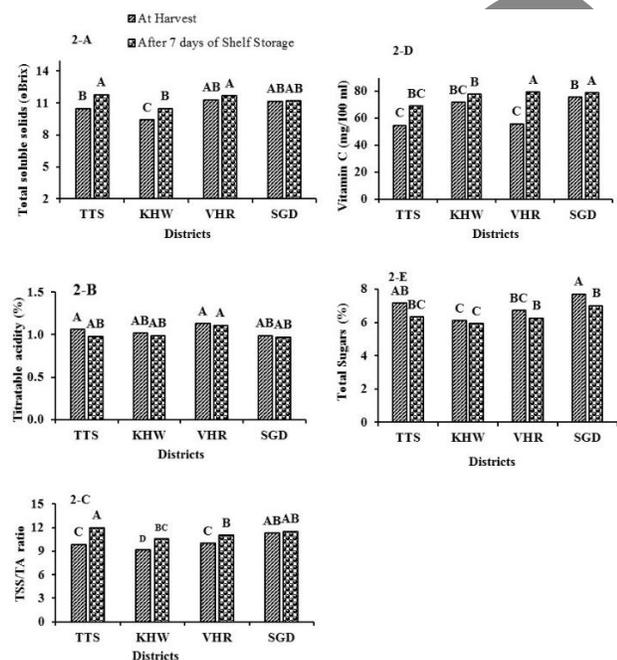


Fig. 2: Internal quality attributes of ‘Kinnow’ fruit from different growing districts under ambient conditions

Here TTS = Toba Tek Singh; KHW = Khanewal; VHR = Vehari; SGD = Sargodha

decreased significantly till 30th days of cold storage; however, upon further storage from 30-60 days’ vitamin C level was raised. Meanwhile, fruits harvested from VHR district exhibited significantly higher Vitamin C (71.5 mg/100 mL)

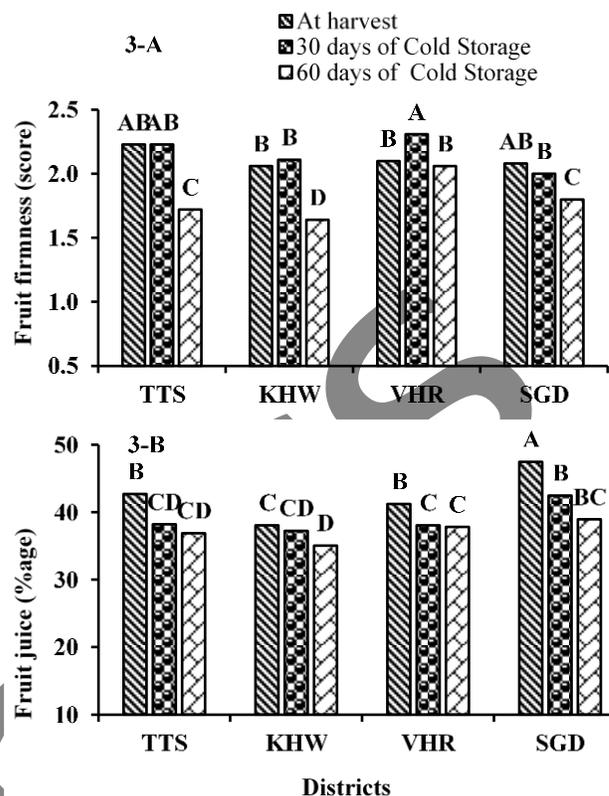


Fig. 3: Comparison of fruit firmness and juice contents (%) at harvest, after 30 and 60 days of cold storage among different ‘Kinnow’ growing districts

Here TTS = Toba Tek Singh; KHW = Khanewal; VHR = Vehari; SGD = Sargodha

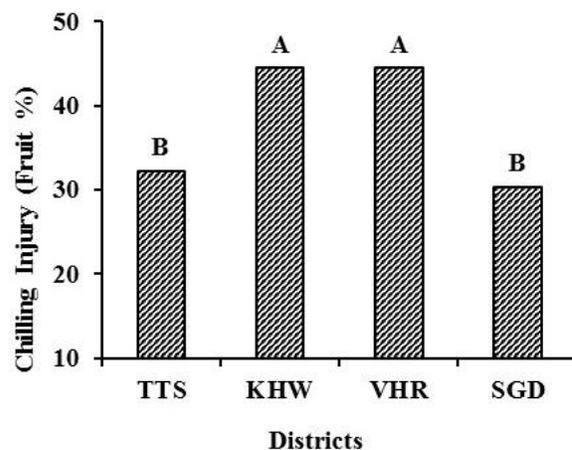


Fig. 4: Chilling injury (fruit %) after 60 days of cold storage

Here TTS = Toba Tek Singh; KHW = Khanewal; VHR = Vehari; SGD = Sargodha

than other districts after 60 days of cold storage (Fig. 5-D). Meanwhile, total sugar percentage of the ‘Kinnow’ fruit was at lowest at the day of harvest and went on to substantially highest level after 60 days of cold storage in all districts that shows the increasing tendency of total sugar under long term cold storage conditions.

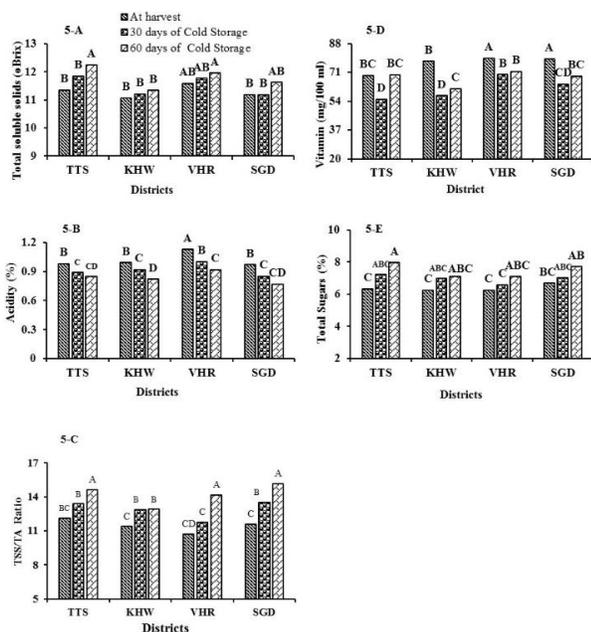


Fig. 5: Internal fruit quality attributes at harvest, after 30 and 60 days of cold storage among different 'Kinnow' growing districts

Here TTS = Toba Tek Singh; KHW = Khanewal; VHR = Vehari; SGD = Sargodha

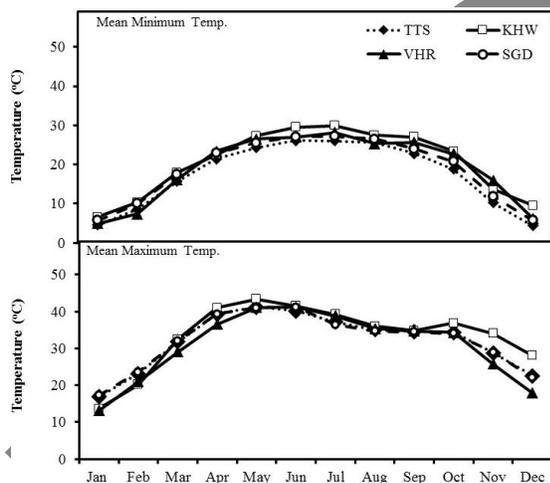


Fig. 6: Monthly mean minimum and maximum temperatures of districts of Punjab during growing season (2010)

Here TTS = Toba Tek Singh; KHW = Khanewal; VHR = Vehari; SGD = Sargodha

Meanwhile, "Kinnow" fruits harvested from TTS district exhibited significantly higher total sugar percentage compared to KHW, VHR and SGD districts (Fig. 5-E). Whereas, "Kinnow" fruit samples also showed increasing tendency in total phenolic contents (TPC) and antioxidants irrespective of district of harvest under storage period. Among all districts, "Kinnow" fruits harvested from TTS exhibited significantly higher TPC than all other districts; while, anti-oxidative capacity of all districts did not exhibit significant variation (Table 1).

Table 1: Total phenolic contents (TPC) and antioxidants activity (IC₅₀) at different storage times in different districts of Punjab

Districts	Storage time (Days)	Total phenolic contents (TPC) ($\mu\text{g/mL}$)	Antioxidants (IC ₅₀)
Toba Tek Singh	0	74.11 f	93.56
	30	110.31 cd	141.21
	60	213.55 a	310.59
Khanewal	0	84.02 ef	86.3
	30	124.38 bc	134.29
	60	133.4 b	292.04
Vehari	0	83 ef	88.48
	30	123.46 bc	136.36
	60	131.29 b	299.49
Sargodha	0	83 ef	91.7
	30	123.46 bc	139.37
	60	131.29 b	309.91
		*	NS

*= Significant, NS = Non-Significant; n=6

Discussion

Mandarin fruit is sensitive to storage conditions and its quality is greatly influenced by storage duration and environment (Singh *et al.*, 2004; Obenland *et al.*, 2011). Results of this study revealed that fruits harvested from district VHR had better quality after 7 days of shelf life storage compared with the fruits harvested from other districts. Better quality of fruits harvested from VHR district could be due to better adaptability and suitable environmental conditions along with availability of soil micro and macronutrients (Meena and Yadav, 2001). Improved peel color of "Kinnow" fruits harvested from VHR district could be due to ideal growing condition like hot summer, cold winter accompanied by low rainfall that contributed better peel color of the fruit. Less relative humidity and rainfall in VHR district (Fig. 8) might have contributed to better physical quality of "Kinnow" fruits. Higher leaf Mn contents in VHR might also have contributed to better peel colour (Zekri and Obreza, 2014; Khalid *et al.*, 2018). Moreover, peel smoothness and firmness might be related higher Ca contents as previously reported by Khalid *et al.* (2018) in "Kinnow" fruits harvested from VHR. Diameter of "Kinnow" fruit did not vary significantly among different districts; however, reduction in diameter of "Kinnow" fruit during storage could be related to loss in moisture and breakdown of tissues due to increased metabolic activities under ambient condition. Previously, Zekri (2011) observed higher firmness in citrus fruits grown under dry climatic conditions.

The prolonged storage time is associated with greater physiological weight loss of fruits. While, juice percentage did not vary significantly in all districts during ambient storage conditions. On the other hand, in 2nd experiment juice percentage of "Kinnow" fruit was significantly higher in SGD district. In both experiments (ambient and cold storage), juice percentage was decreased continuously under ambient as well as under cold storage conditions that

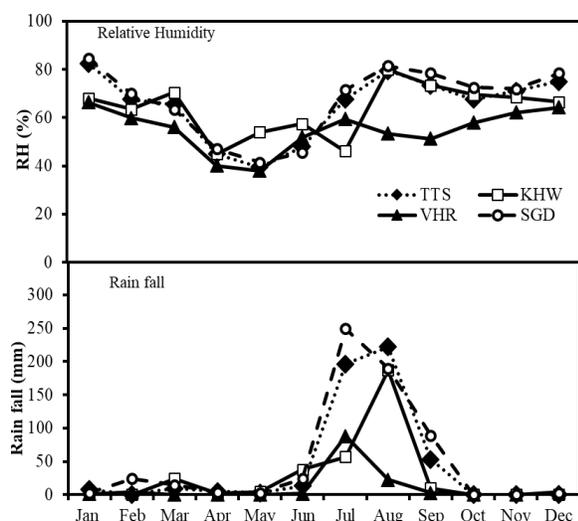


Fig. 7: Monthly mean relative humidity (RH) and precipitation/rainfall in districts of Punjab during growing season (2010)

Here TTS = Toba Tek Singh; KHW = Khanewal; VHR = Vehari; SGD = Sargodha

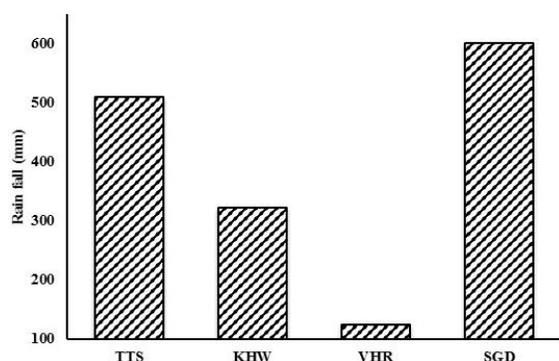


Fig. 8: Total precipitation in districts of Punjab during growing season (2010)

Here TTS = Toba Tek Singh; KHW = Khanewal; VHR = Vehari; SGD = Sargodha

might be due to continuous moisture loss during storage. Comparable findings were reported in Malvasio's mandarin where long term cold storage resulted in substantial weight loss (D'Aquino *et al.*, 2001). Nonetheless, reduction in TA during cold storage might be due to consumption of organic acids as respiration substrates during cold storage as depicted by Xi *et al.* (2016). Leahu *et al.* (2013) and Marcilla *et al.* (2006) also reported similar findings in citrus juice. However, TSS were continuously increased during ambient as well as long term cold storage conditions that might be due to the breakdown of complex organic metabolites into sugars (Mahajan and Goswami, 2004).

Vitamin C contents of "Kinnow" fruits were increased during ambient storage conditions; whereas, in 2nd experiment there was a dip in vitamin C contents during 30 days' interval and then there was continuous elevation in vitamin C contents regardless of district of harvest. Initial decrease in vitamin C during cold storage might be ascribed

to metabolic changes converted organic acids into sugars (Shiri *et al.*, 2011). However, increase in vitamin C at ambient and after 60 days of cold storage describes the response of "Kinnow" fruit under stress conditions and using antioxidant compound in response to oxidative stress. In current study, total sugars at ambient conditions were decreased while total sugars under long-term storage conditions were continuously increased. Increase in total sugars during cold storage could possibly due to conversion of acids in to sugars (Pareek *et al.*, 2015). Similar, results were also reported by Pérez *et al.* (2005) where they observed decline in sugar percentage during storage of *Hernandina* mandarins.

There was no chilling injury after 30 days of cold storage, however, storing "Kinnow" fruits for upto 60 days resulted in significantly higher chilling injury in KHW and VHR districts that could be due to higher temperature and low rainfall conditions during summer (Fig. 6-8). Shyam *et al.* (2004) described that temperature below 10 can cause chilling injury to mandrins; while 'Kinnow' fruits that are exposed to 4°C endure obvious chilling injury symptoms (Bajwa and Anjum, 2007). Higher TPC in TTS district may be attributed to suitable environmental conditions (Fig. 6-8) that resulted in accumulation of more TPC; while continuous elevation in TPC under cold storage describes the defense mechanism of 'Kinnow' fruit under low temperature oxidative stress. Previously, Madiwale *et al.* (2011) reported that phenolic content depends upon the growing location and sample extraction methods (Rumbaoa *et al.*, 2009). While increase in TPC under cold storage conditions could be due to low temperature stress that might have raised phenolic compounds *via* phenylpropanoid pathway by activation of phenylalanine ammonia-lyase (PAL) (McCollum and McDonald, 1992). Some substances might had produced during cold storage that might have resulted in increment in TPC upon reaction with FC reagent (Huang *et al.*, 2005).

Conclusion

Locality and their ecological factor significantly influence the storage/shipping potential and quality of 'Kinnow' fruit. Results revealed that fruits harvested from district VHR and district TTS showed good quality parameters like fruit firmness, total soluble solids (TSS), TSS:TA ratio, lower weight loss *etc.* during both ambient and cold storage conditions. Therefore, fruits harvested from these two districts are also good for export purpose beside SGD.

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