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RESEARCH PAPER

Study to determine the effect of spacing and varieties on fruit quality of citrus

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Key Message: Kinnow and Musambi species of citrus were evaluated by planting at 3 different spacing. Trees planted at 11 × 22 ft spacing showed optimum fruit development and quality attributes.

Abstract: To evaluate spacing impact on fruit quality, Kinnow mandrin (*Citrus reticulata* blanco) and Sweet orange Cv. Musambi (*Citrus sinensis* L.) were selected and planted at three different plant spacing i.e. T_1 (11 × 22 ft), T_2 (11 × 11 ft) and T_3 (22 × 22 ft) at Postgraduate Agriculture Research Station (PARS), University of Agriculture Faisalabad. It was observed that trees planted at 11 × 22 ft spacing showed optimum fruit development and quality attributes. Maximum number of fruits per plant (26.98), average fruit diameter (65.48mm), average fruit weight (141.11 g) with maximum fruit juice percentage (51.48%) pH of juice (5.52), TA (1.31%) and reducing sugars (2.30 %) were observed in 11 × 22 ft plants spacing. Highly dense plantation of trees at 11 × 11 ft resulted in highest average peel weight (36.40 %), average pulp weight (45.08%), ascorbic acid (6.85 mg/100 g), total sugars (8.85%) and non-reducing sugars (7.21%). 22 × 22 ft trees contained more TSS: TA (8.94). It was also observed that Musambi plants showed best results for most of the parameters as compared to Kinnow. The interaction effect of varieties and planting space showed the maximum number of fruits per plant (29.73) and maximum juice weight percentage (64.23%) in Musambi plants planted at 11 × 22 ft. The interaction effect of Musambi planted at 11 × 11 ft distance had more pulp weight percentage (51.07%) and peel weight percentage (41.99%). It was concluded that trees planted at 11 × 22 ft presented better results as compared to 11 × 11 ft and 22 × 22 ft distance for both varieties.

Keywords: Citrus, Kinnow, Musambi, high density, yield, quality, vigor

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Introduction

In 2200 BC, Indo-Chinese origin of citrus was cited first time in the Chinese literature. Citrus is leading fruit crop stands at 13th position in production around the globe. 157,979.26 thousand tonnes is the production of citrus around the world. Citrus is the top ranked fruit crop in Pakistan with respect to both area and production. 22 hundred thousand tonnes of citrus is harvested from the area of 201.47 thousand hectares approximately (Food and Agriculture Organization [FAO], 2019).

Moreover, citrus widely dominates by the farming of Kinnow in Pakistan. Furthermore, sweet orange also dominated the citrus industry, which was harvested on huge scale, few years ago. 98% of the total citrus production is contributed by Punjab in which, production of Kinnow is 70% of the total citrus production of Punjab. The major citrus growing areas are Faisalabad, Hyderabad, T.T. Singh, Sargodha, Multan, Peshawer and Sahiwal districts. Kinnow of Pakistan is exported to Indonesia, Iran, Afghanistan, Russia, Gulf States and China, but unfortunately, Pakistan exports a little to European markets because of high quality demand of European markets (Memon, 2017).

World's population is ever-increasing rapidly; there is always a problem for every government to make availability of fruits for large number of populations within a limited area. Proper plant density is also a better practice for the provision of sunlight, nutrients, and moisture, necessary for quality and better production of crop (Sanjib et al., 2002). Thus, to overcome this issue, it is required to increase the production as well as quality of Kinnow Mandarin with limited agricultural land and inadequate resources. Researchers around the globe have focused their research work towards determining the optimum spacing, agro techniques and varieties in various crops of fruits for the implementation of an important concept known as HDP (Saxena & Sharma, 2004). To utilize limited resources, researchers in Japan, USA, South Africa, Israel, Mexico, Brazil, Italy and Spain have oriented their research towards HDP (Goswami et al., 1993). For easy management of dwarf plants, there must be a check on plants' size in HDP (Bevington & Bacon, 1977). Researchers are focusing on finding out the optimal density of plants to get high quality fruit, yield and profit (Srivastava & Singh, 1999).

Hundreds of elite varieties of citrus with extensively genetic diversities are under the observation of scientists (Amar et al., 2011; Polat et al., 2012). Its popularity due to taste and nutritional values motivates to examine and ameliorate its characteristics (Lagou et al., 2018). It is important commercial crop with high output to farmer (Li et al., 2020). The overall scenario for fruit cultivation in Pakistan is dominated by different cultivars of Citrus in relation with production and area. Among different citrus cultivars, Kinnow shares maximum with 60% production followed by Musambi (Altaf & Khan, 2009). The significance of fruit for human health is reflected by its high consumption per capita (Meza et al., 2020). But unfortunately, production of citrus with respect to requirement is very low due to decrease in agriculture land along with inadequate resources. Various physiological (Cao et al., 2020) and soil problems also reduce yield importantly salinity (Pérez-Jiménez et al., 2020).

Increase in world population also resulted in more demand for citrus fruit because of its nutritional value and ever-loving taste. Citric acid, an important acid of various fruits, found naturally in citrus (Zhou et al., 2020). To overcome low production problem, world trend has been moving towards high density plantation. Appropriate planting distance ameliorates the tree behavior and fruit quality. Different management and biological aspects were considered more important because of their correlation. Light is important external factor to ameliorate fruit health (Ruiz et al., 2020). Planting space has been managed in such a way that proper light penetration along with aeration can be optimized. This results in better fruit quality and tree health. Increased plant population plays important role to overcome water availability problems and low land for fruit crops. Increased density of plants also decreases the unproductivity period and increases fruit yield with good quality (Srivastava & Singh, 1999). Keeping this point of view, the purpose of the study was to find the optimum spacing among Sweet Orange and Kinnow to achieve maximum number of better quality fruits with early return for the agro-ecological environment of Punjab.

Materials and Methods

The experiment was conducted at PARS, IHS, UAF during 2014-2015 02 years. The lab work was done at the Pomology Lab, IHS, UAF. 5 years old 180 trees of Sweet oranges and Kinnow mandarin of moderate health and vigor grafted on *Citrus jambhiri* L. rootstock were grown at 3 different spacings. 1140 gram urea and 1630 gram SSP was applied in all treatments. The experiment consisted of following three treatments.

Factor A:
V_1: Kinnow
V_2: Musambi
Factor B:
T_1: Plant spacings of 11 × 22 ft
T_2: Plant spacings of 11 × 11 ft
T_3: Conventional density 22 × 22 ft

Determination of physical attributes and physiological parameters

Twelve fruit were harvested from all side of full maturity stages. The fruit size (length and Diameter) was measured by Vernier caliper (Oo & Aung, 2018). Fruit yield was measured in terms of average from number of fruits harvested from each plant. Total fruit weight (kg) was calculated by weighing balance and average of result was noted. Peel weight, juice weight and pulp weight were calculated in percentages by using the following formulae:

$$\text{Peel weight } (\%) = \frac{\text{Average peel weight}}{\text{Average fruit weight}} \times 100$$

$$\text{Juice weight } (\%) = \frac{\text{Average juice weight}}{\text{Average fruit weight}} \times 100$$

$$\text{Pulp weight } (\%) = \frac{\text{Average pulp weight}}{\text{Average fruit weight}} \times 100$$

Total soluble solids (TSS), titratable acidity (TA) and ripening index (TSS: TA ratio)

Fruit juice was extracted by manual extractor and filtered with Whatman® filter paper No.1. This juice was used for measurement of pH, TSS and TA. TSS: TA ratio measured by dividing the resulting values of TSS with TA. TSS was observed by using Abby's refractometer (Abbès et al., 2011; Schweiggert et al., 2011; Safdar et al., 2012). For the determination of TA of fruit, 10 ml juice was taken. It was then titrated against 0.1 N NaOH till juice achieved pink color (Hortwitz, 1960) TA was determined according to the following formula:

$$\text{Titratable acidity } (\%) = \frac{0.1\text{N NaOH} \times 0.0064 \times 100}{\text{ml of juice used}}$$

Ascorbic acid (Vitamin C) and pH

pH of juice was measured by using digital pH meter. For vitamin C (mg/100 g), 10.00 ml juice and 90.00 ml of 0.4%

Oxalic acid solution was taken in volumetric flask. It was filtered and 10.00 ml aliquot was separated. Some distilled water was added and titrated against 2.60 dichlorophenolindophenol sodium salt hydrate (0.04%) dye until light pink color appeared. Ascorbic acid was measured by using the following formula:

$$\text{Ascorbic acid} = \frac{1 \times R1 \times V1 \times 100}{R \times W \times V}$$

R = ml of dye for titration against 2.5 ml of reference solution

(l ml standard ascorbic acid + 1.5 ml 0.4% Oxalic acid)

R_1 = ml of dye for titration against V_1 of aliquot

V = Volume of the aliquot made by 0.4% oxalic acid

V_1 = ml of juice taken

W = ml of aliquot used for titration

Determination of sugars

10.00 ml juice was taken in volumetric flask. 10.00 ml potassium oxalate (20%) solution, 100 ml distilled water and 25 ml lead acetate solution (25%) and were added to it. Required volume (250 ml) was made by adding distilled water. This sol was then filtered and filtrate was used for further treatment. This filtrate was titrated against 10 ml Fehling's solution until brick red color appeared. Then added 1% methylene blue (2-3 drops) with continuous addition of aliquot and titrated until brick red color obtained. Total aliquot used was measured and reducing sugars were calculated.

For total sugars determination, 25.00 ml aliquot was taken in volumetric flask. 5 ml concentrated HCl and 20 ml distilled water was added to it. Solution was kept overnight. This resulted in complete hydrolysis and converted the non-reducing sugars into reducing sugars. On next day, 0.1 N NaOH was used to neutralize the solution. Phenolphthalein was also used as indicator and volume was made with distilled water. This solution was titrated against 10.00 ml Fehling solution for calculation of total sugars.

Calculations were made according to the following formulas.

$$\text{Reducing sugars (\%)} = 6.25 \times \frac{X}{Y}$$

$$\text{Total sugars (\%)} = 25 \times \frac{X}{Y}$$

$$\text{Non} - \text{Reducing sugars (\%)} = 0.95 \times (\text{Total sugars \%} - \text{Reducing sugars \%})$$

Where 'X' is the volume (ml) of standard sugar solution titrated against 10 ml Fehling's solution and 'Y' is the volume (ml) of sample aliquot used against 10 ml Fehling's solution.

Statistical analysis

Data was analyzed statistically by two factor factorial design. Treatment means were tested using least significant difference test at 5% probability level (Steel et al., 1997).

Results and Discussion

Fruits per plant

Among all treatments, plants of T_1 (11 × 22 ft), gave the maximum, 26.98 fruits per plant, while minimum fruits were observed in T_2 (11 × 11 ft). Whereas the varieties V_2 (Musambi) gave more production than V_1 i.e. maximum (21.88) fruits per plant in V_1 were observed as compared to 18.11 fruits in V_2 (Fig. 1). It was concluded that Musambi gave maximum fruits per plant in the spacing of (11 × 22 ft) while minimum fruits per plant were observed in denser spacing. Furthermore, our results verified the findings of the Boswell et al. (1970); Nasir et al. (2006); Monga et al. (2011) who came to know that minimum number of fruits would be achieved if the spacing among plants was reduced, owing to the various factors like limited nutrients availability to each plant, reduction of fruiting area and overlapping of shoots and roots.

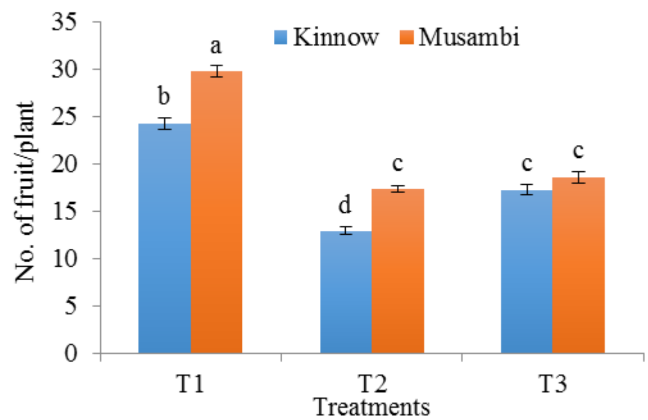

Fig. 1 Effect of plant spacing on of fruit per plant in Kinnow and Musambi

Fruit diameter

Diameter of fruit has commercial importance in trade and marketing of citrus. In plants of treatment T_1 (11 × 22 ft) attained maximum fruit diameter (65.48 mm) that was significantly more than the other treatments while, T_2 (11 × 11 ft) had minimum fruit diameter i.e. 56.69 mm (Fig. 2). Whereas V_1 (Kinnow) variety achieve maximum fruit diameter of 66.91 mm was statistically higher than V_2 (Musambi). In close plantation (11 × 11 ft) reduces in fruit size, may be due to more nitrogen utilization. Furthermore, results of the Currie et al. (2000) verified by our findings that spacing among plants is directly proportional to the fruit size. As, spacing among the plants was increased, fruit size was increased accordingly whereas, by reducing the spacing, reduced fruit diameter was achieved.

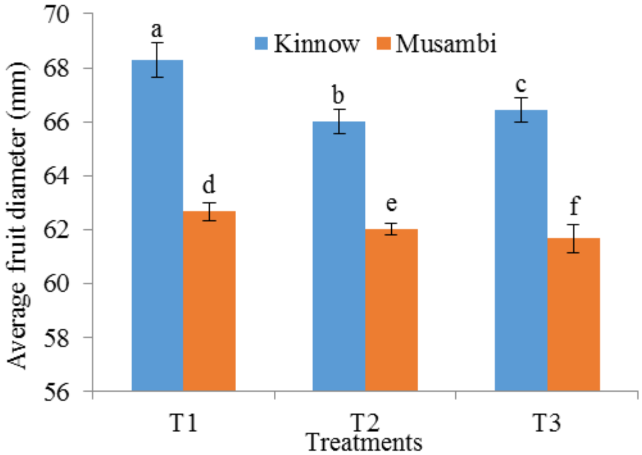

Fig. 2 Effect of plant spacing on average fruit diameter (mm) in Kinnow and Musambi

Fruit weight

Plants of T_1 (11 × 22 ft), gave the maximum, 132.12 g fruits weight that was significantly more than other treatments' plants while, T_2 (11 × 11 ft) had minimum fruit weight i.e. 103.75 g (Fig. 3). Whereas the varieties V_2 (Musambi) gave maximum fruit weight than V_1 i.e. maximum (141.11 g) fruits weight in V1 were observed as compared to V_2 (Kinnow). It was concluded that in dense spacing (11 × 11 ft) fruit weight was reduced because of less number of fruits on plants. The results of Bassal (2009); Nasir et al. (2006) verified our findings that density of plants is inversely proportional to the fruit weight. Fruit weight increased as we decreased the density of plants. Similarly, more fruit weight was achieved by increasing the spacing among plants.

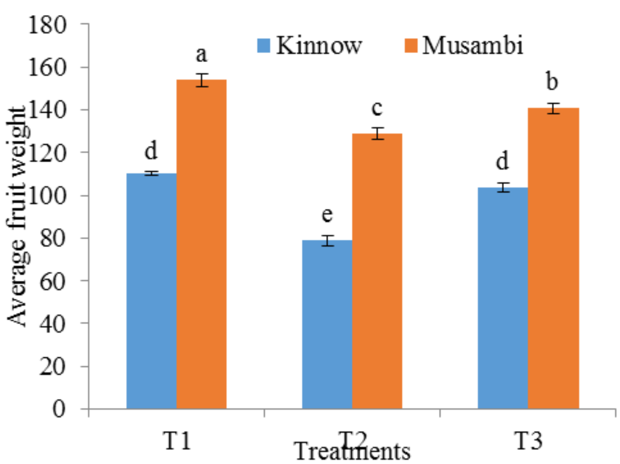

Fig. 3 Effect of plant spacing on average fruit weight (g) in Kinnow and Musambi

Pulp weight percentage

Among treatment's, fruit of T_2 (11 × 11 ft), gave the maximum, 45.08% pulp weight (Fig. 4), while minimum pulp weight 32.526% was observed in T_1 (11 × 12 ft). Whereas the varieties V_2 (Musambi) gave more pulp weight percentage than V_1 (Kinnow) i.e. maximum (42.13 %) pulp weight in V1 were observed as compared to 35.53% fruits in V2. The interaction effect exhibited that in T_2 (11 × 11ft), V_2 (Musambi) achieved maximum pulp weight of 51.07% and V_1 (Kinnow), planted at T_1 (11 × 22 ft), had minimum pulp weight i.e. 30.05%. Pulp weight of Musambi planted at T_2 (11 × 11 ft) was improved significantly. Pulp weight is directly proportional to fruit weight. Our findings are also in accordance with previous research results, which concluded that fruit of high density planted trees produce better fruit diameter and pulp weight (Singh et al., 2012; Umar et al., 2017).

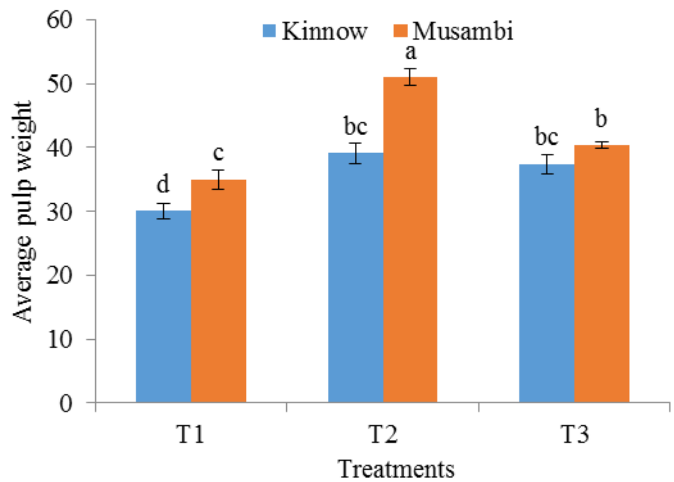

Fig 4 Effect of plant spacing on average pulp weight (%) in Kinnow and Musambi

Peel weight percentage

Treatment T_2 (11 × 11ft) attained maximum peel weight (36.40%) that was significantly more than the other treatment (Fig. 5). T_3 (22 × 22ft) had minimum peel weight i.e. 24.16%. Whereas V_2 (Musambi) variety achieve maximum peel weight of 34.12% was statistically higher than V_1 (Kinnow) 23.29%. Plants were remained un-pruned in all treatments. Our results were also confirmed by Umar et al. (2017) who reported that un-pruned plants at 11 × 11 ft distance produced the maximum peel weight.

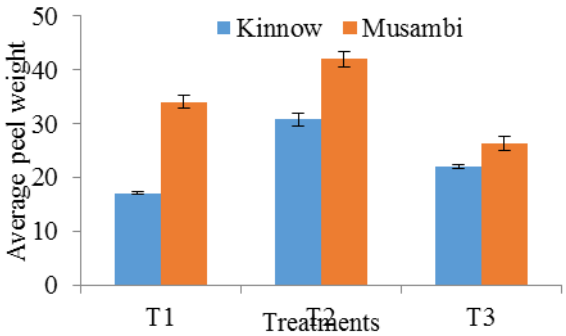

Fig. 5 Effect of plant spacing on average peel weight (%) in Kinnow and Musambi

Juice weight percentage

T_1 (11 × 22 ft) had maximum juice weight i.e. 51.48% as compared to T_2 (11 × 11ft) which had minimum juice weight i.e. 35.79% (Fig. 6). V_2 (Musambi) produced maximum fruit juice i.e. 53.07% as compared to the minimum fruit juice produced by V_1 Kinnow i.e. 33.05%. Interaction effect depicted that maximum juice weight i.e. 64.23% was attained by V_2 (Musambi) plants harvested in T_1(11 × 22 ft) as compared to the minimum juice i.e. 28.64% got from Kinnow harvested in T1(11 × 22 ft). Results depicted that 11 × 22 ft plant density in Musambi improved juice weight significantly. The various factors which have impacts on juice weight include nutritional imbalance, environmental factors and harvesting time of fruits. Moreover, the results of Nasir (2006) verified our findings that juice weight can be improved by the provision of Nitrogen and by the reduction of Phosphorus.

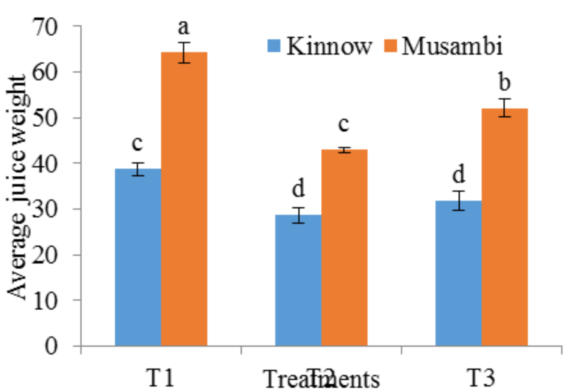

Fig. 6 Effect of plant spacing on average juice weight (%) in Kinnow and Musambi

Total soluble solids

T_3 (22 × 22ft) plants had maximum TSS i.e. 10.61 that was observed higher than T_2 (11 × 11ft) treatment plants having minimum TSS of 8.69. V_1 (Kinnow) had maximum TSS i.e. 10.59 that was significantly more than that of V_2 Musambi (8.68) TSS (Fig. 7). We came to the conclusion that TSS is directly proportional to the density of plants. Furthermore, results of Sharma et al. (1992); Nawaz et al. (2007) were in accordance with our findings that with the increase of spacing among plants the TSS will significantly increase while, by reducing the density of plants, TSS will be reduced accordingly.

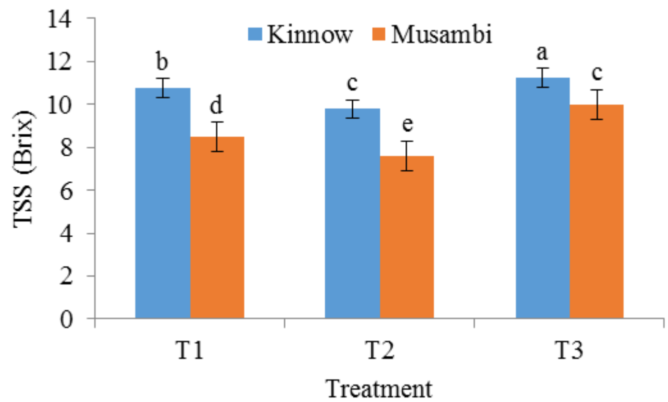

Fig. 7 Effect of planting densities on total soluble solid (˚Brix) in Kinnow and Musambi

pH of juice

In T_1 (11 × 22ft.) plants had maximum pH of juice i.e. 5.53 which was significantly more than other treatments' plants (Fig. 8). T_2 (11 × 11ft) had minimum pH of juice i.e. 4.13.V_2 (Musambi) had higher pH of 5.28 than that of Kinnow which was 4.43. Non-significant results were shown by interaction effect. Results highlighted that pH of juice was significantly increased in 11 × 22ft plant density. Furthermore, the results of Tachibana (1996); Nasir et al. (2006) were in accordance with our findings that plant density is directly proportional to the pH of fruit. If the spacing of the plants is increased, the pH will be increased and by the reducing the spacing among plants, the pH will be reduced accordingly.

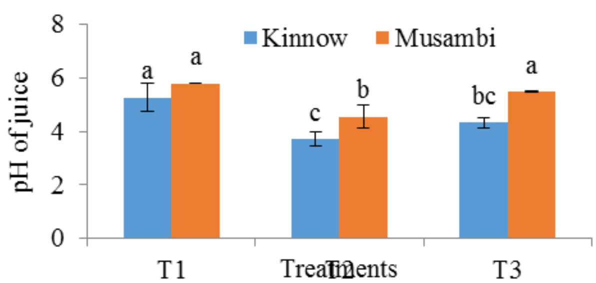

Fig. 8 Effect of plant spacing on pH of juice in Kinnow and Musambi

Titratable acidity

Acidity is important for determine the specific taste of citrus cultivars including Kinnow Mandarin. Acidity in citrus fruit is very important factor in overall juice quality and in determining the time of fruit harvest. Moving forward, acidity plays an important role in determining the taste of Kinnow Mandarin. Further, acidity can not only be ignored in determining the quality of juice but also plays a significant role in finding harvest time of fruit (Fig. 9). T_1 (11 × 22 ft) had the maximum titratable acidity of 1.31% that was significantly more than the other treatments' plants. T_2 (11 × 11 ft) had minimum titratable acidity of 1.18%. V_1 (Kinnow) had maximum titratable acidity of 1.65% that was significantly higher than V_2 Musambi. Non-significant results were shown by interaction effect. Results reveled that plant density (11 × 22 ft) significantly affected titratable acidity. Moreover, results of Nasir et al. (2006) and Nawaz et al. (2007) were in accordance with our findings that titratable acidity is directly proportional to density of plants. By increasing the spacing between plants titratable acidity will be increased accordingly while decreased titratable acidity will be achieved by decreasing the spacing among plants.

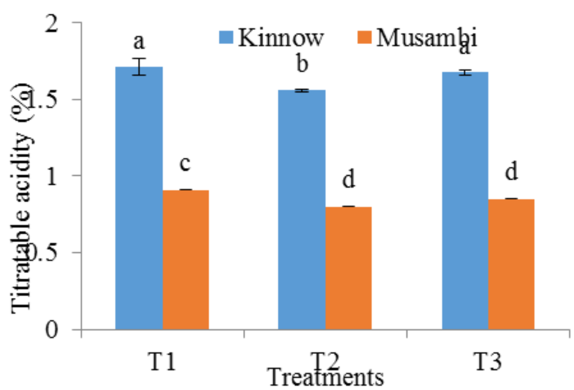

Fig. 9 Effect of plant spacing on TA (%) in Kinnow and Musambi

TSS/TA ratio

Widely spaced (22 × 22ft) plants of T_3 had maximum TSS/TA ratio of 8.94% which was significantly more than the other treatments' plants (Fig. 10). T_1 (11 × 11ft) plants had minimum TSS/TA ratio of 7.41%. V_2 (Musambi) had maximum TSS/TA ratio of 9.48% which was significantly higher than Kinnow whereas, V1 had minimum TSS/TA ratio of 6.60%. Non-significant results were shown by interaction effect. It was concluded that TSS/TA ratio was significantly increased by increase of plant spacing. Furthermore, results of Sharma et al. (1992) and Nasir et al. (2006) verified our results that TSS/TA ratio of fruits is directly proportional to the density of plants. TSS/TA ratio of fruits were increased by the increase of plant spacing while by decreasing the plant spacing, TSS/TA ratio was also reduced accordingly.

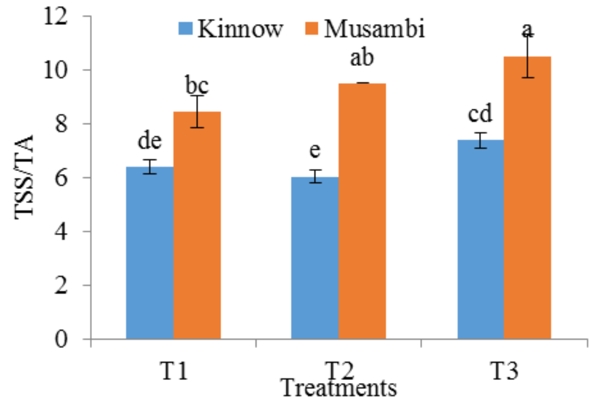

Fig. 10 Effect of plant spacing on TSS: TA in Kinnow and Musambi

Ascorbic acid

Vitamin-C, a useful antioxidant, is a vital constituent of our food. It is helpful in saving humans from various deadly diseases. Each citrus species has specific concentration of vitamin-C. Quantity of vitamin-C is dependent upon different factors out of which harvesting time of fruit, environmental factor, age and vigor of plants are significant. Dense spacing (11 × 11ft) of treatment (T_2) had maximum ascorbic acid of 6.85% which was significantly more than the other treatment plants (Fig. 11). T_1 (11 × 22ft) had minimum ascorbic acid of 5.91%. V1 (Kinnow) had maximum ascorbic acid i.e. 6.64% which was significantly more than the Musambi. V_2 had minimum ascorbic acid of 6.44%. Non-significant results were shown by interaction effect. Thus, concluded that, plant spacing is inversely proportional to the ascorbic acid. Furthermore, results of Nawaz et al. (2007) and Nasir et al. (2006) verified our findings that closer the plants, more will be the ascorbic acid as compared to widely spaced plants.

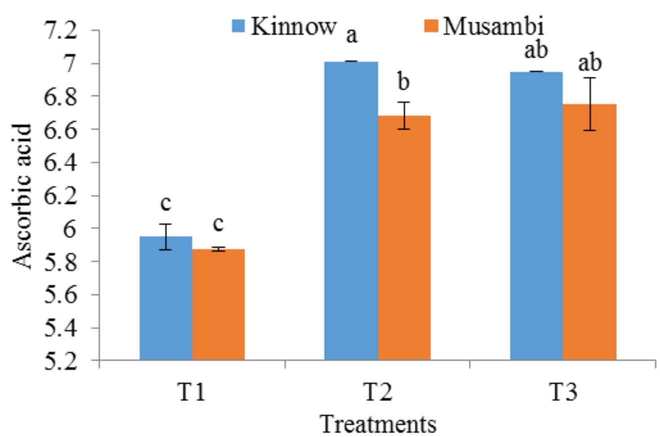

Fig. 11 Effect of plant spacing on ascorbic acid in Kinnow and Musambi

Total sugars

Maximum total sugars 8.85% were noted in T_2 (11 × 11ft) treatment plants which were significantly more than other treatments' plants (Fig. 12). T_1 (11 × 22ft) treatment had minimum total sugar of 7.23% whereas, V_2 (Musambi) variety had maximum total sugar i.e. 8.71% which was significantly higher than the other V_1 Kinnow variety (7.46%). Non-significant results were shown by interaction effect. Thus, concluded that plant spacing is inversely proportional to the total sugar. Moreover, results of Nawaz et al. (2007) were confirmed by our results that as we decrease the plant spacing, total sugars will be increased. Plants harvested in dense spacing had more total sugars as explained by Zekri (2000) and Nasir et al. (2006) that not only Nitrogen utilization has strong effects on the quality of fruits but also the plants harvested at close spaces will have more total sugars.

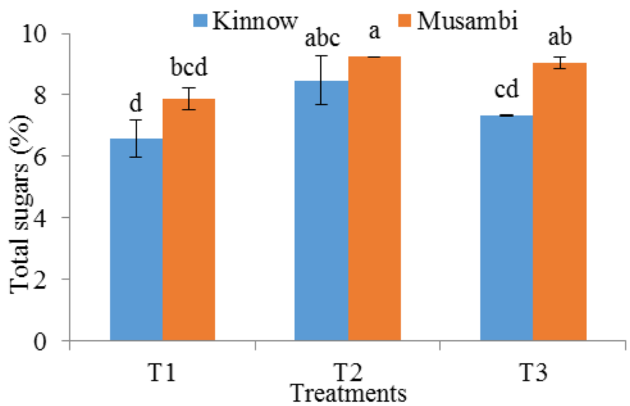

Fig. 12 Effect of plant spacing on Ascorbic acid in Kinnow and Musambi

Reducing sugars

Wider spacing is directly proportional to reducing sugars. Widely spaced plants T_1 (11 × 22ft) resulted in maximum reducing sugars i.e. 2.31% and statistically more than the plants of all other treatments (Fig. 13), along with this T_2 (11 × 11ft) resulted in minimum reducing sugar i.e. 1.65%. 2.26% (maximum reducing sugars) was observed in V_1 (Kinnow) and that was significantly higher than other variety. V2 had minimum reducing sugar of 1.78%. Non-significant results were shown by interaction effect. Thus, concluded that, reducing sugar is directly proportional to wider spacing. Furthermore, results of Nawaz et al. (2007) and Nasir et al. (2006) were confirmed by our findings that maximum reducing sugars will be achieved by maximum widely spaced plants.

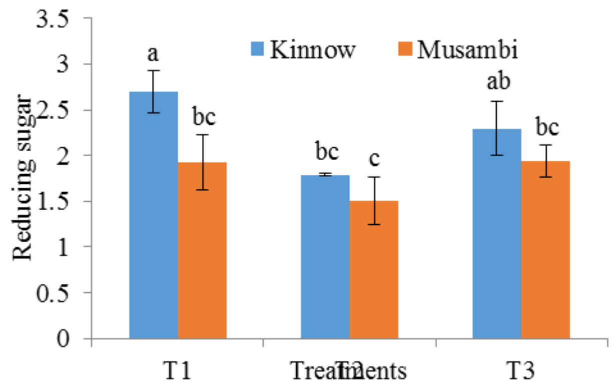

Fig. 13 Effect of plant spacing on reducing sugar in Kinnow and Musambi

Non-reducing sugars

Among all treatments, close spaced plants of T_2 (11 × 11ft), gave the maximum i.e. 7.21% non-reducing sugar (Fig. 14). While minimum non-reducing sugar (4.91%) was measured in T_1 (11 × 12ft). Whereas the variety V_2 (Musambi) gave more non-reducing sugar than V_1 i.e. maximum (6.59%) non-reducing sugar in V_2 was observed as compared to 5.74% in V_2 (Kinnow). But non-significant results were shown by interaction effect. It was concluded that denser plants had considerable impact on non-reducing sugar. Moreover, our results were in accordance with the outcomes of Zekri (2000) who also concluded that dense plant spacing resulted in more non-reducing sugar. Nitrogen source (inorganic and organic) and its utilization might be the reason that affects the quality of high density plants' fruits. Similarly, Nasir et al. (2006) also stated that reduction in plant spacing resulted in the increase of non-reducing sugar of fruits.

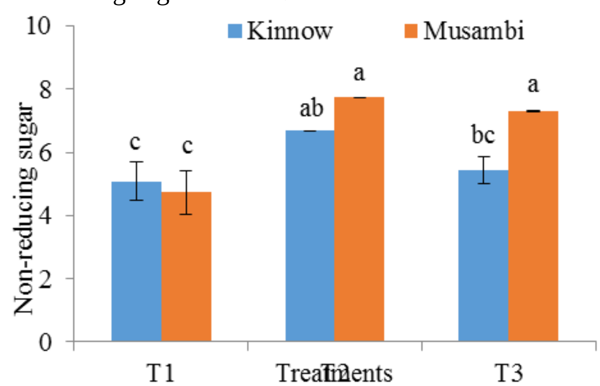

Fig. 14 Effect of plant spacing on non-reducing sugar in Kinnow and Musambi

Conclusion

Importance of high density plantation is increasing due to decrease in agriculture land. By adopting this technique of plantation, fruit bearing volume per hectare increased. In this research, satisfactory results were obtained regarding reproductive growth at 11 × 22 ft. distance plantation. On the other side, good yield was resulted from 11 × 11 ft. distance plantation but with small size fruit. However, more research is required for final recommendation of best planting distance.

Authors Contribution: W.A.D. principal author of the article designed the research study. T.A. helped in write-up. M.U. gathered the data pertaining to different parameters. S.N. analyzed the statistical data. M.A.B. checked the raw data and analyzed the final data. A.N.S. reviewed and edited the manuscript. Z.A. formatted the manuscript according to the guidelines of JPAA.

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