



Optimization and Determination of Doses of Phosphorous and Potassium for *Citrus reticulata* (Blanco) under the Agro-climatic Conditions of Sargodha, Pakistan: Effect on Yield and Fruit Quality of Citrus

Amina^{1,2}, Hira Tariq³, Muhammad Babar Shahzad Afzal^{3*}, Tehseen Ashraf¹ and Shaukat Nawaz³

¹University College of Agriculture, University of Sargodha, Sargodha, Pakistan

²Horticultural Research Institute, Faisalabad, Pakistan

³Citrus Research Institute, Sargodha, Pakistan

*Corresponding Author: Muhammad Babar Shahzad Afzal, Citrus Research Institute, Sargodha, Pakistan.

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Abstract

Kinnow mandarin (*Citrus reticulata* Blanco.) is considered as one of the major fruit crops of Pakistan in terms of area and production. It has supreme taste, quality and health significance. High yield and better quality in citrus fruit are highly dependent upon optimum doses of fertilizer application in the soil. Keeping in view the importance of fertilizers for good quality citrus with maximum yields, this research was conducted to determine the effect of different combinations of phosphorous and potassium fertilizers upon reproductive and chemical characteristics of kinnow mandarin. The experiment was done according to Randomized Complete Block Design (RCBD), with seven treatments and three replications. Soil and plant samples were collected and analyzed for P and K using standard analytical methods. The maximum yield was obtained in T₅ (474.33 fruits/tree) and T₆ (466.33 fruits/tree). In leaf samples, the percentage of phosphorus was maximum for T₁ and T₃, while maximum percentage of potassium was observed in the leaf samples of treatments T₄, T₆ and T₇. None of the treatment affected TSS and TSS acid ratio in Kinnow. The significantly highest value for ascorbic acid was observed for T₅, T₆ and T₇. Maximum phenolic contents were recorded with T₁. It was concluded that both nutrient sources improved the growth and productivity contributing factors. Fertility status of soil and plant was also improved with the application of both sources of nutrients.

Keywords: Citrus; Kinnow; Fertilizer; Yield; Vitamin C

Introduction

Kinnow (*Citrus reticulata* Blanco; *Rutaceae*) is an important fruit among its representatives [1]. It is the heaviest bearer than any other citrus fruits. It is very much liked by all sorts of people due to its excellent taste, dietary values, rich in vitamin-C, and high juice concentration. It is the major fruit crop of Pakistan in terms of area, production and export. It provides 95% share of annual citrus exports from Pakistan [2]. These fruits are commercially important contributing \$6 - 8 billion (US) annually to the world economy and providing jobs to millions of people around the world in harvesting, handling, transportation, storage and marketing [3]. Pakistan is at 12th number with production of 2.36 MT citrus on an area of 206, 569 ha [4]. About 94% of citrus production area is in Punjab, 2.3% in Sindh, 2.4% in the KPK, and 1.3% in Baluchistan. Punjab's share is the biggest due to its climate. Sargodha, Sahiwal and Toba Tek Singh are major citrus producing districts in Punjab. Currently, Sargodha is the main citrus producing district, with about 23% of Pakistan's total citrus plantings [5]. Though, the demand of Pakistan's Kinnow fruit is enormous, but our exporting potential is merely 8% due to a big chunk going waste on account of poor soil, crop and fertilizer management practices, poor quality (fruit with nutrient deficiency and disease symptoms) and poor management during harvesting, transportation, packaging and storage [6,7].

Plant nutrient management can influence flowering, fruit set, fruit size, the amount of vegetative growth and other plant characteristics [8]. By carefully choosing the components of fertilizer pro-

gramme, the grower can nudge a crop toward earlier and heavier fruit set [9-11]. The balanced nutrients have been paid little attention in agricultural areas of the developing world. Nutrients are vital for plant growth and development. They play a significant part in cell growth, cell enlargement and in plant transport nutrients from one part to another. Plant hormones and enzymes stimulate plant growth, flowering, and fruit setting, but those hormones were activated in the presence of nutrients. Citrus needs many nutrients to grow with non-visible sign of deficiency.

The macro-nutrients particularly nitrogen (N), phosphorus (P) and potassium (K) are needed by the citrus plant in greater quantities than others. They play an important role in yield, as well as fruit quality [12-17]. Phosphorus performs many vital functions in the plant photosynthesis, enzyme activity, metabolism and movement of sugars [18]. It is also important for growth and development of flowers, as well as fruits. Low P contents in leaves have been reported to produce deformed poor quality kinnow mandarin fruit with open centers, coarse and thickened peel, low and acidic juice contents [19]. Citrus fruit tree takes up higher amount of potassium (K) as compared to other macronutrients [20,21] because it has a key role in many physiological processes like water relations, opening and closing of stomata, cell division, formation of sugars and starch, neutralization of organic acids, synthesis of proteins, and activation of enzymes [22,23]. Potassium improves citrus fruit quality by enhancing fruit size, juice contents, colour, size and juice flavor [20,24]. Limited supply of K may affect the yield and quality of citrus fruit and accelerate the fruit drop. K

plays a regulatory role in physiological and bio-chemical processes of citrus plant [25]. It is involved in the formation and functioning of proteins, fats, carbohydrates, chlorophyll and maintaining the balance of salts and water in plant cells. The K application has also been reported to play an important role in the acid metabolism of the citrus juice [26].

The growth of young trees is more rapid as compared to fruiting tree, so supply of macronutrients is very important to fulfill the requirement of these rapid growing trees and development. But mostly research has been conducted around the world on mature citrus trees hence there is a dire need to check the results of macronutrients (P and K) on young citrus trees regarding their growth, fruit quality and yield [27]. This study was conducted to determine the fertilizer application program for young Kinnow mandarin plants. The major objectives of this study are to optimize doses of P and K for young citrus trees (4 - 5 years old), to determine the response of different levels of P and K applications on yield and yield contributing factors, and to evaluate the effect of different doses of P and K on fruit quality.

Materials and Methods

Experimental area

This experiment was conducted at the Experimental Research Area of Citrus Research Institute, Sargodha (Latitude: 32°5'1.47" N; Longitude: 72°40'18.69" E), Pakistan with the collaboration of University College of Agriculture Sargodha, Sargodha, Pakistan in the year 2011 - 2013, to study the effect of varying levels of phosphorus and potassium fertilizers on the growth of young Kinnow (*Citrus reticulata*) plants. The study was carried out on five-years-old Kinnow Mandarin plants grafted on rough lemon rootstock.

Experimental design and treatment plan

The experiment was conducted by using Randomized Complete Block Design (RCBD), with seven treatments and three replications. The experiment was done on 21 Kinnow plants. The treatments were applied on Kinnow plants as follows:

$$T_1 = 400\text{g N} + 200\text{g P}_2\text{O}_5 + 200\text{g K}_2\text{O}$$

$$T_2 = 400\text{g N} + 150\text{g P}_2\text{O}_5 + 200\text{g K}_2\text{O}$$

$$T_3 = 400\text{g N} + 250\text{g P}_2\text{O}_5 + 200\text{g K}_2\text{O}$$

$$T_4 = 400\text{g N} + 300\text{g P}_2\text{O}_5 + 200\text{g K}_2\text{O}$$

$$T_5 = 400\text{g N} + 200\text{g P}_2\text{O}_5 + 150\text{g K}_2\text{O}$$

$$T_6 = 400\text{g N} + 200\text{g P}_2\text{O}_5 + 250\text{g K}_2\text{O}$$

$$T_7 = 400\text{g N} + 200\text{g P}_2\text{O}_5 + 300\text{g K}_2\text{O}$$

Cultural practices and application of fertilizer doses

During the course of experiment, cultural practices such as weeding, hoeing, irrigation, fertilization and insect pest management were done where and when needed. The trees were applied with the recommended doses of nitrogen fertilizer into three split doses to maintain the plant vigor and the growth. The soil application of fertilizers was done in February with dose of nitrogen and rests of doses were applied in April and September.

Nutritional status

To determine the status of N, P and K in the leaves of each experimental tree, leaf sampling was done by selecting leaves at random from all sides of each tree; One-hundred mature leaves from branches at shoulder height were collected of 4 - 6 months old, free from disease and insect pest attack. Leaf samples were washed with detergent and rinsed with distilled water for 2 - 3 times. The sampled leaves were dried in shade and then oven dried in perforated paper bags at 65°C for 48 hours. Completely dried leaves were meshed to fine powder and stored in airtight plastic bottles for analysis. Total nitrogen was determined according to the procedure defined by Chapman and Parker [28]; whereas P and K analysis were carried out according to the method described by Yoshida, *et al.* [29].

Soil sampling

Sampling time and procedure

Samples were collected in January prior to fertilizer application and treated samples were collected in September. Each soil sample from each replicated plant of treatment was collected from the drip line of plant or within the area where the maximum root system activity occurs. The depth of soil sample was 0 - 6 inches and 6 - 12 inches. Sampled soil of each replicated plant of treatment of same depth were homogenized or mixed into a non-metal bucket to form a composite sample. Confined samples were taken from composite sample and kept into a labeled paper bag. Soil samples were brought to the laboratory for analysis of the nutrient status of soil.

Soil analysis

Soil samples were collected, air dried and passed through 2 mm sieve. Laboratory analysis was carried out by following the analytical methods of U.S Salinity Laboratory Staff [30]. All calculations were made on oven dried soil weight basis.

Preparation of saturated soil paste

Saturated soil paste was prepared according to method 2.

Determination of phosphorous percentage in the soil

Available phosphorous from soil sample was determined by Olsen's method [31]. Weighed sample of 2.5g was air dried, grinded and 50 ml of extracting solution (sodium bicarbonate) was added. This mixture was shaken for 30 minutes and filtered with Whatman's No. 42 filter paper. About 5 ml aliquot was pipetted out and 5 ml of colour developing reagent was added in 25 ml volumetric flask. This solution was again shaken to remove gas bubbles. The solution was left for 15 minutes. The volume was made up to mark and bluish colour developed. Concentration of phosphorus in soil is directly proportional to the intensity of blue colour developed. Reading was taken on concentration mode at 880 nm wave length on Spectrophotometer.

$$\text{Extractable phosphorus in Soil (ppm)} = \text{ppm p}^* \times \frac{A \times 5}{\text{Wt} \times V}$$

A = Total volume of extractant (ml)

Wt = Weight of Air-dry soil (g)

V = Volume of extract used (ml)

Determination of potassium percentage in the soil

The 2.5g soil sample was weighed, air dried and grinded. It was passed through 2 mm sieve; 50 ml extracted reagent was added and placed on a flatbed reciprocating shaker for 30 minutes, then the extract was filtered. The available K was determined by flame photometer in ppm using graph reading.

Available K (ppm) = reading (ppm) × 20

Here 20 is dilution factor.

Leaf analysis

Collection of leaves and sample preparation

The 4 - 6 months old leaves, free from disease and insect pest infestation were collected from non-fruiting shoot. About 100 leaves were collected from all sides of the tree at shoulder height. Leaves were brought to the laboratory in labeled bags for further analysis. These leaves were washed with detergents and rinsed with distilled water 2 - 3 times. The washed leaves were placed in paper bags and dried in an oven at 65°C for 48 hours. The dried samples were grinded to a fine powder form.

Determination of nutrient elements in leaf

Wet digestion for phosphorus and potassium

The digestion for estimation of phosphorus and potassium was done according to the method described by Yoshida, *et al.* [29]. The beakers were washed with commercial detergents carefully, dipped in HNO₃ for 2 - 3 hours and then rinsed with distilled water 2 - 3 times. These beakers were placed in oven for drying. One-gram leaf sample was transferred to 10 ml of Tri-acid mixture (HNO₃, HClO₄, H₂SO₄ in the ratio 5:2:1) and allowed to stand still for about four hours till all the initial reaction subsided. It was heated gently until the solid material disappeared, then heated vigorously till a clear colorless solution formed.

Phosphorus estimation

Phosphorus was determined according to the method described by Chapman and Parker [28]. Colour of sample was developed by adding 5 ml of diluted sulphuric acid (H₂SO₄) (1:6), 5 ml of 5% Ammonium molybdate [(NH₄)₂MoO₄] and 0.25% Ammonium vanadate (NH₄VO₃). The standard curve was prepared by using different concentrations of potassium dihydrogen phosphate. The coloured samples were placed inside spectrophotometer at a wave length of 420 nm and transmittance was noted which compared with the standard curve to find out the quantity of the element in ppm. It was then converted into percentage by using the following formula:

$$P (\%) = \frac{\text{ppm on graph} \times \text{dilution}}{10^6} \times 100$$

Potassium estimation

Potassium was determined by flame photometer according to the method described by Chapman and Parker [28] Quantity of the element was estimated in ppm by comparing the emission of flame photometer with that of standard curve, which was converted into percentage by using the following formula:

$$K (\%) = \frac{\text{ppm on graph} \times \text{dilution}}{10^6} \times 100$$

Reproductive parameters

Number of flowers per flush

For calculating the number of flowers per flushes, five flushes were tagged from per square meter covering four sides of tree and their mean values were calculated.

Yield

Total number of fruits harvested per treatment and multiplied with the average mean values of weight.

Chemical analysis

Acidity %

Acidity of juice was determined by method given by Hortwitz [32]. 5 ml of juice was taken in 100 ml conical flask and diluted up to 50 ml with distilled water. It was titrated against 0.1 N NaOH using 2 - 3 drops of phenolphthalein as an indicator till pink colour end point was achieved. To determine acidity calculations were made according to the formula:

$$\text{Acidity} (\%) = \frac{0.1N \text{ NaOH used} \times 0.0064}{\text{ml of Juice taken for titration}} \times 100$$

Total soluble solids (%)

Total soluble solids (TSS) of juice were estimated with Automatic Digital Refractometer (Atago Pal-13810). Filtered juice was placed on clean prism and results were expressed as Brix (°Bx).

TSS/Acid ratio

TSS/acid ratio was calculated by dividing the TSS over acidity.

Determination of Vitamin- C (Ascorbic acid)

For the determination of vitamin-C (ascorbic acid) in juice, the method described by Ruck [33] was used. Ten ml juice was taken into 250 ml conical flask and volume was made up to the mark using 0.4% Oxalic acid solution. Five ml of filtered aliquot was taken in a flask and titrated against 2,6-Dichlorophenolindophenoldye to a light pink colour which persisted for 10 - 15 seconds. Vitamin-C was calculated as:

$$\text{Vitamin-C} = \frac{1 \times R_1 \times V}{R \times W \times V_1} \times 100 \text{ mg ascorbic acid per 100 ml juice}$$

Where;

R₁ = ml dye used in titration of aliquot

R = ml dye used in titration of 1 ml of standard ascorbic acid solution prepared by adding 1 ml of 0.1% ascorbic acid + 1.5 ml of 0.4% oxalic acid.

V₁ = ml of juice used.

V = Volume of aliquot made by addition of 0.4% Oxalic acid.

W = ml of aliquot used for titration.

Total phenolic contents

The phenolic content in the fruit juices was estimated by Folin-Ciocalteu method described by Ribeiro, *et al.* [34]. 0.5 mL of each fruit juice was mixed with 2.5 ml distilled water. In this solution, 0.5 ml of F-C reagent (1:1) was added and incubated for 3 minutes. In each tube, 2 ml of 20% sodium carbonate was added and the tubes were kept in boiling water bath for 1 minute. Tubes were cooled, and the absorbance of reaction mixture was read at 650 nm by spectrophotometer. A standard curve was plotted using different concentrations of Gallic acid (standard, 0 - 1000 µg/ml). Total phenolic contents were estimated as µg Gallic Acid Equivalents (GAE)/ml of fruit juice.

Statistical design and analysis

The data was analyzed by using statistics software (version 8.1) through Randomized Complete Block Design (RCBD) to obtain analysis of variance (ANOVA) [35]. Means of all treatments were compared pair wise by least significant difference test (LSD test) and significance was tested at 5% significance level.

Results and Discussion

The experiment was performed to determine the optimum doses of P and K for young Citrus plants with basic nutrient elements i.e. Nitrogen and Phosphorous. The salient objectives of this experiment were to study the response of P and K in improving the fruit quality and yield parameters. P and K were determined by soil and leave analysis, and chemical characteristics of citrus fruit affected by these fertilizers were also studied.

No. of flowers

Application of P and K fertilizers in different combinations affected the number of flowers produced in *Citrus reticulata*. Average number of flowers produced in citrus plants did not show any significant difference (DF = 6, P = 0.428, F = 1.08) as a result of application of different doses of P and K fertilizer (Table 1).

Treatments	No. of flowers	Yield (Fruit/tree)
T1 (200g P ₂ O ₅ + 200g K ₂ O)	36.89 a	331.67 ab
T2 (150g P ₂ O ₅ + 200g K ₂ O)	22.67 a	309.33 ab
T3 (250g P ₂ O ₅ + 200g K ₂ O)	49.43 a	237.67 b
T4 (300g P ₂ O ₅ + 200g K ₂ O)	20.23 a	342.00 ab
T5 (250g P ₂ O ₅ + 150g K ₂ O)	39.13 a	474.33 a
T6 (200g P ₂ O ₅ + 250g K ₂ O)	28.57 a	466.33 a
T7 (200g P ₂ O ₅ + 300g K ₂ O)	24.92 a	390.33 ab

Table 1: Effect of P and K application on reproductive parameters of citrus reticulata.

Means in a column followed by same letters are non-significant; means not followed by like letters differ at 5% level of significance.

Yield (fruit/tree)

Citrus trees required large quantities of mineral nutrients to attain adequate growth and yield, and the requirements for some of the nutrients vary with soil fertility and type [36]. The fruit yield (no of fruits/tree) ranges from 237.67 - 474.33 fruits/tree by using different combinations of P and K fertilizers. Significantly higher yield was obtained in T5 (250g P₂O₅ + 150g K₂O) and T6 (200g P₂O₅

+ 250g K₂O) as compared to all other treatments. However, there was no significant difference in the yield among other treatment combinations (Df = 6, P = 0.419, F = 0.419) (Table 1). Our results are not in accordance with the findings of Dubey and Yadav [37] who studied the effect of phosphorus and potassium on yield was significant and highest yields were obtained at higher doses. The similar results on fruit yield and quality were observed in Nagpur mandarin and acid lime [38-41] and it was reported that with the application of different levels of phosphorus and potassium, high-est yield of Kinnow per plant was 493.3 fruits.

Status of P percentage in leaf and soil samples after fertilizer application

After application of each treatment, leaf analysis showed significant results. Maximum percentage of phosphorus in leaf samples was recorded for T₁ and T₃ (Df = 6, P = 0.034, F = 3.369) (Table 2). The optimum range of P in citrus leaves is 0.12% - 0.16 % [42]. However, as a result of phosphorus fertilizer application (after each treatment), tree responded very well in terms of increase in leaf phosphorus holding optimum level as compared to before application of P fertilizer. These findings are similar with those of Saleem, *et al.* [43] who reported that fertilizer application improved the leaf phosphorus status of both Kinnow and Feutrell's Early trees. Barakat, *et al.* [44] reported that a direct relationship was found between fertilization levels and leaf phosphorus (%). Highest percentage of 0.28% phosphorus was observed with the application of minerals fertilizer in Navel orange. However, in the soil samples after application of fertilizers, none of the treatment showed significant response and their behaviour was statistically similar (DF = 6, P = 0.693, F = 0.6459) (Table 3). These findings are not in accordance with the findings of Ebrahimian and Soleymani [45], and Smith, *et al.* [46] who concluded that soil P concentrations showed significant differences.

Treatments	P% in leaf sample	P% in soil sample	K% in leaf sample	K% in soil sample
T1	0.29 a	4.09 bc	0.96 f	194.33 a
T2	0.11 b	3.37 c	1.26 cd	156.00 bc
T3	0.19 ab	5.07 bc	1.08 e	152.00 c
T4	0.17 ab	5.95 abc	1.31 c	164.33 b
T5	0.15 b	6.33ab	1.23 d	139.67 d
T6	0.13 b	8.27 a	1.38 b	152.33 c
T7	0.17 b	5.45 abc	1.44 a	202.00 a

Table 2: Status of P and K (%) in leaf and soil samples before fertilizer application in the experimental area.

Means in a column followed by same letters are non-significant; means not followed by like letters differ at 5% level of significance.

Leaf and Soil potassium status after fertilizer application (%)

Analysis of variance indicated that the level of K% in citrus leaves was highly significant among the treatments after application of different levels of potassium (Df = 6, F = 25.28, P = 0.0000). Maximum percentage of potassium was observed in the leaf samples of treatments T₄, T₆ and T₇. However, these treatments were statistically similar to each other (Table 3). These findings are similar to those of Obreza [47] who determined that in grapefruit and oranges, increase in K fertilizer rates resulted in a concur-

rent increase of K level in the leaf tissues. Our results are also in accordance with the findings of Shirgure and Srivastava [48] who reported that in Nagpur mandarin leaves K% ranges 0.97 - 1.18% as a result of potash application at higher rates. Similarly, Barakat, *et al.* [43] observed that with the application of mineral fertilizers, K% in leaves of Navel oranges was determined to be 1.57% due to increase in fertilizer dose. Basharat, *et al.* [49] concluded that fertilizer application resulted in improved potash contents in both kinnow and Feutrell's Early, although the response to different fertilizer applications did not differ. The results for K percentage in soil after fertilizer application showed significant increase of K in the soil for T₇ when compared with other treatments (DF =6, F= 5.146, P= 0.007) (Table 3). These findings are in parallel with those of Ashraf, *et al.* [50], and Ashraf, *et al.* [20] whose results indicated that the application of K was beneficial and effective in promoting the growth as well as K contents in the soil.

Treatments	P% in leaf sample	P% in soil sample	K% in soil sample	K% in leaf sample
T1(200g P ₂ O ₅ + 200g K ₂ O)	0.40 a	9.23 a	200.67ab	0.92 d
T2(150g P ₂ O ₅ + 200g K ₂ O)	0.14 b	8.70 a	176.67 c	1.31 b
T3 (250g P ₂ O ₅ + 200g K ₂ O)	0.33 a	9.89 a	166.67 c	1.14 c
T4 (300g P ₂ O ₅ + 200g K ₂ O)	0.20 b	8.07 a	181.00 c	1.46 a
T5 (250g P ₂ O ₅ + 150g K ₂ O)	0.15 b	9.13 a	168.67 c	1.23bc
T6 (200g P ₂ O ₅ + 250g K ₂ O)	0.18 b	8.50 a	182.67bc	1.48 a
T7 (200g P ₂ O ₅ + 300g K ₂ O)	0.20 b	7.97 a	218.00 a	1.52 a

Table 3: P and K (%) in leaf and soil samples after fertilizer application.

Means in a column followed by same letters are non-significant; means not followed by like letters differ at 5% level of significance.

Total soluble solids (TSS)

Analysis of variance showed non-significant results of TSS. None of the treatments showed significant response in TSS after application of fertilizers (Df = 6, F = 1.57, P = 0.2385). The findings were found in close consonance with Shirgure and Srivastava [48] who studied that in Nagpur mandarin with the application of phosphorus and potassium fertilizers, maximum TSS (10.49%) was observed. Our results also confirmed the findings of Salik, *et al.* [41] who also reported that application of phosphorus and potassium in different doses has no significant effect on kinnow juice TSS. However, our results are not in line with the results of Ashkevari, *et al.* [51] in Thomson novel and Dudi, *et al.* [52] in kinnow, who found that application of 320g P₂O₅ and 210g K₂O significantly improved the TSS as compared to control. These findings contradict the results of Mostafa, *et al.* [53] who found that TSS content in the fruit juice increased with potassium in Balady mandarin trees. Dubey and Yadav [37] reported that TSS differed significantly with application of K. The highest TSS (11.77%) was recorded with higher doses of potassium application.

TSS/acid ratio

The results of TSS/acid ratio revealed non-significant differences among the treatments (Df = 6, F = 2.35, P = 0.097). However, the higher TSS/acid ratio value was observed in the fruits of plants treated with T₇ and T₆. Though, TSS/acid ratio remained statistically similar in both treatments. As fruits mature and ripe, the carbohydrates are converted into sugars, increasing the sugar content with the decrease in acidity, resulting in a higher sugar: acid ratio. As TSS/acid ratio is a flavoring factor, these results depicted that with increase in the ratio, there was a decrease in the acidity so with low TSS/acid ratio, quality of fruit is poor, and it becomes tasteless. The ratio is used to determine the fruit maturity standards, where the ratio is high, fruit will mature earlier. The higher the Brix: acid ratio, the earlier is the fruit maturity [54]. Our results confirmed the findings of Dubey and Yadav [37] who reported that TSS/acid ratio was not affected significantly by the application of potassium. Our results also confirmed the findings of Rahman, *et al.* [55] who recorded the highest values for TSS/acid ratio with application of potassium. In this context, our results are not consistent with the findings of Ashkevari, *et al.* [51] and Wang, *et al.* [56] who determined that the ratio of soluble solids to acid in the fruit was significantly affected with the application of phosphorus and potassium.

Vitamin-C (mg/100ml)

The effect of phosphorus and potassium on the ascorbic acid contents of Kinnow mandarin juice revealed significant results (DF = 6, F = 13.65, P = 0.0001). The significantly highest value for ascorbic acid was observed for T₅, T₆ and T₇ (Table 4). Vitamin-C is a powerful antioxidant and is an important part of human food. It helps to save the human from many serious diseases and scavenges the reactive oxygen species (ROS) produced in the body. Vitamin-C contents in fruits vary in concentration for various citrus species. Vitamin-C is being affected by the environmental factors, level of nutrition, time of fruit harvest and plant age. K is very effective in improving the ascorbic acid contents in kinnow fruit [57]. In this context, the results are consistent with other previous studies, i.e. Ashraf, *et al.* [21], Ibrahim, *et al.* [58] and Ashraf, *et al.* [20] as they found that K was effective in enhancing the ascorbic acid contents in citrus fruits. Our results also confirmed the findings of Saleem, *et al.* [59], Abd El-Moneim, *et al.* [60] and Wang, *et al.* [56] who found that phosphorus and potassium had a significant effect on vitamin-C contents in citrus fruits. Dudi, *et al.* [52] found that ascorbic acid level improved in kinnow. Maximum value of ascorbic acid ranges from 24.38 to 28.29 mg/100 ml was obtained with the application of different level of P₂O₅ and K₂O.

Total phenolic contents

The highly significant results (DF = 6, F = 857.84, P = 0.0000) were obtained as for as total phenolic contents are concerned (Table 4). Mean values for total phenolic contents ranges from 530 - 710µg GAE/ml in our work. Maximum phenolic contents value 710µg GAE/ml was found in T₁ (200g P₂O₅ + 200g K₂O) which was significant and different from all other treatments. Moreover, T₃ (250g P₂O₅ + 200g K₂O) and T₇ (200g P₂O₅ + 300g K₂O) also showed significantly higher values of total phenolic contents as compared to T₂, T₄, T₅ and T₆ with total phenolic contents of 690µg GAE/ml and 685µg GAE/ml, respectively. Citrus fruits not only have their delicious flavors, but also have their antioxidant capacity with

health benefits [61,62]. Recently more attentions had been paid on phenolic compounds of citrus fruits [63]. The antioxidant property of phenolic is mainly due to their REDOX properties. They act as reducing agents, hydrogen donors, singlet oxygen quenchers and metal chelators. Our results are in accordance to Wu, *et al.* [64] who reported that with the application of phosphorus and potassium there is a significant effect on total phenolic contents. Especially potassium as an individual nutrient has improved the total phenolic contents. Our results confirmed the findings of Radi, *et al.* [65] who reported that in apricot fruit fertilization with the lowest K level resulted in lower concentrations in phenolic compounds. Such impact could be partially due to potassium activating physiological metabolism reactions in plants and playing an important role in growth and water-use efficiency [66].

Treatments	TSS	TSS/acid ratio	Vit-C (mg/100ml)	Total phenolic contents (µg GAE/ml)
T1 (200g P ₂ O ₅ + 200g K ₂ O)	9.80ab	7.87b	21.56bc	710a
T2 (150g P ₂ O ₅ + 200g K ₂ O)	9.70ab	9.34ab	18.30c	590d
T3 (250g P ₂ O ₅ + 200g K ₂ O)	11.37a	9.54ab	26.14b	690b
T4 (300g P ₂ O ₅ + 200g K ₂ O)	8.63b	7.99b	24.18b	530f
T5 (250g P ₂ O ₅ + 150g K ₂ O)	9.17b	8.55b	32.68a	650c
T6 (200g P ₂ O ₅ + 250g K ₂ O)	10.27ab	10.63ab	35.29a	580e
T7 (200g P ₂ O ₅ + 300g K ₂ O)	9.67ab	12.05a	32.33a	685b

Table 4: Effect of P and K fertilizer application on chemical characteristics of citrus fruit.

Means in a column followed by like letters are non-significant; means not followed by like letters differ at 5% level of significance.

Conclusion

This study unravel the effect of different combinations and doses of phosphorous and nitrogen fertilizers upon different parameters of Kinnow Mandarin in the citrus orchards of Sargodha. The tested parameters were number of flowers per flush, yield, acidity, TSS, TSS/ acid ratio, ascorbic acid and phenolic contents. The treatment combinations 250g P₂O₅ + 150g K₂O and 200g P₂O₅ + 250g K₂O are recommended to get higher yields per plant. However, the parameters like number of flowers, TSS and TSS/acid ratio were not affected by any treatment combinations. Maximum phenolic contents were achieved by using dose of 200g P₂O₅ + 200g K₂O per plant.

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