Influence of Fertilizers and Plant Growth Regulators Application on Physicochemical Attributes of ‘Kinnow’ Mandarin Fruit

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**ABSTRACT**

Plant growth regulators (Kinetin, GA\textsubscript{3} and Wokozim) and fertilizers (nitrogen, phosphorus, potassium and calcium) alone and in various combinations were applied to young ‘Kinnow’ mandarin trees to improve their quality. Fruit harvested at commercial maturity was analyzed at harvest and seven days after storage at ambient conditions. Results revealed that higher seed number (20) was recorded with Kinetin at 60 mg l\textsuperscript{-1} and minimum seed number (10) with Kinetin at 30 mg l\textsuperscript{-1}+ GA\textsubscript{3} at 10 mg l\textsuperscript{-1}. More healthy and less aborted seeds were recorded with Wokozim (1500 g tree\textsuperscript{-1})+2 ml l\textsuperscript{-1} foliar spray. High juice mass (%) and low rag mass (%) were observed with control and 250 g tree\textsuperscript{-1} phosphorous (P) application as single super phosphate. The TSS were higher with 150 g tree\textsuperscript{-1} calcium (Ca) applied as calcium ammonium nitrate and lower with P application at 500 g tree\textsuperscript{-1}. Titratable acidity was higher in fruit treated with Kinetin at 60 mg l\textsuperscript{-1} and lower in fruit treated with NPK (230 + 250 + 250 g tree\textsuperscript{-1})+Ca (150 g tree\textsuperscript{-1})+Kinetin at 30 mg l\textsuperscript{-1}. The TSS:TA ratio was higher in fruit treated with NPK (230 + 250 + 250 g tree\textsuperscript{-1})+ Ca (150 g tree\textsuperscript{-1})+ Kinetin at 30 mg l\textsuperscript{-1} and lower in fruit treated with P (250 g tree\textsuperscript{-1})+ Kinetin at 30 mg l\textsuperscript{-1}. Ascorbic acid was higher in fruit treated with Ca (150 g tree\textsuperscript{-1})+P (250 g tree\textsuperscript{-1})+Kinetin at 30 mg l\textsuperscript{-1} and lower with Wokozim (1500 g tree\textsuperscript{-1})+2 ml l\textsuperscript{-1} foliar applications.

**KEYWORDS**

Fruit quality; fertilizers; ‘Kinnow’ mandarin; plant growth regulators; tree age

**Introduction**

‘Kinnow’ mandarin leads the citrus industry of Pakistan because of its excellent yield, more juice contents, refreshing flavor and adaptation to the indigenous agro climatic circumstances of Punjab, Pakistan since its introduction to sub continent from USA. In Pakistan during the year 2015–16 citrus fruit production was 2.34 million tonnes from an area of 192.23 thousand hectares (Anonymous, 2016). Pakistan exported 0.37 million tonnes of citrus and earned foreign exchange of 222 USD million during the year 2017–18 (Abbas, 2018). Presently, the area under ‘Kinnow’ mandarin cultivation is rising; but, owing to decline-related problems productive lifespan of citrus tree is very short and thus the growers frequently have to replace their old and vulnerable trees with new ones. On the other hand, fruit from young trees are considered inferior in fruit quality due to less juice and total soluble solid contents and thicker and rough rind (Khalid et al., 2012) due to which exporters are reluctant to purchase fruit from juvenile orchards.

The role of plant growth regulators (PGRs) and nutrients in citrus fruit quality is well recognized in mature trees (Ashraf et al., 2010; Fidelibus et al., 2002; Pozo et al., 2000; Singh and Sharma, 2011). The...
PGRs has been used to influence flowering, fruit set and fruit drop (Berhow, 2000) and have also been used to influence rind and juice quality, fruit color and size and to improve total soluble solid in different citrus species. Helal et al. (2019) reported that GA3 application at 50 ppm improved juice volume in Valencia oranges. In citrus plants endogenous PGRs and nutrient levels vary with vegetative and reproductive growth, which influences fruit set and fruit quality. Previous studies revealed that juvenile plants of Kalanchoe and Valencia orange were low in endogenous GA3 and cytokinins (Hendry et al., 1982; Wadhi and Ram, 1967) respectively as related to adult plants. Plant growth regulators (PGR) and nutrients were proved to affect the fruit rind thickness. Erner et al. (1976) described that in mature 18 year old Shamouti orange trees, rough fruit had higher endogenous cytokinin and GA3 contents as compared to smooth fruit. Goldschmidt (1983) reported that confined application of lanolin pastes comprising 0.02–1.0% of gibberellin A4+7 to developing citrus fruitlets produced peel thickening. However, Pozo et al. (2000) stated that foliar spray of GA3 on citrus fruit reduced rind thickness.

Macro (N, P, K and Ca) nutrients are required in large amounts and micro (B, Cu, Fe, Mg, Mn, Mo, Ni and Zn) nutrients are required in small amounts to play an important role in citrus fruit quality (Kaur et al., 2015). Phosphorous (P) being a constituent of nucleoprotein is important in cell division and plays important role in many physiological processes and enzymatic reactions in plants (Azeem et al. 2018). Similarly potassium also has a major role in many physiological processes in citrus plants like water relations, opening and closing of stomata, activation of enzymes, cell division, synthesis of proteins, formation of sugars and starch, and acid metabolism of citrus juice (Liu et al., 2000; Srivastava and Singh, 2005). Potassium has a positive influence on various citrus fruit quality attributes like fruit size, juice contents, color, size and juice flavor (Ashraf et al., 2010; Tiwari, 2005). Amina et al. (2018) reported that NPK applications significantly improve ascorbic acid contents of ‘Kinnow’ mandarin fruit. Calcium in the form of calcium pectate enhances the mechanical power of cell wall and plays an important role in the formation of pedicel attachment to proximal of fruit, hence resulted in a reduced fruit drop (Guardiola and Garcia, 2000). Zaman et al. (2019) reported that calcium application improves ‘Kinnow’ mandarin fruit yield and quality. Ascorbic acid contents increased with application of zinc and its combination with manganese and boron (Tariq et al. 2007) and with zinc and GA3 applications (Eman et al., 2007) in Washington navel orange and sweet orange respectively. Khalid et al. (2012) disclosed that young ‘Kinnow’ mandarin trees which were lower in endogenous Ca, N and P had rough and thick rind and had poor biochemical (TSS, titratable acidity, TSS: titratable acidity ratio and total sugars) fruit quality. Therefore the variation in endogenous levels of PGRs and nutrients among juvenile and adult plants could be a potential cause of inferior quality fruit in young orchards.

However, sporadic work has been done on exogenous use of PGRs and nutrients on citrus fruit quality grown on young plants. This study was carried out to determine the potential of exogenous use of PGRs (Kinetin and gibberellic acid), macronutrients (N, P, K and Ca) along with Wokozim (a commercial product which comprised of auxins, cytokinin, and nutrients such as B, Cu, Fe, Mg, Mn, Mo, Ni and Zn) in modifying fruit quality characteristics of ‘Kinnow’ mandarin fruits procured from young trees.

**Materials and Methods**

**Plant Material and Site Selection**

The trial was conducted on juvenile (5–6 years old) ‘Kinnow’ mandarin (Citrus nobilis Lour × Citrus deliciosa Tenora) trees budded on to rough lemon (Citrus jambhiri) rootstock, at Silanwali tehsil of Sargodha district (32°03’ N; 72°40’ E), Punjab, Pakistan.
**Experimental Treatments**

Fertilizers were applied to the plants in the first week of March, whereas PGRs were sprayed in the first week of April during fruit setting stage. Fifteen treatments comprising fertilizers and PGRs applied in this experiment viz. Control (without fertilizer and growth regulator), P (250 g tree⁻¹), P (500 g tree⁻¹), NPK (230 + 250 + 250 g tree⁻¹)+Ca (150 g tree⁻¹), Wokozim (granular) (1500 g tree⁻¹), Wokozim (1500 g tree⁻¹) + 2 ml l⁻¹ foliar spray, Kinetin (30 mg l⁻¹), Kinetin (60 mg l⁻¹), Kinetin (120 mg l⁻¹), P (250 g tree⁻¹) + Kinetin (30 mg l⁻¹), Kinetin (30 mg l⁻¹)+GA₃ (10 mg l⁻¹), P (250 g tree⁻¹)+Kinetin (30 mg l⁻¹)+GA₃ (10 mg l⁻¹), NPK (230 + 250 + 250 g tree⁻¹)+Ca (150 g tree⁻¹)+Kinetin (30 mg l⁻¹), Ca (150 g tree⁻¹)+ P (250 g tree⁻¹)+Kinetin (30 mg l⁻¹), Ca (150 g tree⁻¹).

**Preparation and Application of Treatments**

The source of N was urea (46% N), P was single supper phosphate (20% P), K was sulfate of potash (50% K) and calcium was calcium ammonium nitrate (10% Ca). Aqueous solution of PGRs having 0.1% wetting agent (Tween 20) was sprayed by hand held sprayer on entire tree to the point of run off. Simple water containing the same concentration of Tween 20 was used to treat control plants. Randomized complete block design (RCBD) was used to lay down the experiment having three replications with single tree per replication. Twenty fruits per treatment per replication were randomly picked at marketable harvest maturity (100% orange color) and transported to the Postharvest Research and Training Center (PRTC), Institute of Horticultural Sciences (IHS) University of Agriculture, Faisalabad (UAf), Pakistan. Fruits were washed with tap water, ten fruits per treatment per replication were analyzed for various fruit quality attributes just after harvest and ten fruits were retained at ambient environment (20 ± 2°C) for seven days and analyzed for physicochemical attributes.

**Physical Parameters**

Physical fruit quality parameters like fruit mass (g), fruit mass loss (%), juice mass (%), rag mass (%), rind mass (%), seed mass (%), fruit diameter (mm), rind thickness (mm), seed number, seed mass (%), aborted and healthy seed (%) were determined as described by Khalid (2013) with some modifications. Mass of ten fruit was determined and their average was calculated to determine fruit mass (g). Fruit mass loss (%) was calculated by subtracting the final mass from initial mass and their percentage was calculated. Rind, rag and seed mass (%) were determined by dividing the individual component mass by fruit mass multiplied by 100. Juice mass (%) was calculated by subtracting the sum of rind, rag and seed mass from fruit mass and expressed as a percentage. Total number of seeds from ten fruits was counted and their average was determined. Healthy and aborted seed from total seeds were separated and their percentage was determined.

**Chemical Composition**

Titratable acidity (TA) (%), TSS (°Brix), TSS:TA ratio and ascorbic acid (AA) (mg100 ml⁻¹) were evaluated as described by Khalid et al. (2012) with some modifications. Titratable acidity (%) was calculated by titrating juice sample against 0.1 N NaOH using two to three drops of phenolphthalein as an indicator (Hortwitz, 1960). Total soluble solids of juice sample were determined with hand refractometer (Atago, ATC-1, Tokyo, Japan). TSS:TA ratio was calculated by dividing TSS with TA. Ascorbic acid (AA) (mg100 ml⁻¹) was determined by titrating five ml of juice aliquot with 2, 6-dichlorophenolindophenol dye solution (Rusk, 1961).
**Statistical Analysis**

Treatments were organized in randomized complete block design (RCBD) with two factors (treatments and storage duration) factorial arrangement and treatment means were separated by Duncan’s Multiple Range test (DMRT). Further, different treatment combinations were compared using orthogonal contrasts.

**Results and Discussion**

**Physical Parameters**

Seed quality data exhibited in Table 1 showed that higher number of seeds per fruit (20) were extracted from plants treated with Kinetin 60 mg l⁻¹, while the lesser seed number per fruit (10) was found when both Kinetin and GA₃ were applied at 30 mg l⁻¹ + GA₃ at 10 mg l⁻¹. Reduction in seed number by GA₃ application might be due to reduced fertilization by either increasing ovule abortion or decreasing pollen tube germination (Mesejo et al., 2008). Similarly a reduction in seed number by GA₃ application was reported in Murcott Tangor (Domínguez and Rodríguez, 2007) and Afourer mandarin (Gambetta et al., 2013). More seed number in Kinetin treated fruit might be due to the positive impact of Kinetin on source sink relation in reproductive development by prompting photo assimilates production and partitioning (Kriedemann, 1968). Increased in seed number by Kinetin application was also reported in lentil by Khalil et al. (2006). More healthy (96%) and less aborted seeds (4%) were obtained from Wokozim (1500 g tree⁻¹)+2 ml l⁻¹ foliar spray, whereas P (250 g tree⁻¹) + Kinetin at 30 mg l⁻¹ gave lesser healthy (89%) and higher aborted seeds (11%) per fruit. Higher aborted seed in P treated plants might be due to reduced uptake of Boron as both nutrients are antagonistic to each other (Kaya et al., 2009). Boron is necessary for reproductive growth and

| Treatments | Seed mass (g) | Total seed | Healthy seed (%) | Aborted seed (%) |
|------------|---------------|------------|------------------|-----------------|
| Control    | 1.8           | 14 cd      | 94abcd           | 6bcde           |
| P (250 g tree⁻¹) | 2.0           | 14 cd      | 91cde            | 9abc            |
| P (500 g tree⁻¹) | 2.1           | 15 cd      | 90de             | 10ab            |
| NPK (230 + 250 + 250 g tree⁻¹ + Ca (150 g tree⁻¹)) | 2.6           | 19a        | 90e              | 10a             |
| Wokozim (granular) (1500 g tree⁻¹) | 2.1           | 18abc     | 92abcde          | 8abbcde         |
| Wokozim (1500 g tree⁻¹) + 2 ml l⁻¹ foliar spray | 2.1           | 13de      | 96a              | 4e              |
| Kinetin at 30 mg l⁻¹ | 2.6           | 12de      | 93abcde          | 7abcde          |
| Kinetin at 60 mg l⁻¹ | 3.0           | 20a        | 94abcd           | 6bcde           |
| Kinetin 120 mg l⁻¹ | 2.2           | 13de      | 95abc            | 5cde            |
| P (250 g tree⁻¹) + Kinetin at 30 mg l⁻¹ | 1.8           | 15bcd     | 89e              | 11a             |
| Kinetin at 30 mg l⁻¹ + GA₃ at 10 mg l⁻¹ | 2.6           | 10e        | 95a              | 5e              |
| P (250 g tree⁻¹) + Kinetin at 30 mg l⁻¹ + GA₃ at 10 mg l⁻¹ | 2.5           | 18ab      | 95ab             | 5de             |
| NPK (230 + 250 + 250 g tree⁻¹ + Ca 150 g tree⁻¹) + Kinetin at 30 mg l⁻¹ | 2.0           | 15cd      | 91bcd            | 9abcd           |
| Ca (150 g tree⁻¹) + P (250 g tree⁻¹) + Kinetin at 30 mg l⁻¹ | 2.5           | 19a        | 94abcd           | 6bcde           |
| Ca (150 g tree⁻¹) | 2.3           | 18ab      | 93abcde          | 7abcde          |
| LSD (P ≤ 0.05) | NS            | 3.49       | 4.07             | 4.07            |

**Significance of selected contrasts on D-1**

| Treatments | Seed mass (g) | Total seed | Healthy seed (%) | Aborted seed (%) |
|------------|---------------|------------|------------------|-----------------|
| Control vs P | NS           | NS         | 0.04*            | 0.04*           |
| Control vs Kinetin | 0.007*       | NS         | NS               | NS              |
| Control vs Wokozim | NS          | NS         | NS               | NS              |
| P vs Kinetin | 0.01*        | NS         | 0.01*            | 0.01*           |
| P vs Wokozim  | NS           | NS         | 0.01*            | 0.01*           |
| Wokozim vs Kinetin | 0.02*       | NS         | NS               | NS              |

NS = Non significant; N = Nitrogen; P = Phosphorous; K = Potassium; Ca = Calcium; PGRs = Plant Growth Regulators

*Similar letter in columns represents statistically similar results.*
fertilization (Rerkasem et al., 1993) and its deficiency may cause seed abortion. The contrast between control and Kinetin, Kinetin and Wokozim and P and Kinetin showed that seed mass (g) was significantly increased with Kinetin treatments (Table 1). Contrasts between control and P, P and Kinetin and P and Wokozim discovered that P applications significantly decreased healthy seed (%) and improved aborted seed (%) as compared to control, Kinetin and Wokozim applications.

Fruit mass (g), rind mass (%), fruit diameter (mm) and rind thickness (mm) remained statistically at par with plant growth regulators and nutrients applications (Tables 2 and 3). Juice mass (%) was significantly reduced (44%) while rag mass (%) was significantly improved (30%) by higher dose of P and Wokozim applications in the form of fertilizer and foliar spray respectively (Table 4). Contrast analysis revealed that rag mass was significantly improved by Wokozim application. This could be due to rise in sink strength as Wokozim is comprised of PGRs and nutrients.

**Chemical Composition**

Table 5 revealed that Ca application at 150 g tree⁻¹ had higher TSS (10.8°Brix), whereas lower TSS (9.2°Brix) was depicted with P at 500 g tree⁻¹ (Table 5). Decline in TSS by P application could be due to reduced activity of sucrose phosphate synthase (SPS) enzyme by the application of P (Qiu and Israel, 1992). This enzyme synthesizes sucrose, which is the key component of TSS of juice (Huang et al., 2009; Huber and Israel, 1982). Therefore decrease in the synthesis of sucrose might decrease the TSS of juice. Analogous outcomes of lower the TSS in Flam grapefruit juice by phosphorus application were also stated by Dou et al. (2005). Raise in the TSS of juice by calcium treatment could be due to its boosting effect on enzymes, which synthesize sucrose (Bhatia and Singh, 2000) and therefore improved TSS of juice. Likewise, Singh and Sharma (2011) also described that in citrus pre-harvest applications of calcium increased juice TSS (*Brix). Interaction of treatments and shelf life period showed that on seventh day maximum TSS (11.8°Brix) was recorded with Ca (150 g tree⁻¹)+P (250 g

| Table 2. Fruit physical quality variables influenced by PGRs and nutrients. |
|---|
| **Treatments** | Fruit mass (g) | Fruit diameter (mm) | Rind thickness (mm) |
| Control | 146.1 | 69.4 | 4.1 |
| P (250 g tree⁻¹) | 149.1 | 69.2 | 4.6 |
| P (500 g tree⁻¹) | 141.7 | 69.0 | 4.4 |
| NPK (230 + 250 + 250 g tree⁻¹) + Ca (150 g tree⁻¹) | 134.7 | 67.2 | 4.1 |
| Wokozim (granular) (1500 g tree⁻¹) | 131.9 | 66.1 | 4.1 |
| Wokozim (1500 g tree⁻¹) + 2 ml l⁻¹ foliar spray | 159.2 | 71.3 | 4.4 |
| Kinetin at 30 mg l⁻¹ | 150.1 | 70.6 | 4.6 |
| Kinetin at 60 mg l⁻¹ | 132.8 | 66.8 | 4.3 |
| Kinetin 120 mg l⁻¹ | 154.1 | 70.9 | 4.7 |
| P (250 g tree⁻¹) + Kinetin at 30 mg l⁻¹ | 142.1 | 68.4 | 4.0 |
| Kinetin at 30 mg l⁻¹ + GA₃ at 10 mg l⁻¹ | 144.1 | 69.5 | 4.2 |
| P (250 g tree⁻¹)+Kinetin at 30 mg l⁻¹ + GA₃ at 10 mg l⁻¹ | 156.8 | 71.3 | 4.5 |
| NPK (230 + 250 + 250 g tree⁻¹)+Ca (150 g tree⁻¹)+Kinetin at 30 mg l⁻¹ | 149.7 | 70.2 | 4.4 |
| Ca (150 g tree⁻¹)+ P (250 g tree⁻¹) + Kinetin at 30 mg l⁻¹ | 159.0 | 70.6 | 5.1 |
| Ca (150 g tree⁻¹) | 141.2 | 67.7 | 4.6 |
| LSD (P ≤ 0.05) | NS | NS | NS |

**Significance of selected contrasts on D-1**

| Fruit (g) | Fruit mass diameter (mm) | Rind thickness (mm) |
|---|---|---|
| Control vs P | NS | NS | NS |
| Control vs Kinetin | NS | NS | NS |
| Control vs Wokozim | NS | NS | NS |
| P vs Kinetin | NS | NS | NS |
| P vs Wokozim | NS | NS | NS |
| Wokozim vs Kinetin | NS | NS | NS |

NS = Non significant; N = Nitrogen; P = Phosphorous; K = Potassium; Ca = Calcium; PGRs = Plant Growth Regulators

*Similar letter in columns represents statistically similar results.*
Table 3. Effect of PGRs and nutrients on rind mass (%) of ‘Kinnow’ mandarin.

| Treatments                                      | Rind mass (%) |       |       |
|------------------------------------------------|---------------|-------|-------|
|                                                 | * Day-1 | ** Day-7 | Mean (Treatment) |
| Control                                         | 27       | 23     | 25    |
| P (250 g tree^{-1})                             | 25       | 22     | 24    |
| P (500 g tree^{-1})                             | 27       | 23     | 25    |
| NPK (230 + 250 + 250 g tree^{-1})+Ca (150 g tree^{-1}) | 26   | 25     | 25    |
| Wokozim (granular) (1500 g tree^{-1})            | 27       | 26     | 26    |
| Wokozim (1500 g tree^{-1}) + 2 ml l^{-1} foliar spray | 27   | 25     | 26    |
| Kinetin at 30 mg l^{-1}                         | 27       | 25     | 26    |
| Kinetin at 60 mg l^{-1}                         | 25       | 23     | 24    |
| Kinetin 120 mg l^{-1}                           | 28       | 24     | 26    |
| P (250 g tree^{-1}) + Kinetin at 30 mg l^{-1}    | 26       | 24     | 25    |
| P (500 g tree^{-1}) + Kinetin at 30 mg l^{-1}    | 27       | 24     | 25    |
| P (250 g tree^{-1}) + Kinetin at 30 mg l^{-1} + GA_{3} at 10 mg l^{-1} | 26   | 26     | 26    |
| NPK (230 + 250 + 250 g tree^{-1})+Ca (150 g tree^{-1})+Kinetin at 30 mg l^{-1} | 27   | 27     | 27    |
| Ca (150 g tree^{-1})+ P (250 g tree^{-1})+Kinetin at 30 mg l^{-1} | 27       | 25     | 26    |
| Ca (150 g tree^{-1})                           | 27       | 25     | 26    |
| Controls vs P                                  | NS       |        |       |
| Controls vs Kinetin                           | NS       |        |       |
| Controls vs Wokozim                            | NS       |        |       |
| P vs Kinetin                                   | NS       |        |       |
| P vs Wokozim                                   | NS       |        |       |
| Wokozim vs Kinetin                             | NS       |        |       |

NS = Non significant; N = Nitrogen; P = Phosphorous; K = Potassium; Ca = Calcium; PGRs = Plant Growth Regulators

Similar letter in columns represents statistically similar results.

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Table 4. Influence of PGRs and nutrients on rag mass (%) and juice mass (%) of ‘Kinnow’ mandarin.

| Treatments                                      | Rag mass (%) |       | Juice mass (%) |       |
|------------------------------------------------|--------------|-------|----------------|-------|
|                                                 | *Day-1 | **Day-7 | Mean | *Day-1 | **Day-7 | Mean |
| Control                                         | 24     | 26     | 25d  | 48    | 51     | 50ab |
| P (250 g tree^{-1})                             | 24     | 28     | 26cd | 50    | 50     | 50a  |
| P (500 g tree^{-1})                             | 28     | 35     | 32a  | 45    | 42     | 44e  |
| NPK (230 + 250 + 250 g tree^{-1})+Ca (150 g tree^{-1}) | 26   | 26     | 26bcd| 47    | 49     | 48abcd|
| Wokozim (granular) (1500 g tree^{-1})            | 28     | 30     | 29abc| 45    | 44     | 44de |
| Wokozim (1500 g tree^{-1}) + 2 ml l^{-1} foliar spray | 29   | 30     | 30ab | 44    | 45     | 44cde|
| Kinetin at 30 mg l^{-1}                         | 26     | 28     | 27bcd| 47    | 47     | 47abced|
| Kinetin at 60 mg l^{-1}                         | 25     | 28     | 27bcd| 50    | 49     | 49abc|
| Kinetin 120 mg l^{-1}                           | 29     | 28     | 29abc| 43    | 47     | 45bced|
| P (250 g tree^{-1}) + Kinetin at 30 mg l^{-1}    | 29     | 26     | 27bcd| 46    | 50     | 48abcd|
| P (250 g tree^{-1}) + Kinetin at 30 mg l^{-1} + GA_{3} at 10 mg l^{-1} | 28   | 25     | 27bcd| 45    | 51     | 48abcd|
| P (250 g tree^{-1}) + Kinetin at 30 mg l^{-1} + GA_{3} at 10 mg l^{-1} | 25   | 26     | 26d  | 48    | 47     | 47abced|
| NPK (230 + 250 + 250 g tree^{-1})+Ca (150 g tree^{-1})+Kinetin at 30 mg l^{-1} | 26   | 26b    | 26cd | 47    | 47     | 47abced|
| Ca (150 g tree^{-1})+ P (250 g tree^{-1})+Kinetin at 30 mg l^{-1} | 26     | 26b    | 26cd | 46    | 49     | 48abcd|
| Controls vs P                                  | NS     |        |       |       |
| Controls vs Kinetin                           | NS     |        |       |       |
| Controls vs Wokozim                            | 0.03*  |        |       |       |
| P vs Kinetin                                   | NS     |        |       |       |
| P vs Wokozim                                   | NS     |        |       |       |
| Wokozim vs Kinetin                             | NS     |        |       |       |

NS = Non significant; N = Nitrogen; P = Phosphorous; K = Potassium; Ca = Calcium; PGRs = Plant Growth Regulators

Similar letter in columns represents statistically similar results.
Table 5. Nutrients and PGRs effect on TSS (Brix) and acidity (%) of 'Kinnow' mandarin juice.

| Treatments | TSS (‘Brix) | Titratable acidity (%) |
|------------|-------------|------------------------|
|            | *Day-1      | **Day-7 | Mean | *Day-1 | **Day-7 | Mean |
| Control    | 9.1ef       | 9.8cdf   | 9.4de | 0.62   | 0.50    | 0.56abcd |
| P (250 g tree⁻¹) | 9.2de       | 9.7cdf   | 9.5de | 0.60   | 0.45    | 0.53d   |
| P (500 g tree⁻¹) | 8.8f       | 9.6cdf   | 9.2e  | 0.59   | 0.52    | 0.55abcd |
| NPK (230 + 250 + 250 g tree⁻¹+ Ca (150 g tree⁻¹) | 9.7cdfef | 9.2def   | 9.5de | 0.63   | 0.45    | 0.54bde |
| Wokozim (granular) (1500 g tree⁻¹) | 9.9 cdef | 9.7cdef  | 9.8cde | 0.74   | 0.49    | 0.62abc |
| Wokozim (1500 g tree⁻¹) + 2 ml l⁻¹ foliar spray | 10.2cd | 9.9cde | 10.1bcd | 0.75 | 0.50 | 0.62ab |
| Kinin at 30 mg l⁻¹ | 9.9cdef | 9.8cde | 9.8cde | 0.60 | 0.48 | 0.57abde |
| Kinetin at 60 mg l⁻¹ | 9.8cdef | 10.3bc | 10.1bcd | 0.72 | 0.56 | 0.64a |
| Kinetin 120 mg l⁻¹ | 9.7cdef | 9.9cde | 9.8cde | 0.64 | 0.54 | 0.59abde |
| P (250 g tree⁻¹) + Kinetin at 30 mg l⁻¹ | 9.4cdef | 9.9cde | 9.7cde | 0.72 | 0.54 | 0.63a |
| Kinetin at 30 mg l⁻¹+ Ga₃ at 10 mg l⁻¹ | 9.1ef | 11.2ab | 10.2abcd | 0.71 | 0.55 | 0.63a |
| P (250 g tree⁻¹)+Kinetin at 30 mg l⁻¹ at 10 mg l⁻¹ | 9.4cdef | 11.2ab | 10.3abc | 0.57 | 0.50 | 0.54cd |
| NPK (230 + 250 + 250 g tree⁻¹)+Ca (150 g tree⁻¹)+Kinetin at 30 mg l⁻¹ | 9.1ef | 11.5a | 10.3abc | 0.60 | 0.49 | 0.52d |
| Ca (150 g tree⁻¹) | 9.5cdef | 11.80a | 10.6ab | 0.67 | 0.51 | 0.59abcd |
| Ca (150 g tree⁻¹) | 10.0cde | 11.50a | 10.8a | 0.67 | 0.53 | 0.60abcd |
| Treatments (LSD) | 0.61 | 0.07 |
| Treat. × SD (LSD) | 0.87 | NS |
| Significance of selected contrasts on D-1 | TSS | Titratable acidity (TA) (%) |
| Control vs P | NS | NS |
| Control vs Kinetin | NS | NS |
| Control vs Wokozim | 0.04* | 0.02* |
| P vs Kinetin | 0.03* | NS |
| P vs Wokozim | 0.01* | 0.001* |
| Wokozin vs Kinetin | NS | NS |

NS = Non significant; N = Nitrogen; P = Phosphorous; K = Potassium; Ca = Calcium; PGRs = Plant Growth Regulators

Similar letter in columns represents statistically similar results.

tree⁻¹)+Kinetin at 30 mg l⁻¹, whereas minimum TSS (8.77°Brix) on day-1 was recorded with P (500 g tree⁻¹). The TSS of juice increased during seven days of ambient storage and significant increment was recorded in Ca and Ga₃ treated fruits. This increase in TSS may perhaps be due to conversion of complex carbohydrates into simple sugar (Singh and Sharma, 2011). Maximum titratable acidity (0.64%) of juice was assessed by Kinetin application at 60 mg l⁻¹, whereas minimum titratable acidity (0.53%) was detected by P application at 250 g tree⁻¹. Titratable acidity (%) declined during seven days ambient storage (Table 5). This decrease in acidity could be due to more breakdowns of organic acids into sugar and salts during the processes of respiration (Rutter et al., 1975).

Contrasts analysis exposed that use of Wokozim significantly enhanced TSS when compared with control (Table 5). Phosphorus fertilizer considerably reduced TSS compared to Wokozim and Kinetin treatments.

Maximum TSS:TA (19.8) was noted with NPK (230 + 250 + 250 g tree⁻¹)+Ca (150 g tree⁻¹) + Kinetin at 30 ml l⁻¹, whereas minimum TSS:TA ratio (15.8) was detected with P (250 g tree⁻¹)+Kinetin at 30 ml l⁻¹ (Table 6). The TSS:TA ratio significantly improved during the shelf life duration. Increased TSS:TA in the course of ambient storage may possibly be due to rise in TSS and fall in acidity (Table 5) during storage. Maximum ascorbic acid (57 mg 100 ml⁻¹) was perceived with application of Ca (150 g tree⁻¹)+P (250 g tree⁻¹)+Kinetin at 30 ml l⁻¹, whereas minimum ascorbic acid (37.6 mg 100 ml⁻¹) was observed with Wokozim (1500 g tree⁻¹)+2 ml l⁻¹ foliar spray (Table 4). Decrease in ascorbic acid in the Wokozim treatment (Table 6) could be due to the presence of auxin and cytokinin in Wokozim formulation. These two PGRs might improve reproductive growth (sink strength) of the plant. As increased reproductive growth in Arabidopsis mutant vtc 1 resulted in reduced ascorbic acid concentrations (Barth et al., 2006). Interaction of treatment and storage interval showed that more ascorbic acid
Table 6. Influence of PGRs and nutrients on TSS:TA and ascorbic acid (mg 100 ml⁻¹) of ’Kinnow’ mandarin juice.

| Treatments                                                      | TSS:TA | Ascorbic acid (mg 100 ml⁻¹) |
|-----------------------------------------------------------------|--------|---------------------------|
|                                                                 | *Day-1 | **Day-7 | Mean | *Day-1 | **Day-7 | Mean |
| Control                                                         | 14.8   | 20.0 | 17.4bcdef | 36.6fg | 47.3cdef | 41.9cd |
| P (250 g tree⁻¹)                                                | 15.4   | 21.5 | 18.4abcd | 36.6fg | 58.1abcd | 47.4abc |
| P (500 g tree⁻¹)                                                | 14.8   | 18.5 | 16.8cdef | 45.2defg | 58.1abcd | 51.6abc |
| NPK (230 + 250 + 250 g tree⁻¹)+Ca (150 g tree⁻¹)                | 15.4   | 20.8 | 18.1abcede | 43.0efg | 60.2abc | 51.6abc |
| Wokozim (granular) (1500 g tree⁻¹)                              | 13.4   | 19.7 | 16.6def | 36.6fg | 47.3cdef | 42.0cd |
| Wokozim (1500 g tree⁻¹) + 2 ml l⁻¹ foliar spray                 | 13.8   | 20.0 | 16.9cdef | 34.3 | 40.9efg | 37.6d |
| Kinetin at 30 mg l⁻¹                                           | 15.1   | 20.4 | 17.8bcdef | 45.2defg | 53.8abced | 49.5abc |
| Kinetin at 60 mg l⁻¹                                          | 13.6   | 18.5 | 16.1ef | 40.9efg | 58.1abcd | 49.5abc |
| Kinetin 120 mg l⁻¹                                            | 15.1   | 18.2 | 16.7cdef | 40.9efg | 51.6bcd | 46.2bcd |
| P (250 g tree⁻¹) + Kinetin at 30 mg l⁻¹                         | 13.1   | 18.5 | 15.8f | 47.3cdef | 62.4ab | 54.8ab |
| Kinetin at 30 mg l⁻¹+ GA₃ at 10 mg l⁻¹                          | 13.2   | 20.9 | 17.0cdef | 45.2defg | 53.8abced | 49.5abc |
| P (250 g tree⁻¹)+Kinetin at 30 mg l⁻¹+ GA₃ at 10 mg l⁻¹         | 16.7   | 22.3 | 19.5ab | 49.5bcdef | 58.1abcd | 53.8ab |
| NPK (230 + 250 + 250 g tree⁻¹)+Ca (150 g tree⁻¹)+Kinetin at 30 mg l⁻¹ | 16.2   | 23.4 | 19.8a | 51.6bcde | 47.3cdefg | 49.4abc |
| Ca (150 g tree⁻¹) + P (250 g tree⁻¹)+Kinetin at 30 mg l⁻¹       | 14.4   | 23.4 | 18.9abc | 66.7a | 47.3cdefg | 57.0a |
| Ca (150 g tree⁻¹)                                               | 15.0   | 22.0 | 18.5abcde | 53.8abced | 43.0efg | 48.4abc |
| Treatments (LSD)                                               | 1.95   |     | 8.56 |
| Treat. × SD (LSD)                                               | NS     | 12.11 |
| Significance of selected contrasts on D-1                      | TSS:TA | Ascorbic acid (mg 100 ml⁻¹) |
| Control vs P                                                   | NS     | NS |
| Control vs Kinetin                                             | NS     | NS |
| Control vs Wokozim                                            | NS     | NS |
| P vs Kinetin                                                   | NS     | NS |
| P vs Wokozim                                                   | NS     | NS |
| Wokozim vs Kinetin                                             | NS     | NS |

NS = Non significant; N = Nitrogen; P = Phosphorous; K = Potassium; Ca = Calcium; PGRs = Plant Growth Regulators

*Similiar letter in columns represents statistically similar results.*

(66.7 mg 100 ml⁻¹) was detected on the first day, with Ca (150 g tree⁻¹)+P (250 g tree⁻¹)+Kinetin at 30 ml l⁻¹ whereas less ascorbic acid (34.3 mg 100 ml⁻¹) was noted on the first day with Wokozim (1500 g tree⁻¹)+2 ml l⁻¹ foliar spray. Ascorbic acid significantly enhanced during ambient storage for seven days.

**Conclusion**

In conclusion, Ca and low P improved aborted seed (%) and ascorbic acid, whereas Kinetin, GA₃ and Wokozim improved healthy seed (%), but reduced ascorbic acid concentrations in ’Kinnow’ mandarin juice.

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The authors announce that they have no conflict of interest.

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