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ROLE OF GROWTH REGULATORS ON PREHARVEST FRUIT DROP, YIELD AND QUALITY IN KINNOW MANDARIN

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Abstract

The study was undertaken to envisage the effect of growth regulators on preharvest fruit drop and quality of Kinnow mandrin (*Citrus reticulata* Blanco). Various concentration of 2, 4-D, GA_3 and NAA were applied during the last week of November, 2005 to check the impact of various treatments on preharvest fruit drop, yield and fruit quality. Exogenous application of growth regulators significantly decreased preharvest fruit drop percentage, leading to increase in total number of fruits per plant, fruit weight, juice percentage, total soluble solids, acidity, vitamin-C, reducing sugars and non reducing sugars %age while no effect was observed on fruit size. Auxin (2,4-D and NAA) performed better compared to gibberellins.

Introduction

Citrus is an important genus of the family Rutaceae in the plant kingdom. Its importance is demonstrated by its wide distribution and large-scale production. It is highly prized and economically remunerative fruit. Citrus fruits have special importance due to their distinct flavors and therapeutic values. These are rich in vitamin-C with fair amounts of vitamins A & B. Besides this, they are rich source of minerals (calcium, phosphorus and iron). The juice is very refreshing, delicious and soothing (Ahmed *et al.*, 2007). Alongwith their consumption as fresh fruit, a large number of products and byproducts are prepared and marketed with premium price (Niaz *et al*, 2004). Citrus production is 108 million tons in the world. Brazil is the largest producer of citrus worldwide followed by USA, China and Mexico. Pakistan is among the top thirteen citrus producing countries of the world. In the country citrus is cultivated over an area of 185, 000 hectares with an annual production of 1.67 MT tones (Anon., 2005). Citrus fruits hold first position in production and have a share of about 34% in total fruit production of the country; over 93% of the total cultivated area for citrus in Punjab is under Kinnow mandarin. Kinnow is commercially cultivated due to its good yield, high processing quality, fresh consumption, aromatic flavor and better adaptation to agro-environmental conditions of Punjab (Ahmed *et al.,* 2006).

Progressive farmers prefer to grow kinnow because of its high yielding characteristics and its attractive quality that possesses the potential to give the lucrative return in form of profit. But even then, average yield of Kinnow in Pakistan is just 9.0 tons ha^{-1}, while in many other citrus growing countries it is much higher, as average yield of Brazil is 21.64 tones ha^{-1}, USA 25.98 tones ha^{-1} and of Turkey 26.73 tones ha^{-1}. Preharvest fruit drop is major reason of low yield in Pakistan, this drop of fruit at various stages of fruit development is due to malnutrition, water stress, excessive insect pest attack and most important is the hormonal imbalance. Tree drops its fruit when the concentration of auxins decreases and the concentration of abscissic acid (ABA) increases (Browning, 1986; Marinho *et al.*, 2005) as the endogenous hormones and their balance play a modulating role in the mobilization of nutrients to the developing organs.

The use of growth regulators has become an important component of agrotechnical procedures for most of the cultivated plants and especially for fruit plants (Monselise, 1979). So in citrus fruits, excessive fruit drop can be controlled by the exogenous application of plant growth regulators. The auxins and gibberillins are used to control the fruit drop in citrus and to improve the quality of fruit (Almeida *et al.*, 2004). Although some references are available in the literature and efforts have been made to control the fruit drop by exogenous application of growth regulators but there is no precise recommendation for the control of fruit drop in Kinnow mandarin. Moreover it has also been reported that the pre-harvest drop of Valencia orange in Florida was not reduced by the application of plant growth regulators, stressing the importance of climatic conditions on the effectiveness of growth regulators treatment (Greenberg *et al.*, 1975).

So there was a need to test the efficacy of plant growth regulators to reduce fruit drop and improve the quality and yield under agro-environmental conditions of Punjab-Pakistan. This research was initiated as a preliminary effort however it is well planned and it provides necessary efficacy data for the registration of use of plant growth regulators (PGRs) on Kinnow mandarin in Pakistan.

Materials and Methods

The research work was conducted at fruit experimental Orchard Sq. No. 9, Institute of Horticultural Sciences, University of Agriculture, Faisalabad during 2005-2006. Laboratory work was carried out in Postgraduate Pomology Laboratory, Institute of Horticultural Sciences, University of Agriculture, Faisalabad. Ten years old, 30 plants of Kinnow mandarin (*Citrus reticulata* Blanco) grafted on Rough lemon rootstock of uniform size and age were selected for this experiment. Three growth regulators 2, 4-D (10, 20, 30ppm), GA_3 (10, 50, 100ppm) and NAA (10, 15, 20ppm) were applied during the last week of November, 2005 to check the effect on preharvest fruit drop and physiochemical properties of the fruit. The experiment consisted of 10 treatments including control, replicated thrice and single tree was taken as an experimental unit. All the experimental trees were maintained under similar agro-climatic condition.

Number of fruits per plant at spray time and after spray was counted by tagging 4 braches of one inch diameter on each side of the tree. To calculate fruit drop %age from tagged branches of the experimental tree, number of fruits were counted and fruit drop percentage was calculated using the following formula:

$$\text{Fruit drop \%age} = \frac{\text{Total number of dropped fruits}}{\text{Total number of fruits before application}} \times 100$$

Yield per tree was recorded by weighing and counting total number of fruits per tree at the time of harvesting. Fruit size was measured by measuring the diameter of 10 fruits per tree randomly with the help of Vernier caliper from each experimental tree. Average fruit weight was calculated by weighing ten fruits per tree on digital UWE–ESP Digital Electric Balance and average weight was calculated.

Juice of each of 10 harvested fruit was extracted and weighed; average juice weight was calculated separately for each treatment. The average juice percentage per fruit was obtained from the following formula:

$$\text{Juice \%age} = \frac{\text{Juice weight per fruit}}{\text{Average fruit weight}} \times 100$$

Total Soluble Solids were measured by automatic digital refractometer (ATAGO, RX 5000) by placing 1-2 drops of juice on the prism of refractometer. Acidity of juice was determined by taking 10 ml of juice from each sample and diluted with distilled water in a 100 ml beaker; 2-3 drops of phenolphthalein were added for end point. The samples were titrated against N/10 NaOH (Hortwitz, 1960). The results were expressed as percent citric acid.

$$\text{Acidity \%age} = \frac{\text{N/10 NaOH used} \times 0.0064}{\text{Volume or weight of sample used}} \times 100$$

Vitamin-C in juice was estimated according to the method described by Ruck (1961) whereas sugars in juice were estimated by using Lane & Eynon method (1923) described by Hortwitz (1960).

Results and Discussion

There is no argument for the importance of citrus cultivars and growth regulators to desert citrus production. Growth regulators in citriculture are known to have a profound effect on the tree vigor, health, yield and quality of fruit.

Physical characteristics

Preharvest fruit drop: Preharvest drop of the fruit is of commercial loss to farmer as the drop occurs just before harvesting when fruit is physiologically mature. The perusal of the Table 1 shows that all the growth regulators treatments (2, 4-D, GA_3 and NAA) significantly reduced the preharvest drop compared to control. The lowest fruit drop of 12.95% was observed in T_3 (20 ppm 2, 4-D) followed by T_2 (10 ppm 2, 4-D) and T_7 (20 ppm GA_3) with a fruit drop of 15.02 and 16.20% whereas, maximum fruit drop (49.03%) was found in T_1 (Control). It is also clear from the results that 2, 4-D treatments proved better compared to GA_3 and NAA but when the concentration of 2,4-D was increased upto 30ppm, fruit drop was also increased. GA_3 was at 2nd position in controlling the preharvest fruit drop. Our results were found to be in agreement with that of Almedia *et al.,* (2004) and Davies & Zalman (2006) who reported that application of 2,4-D, GA_3 some other plant growth regulators significantly reduced the preharvest fruit drop in citrus species. Keeping in view the above results it can safely be recommended that 2,4-D can be applied at 20ppm to control the preharvest fruit drop in Kinnow mandarin.

Fruit diameter (mm): Fruit diameter is of commercial importance for citrus fruits marketing and trade/business. It is generally considered that in citrus with excessive increase in size the quality is impaired, while on the other side small sized fruits are of low quality. The results showed non-significant differences. On 27-12-2005 maximum fruit size (71.20 mm) was found in case of T_5 (10 ppm GA_3) followed by T_2 (10 ppm 2, 4-D) and T_3 (20 ppm 2, 4-D) with a fruit size of 70.48 and 70.05 mm respectively. The minimum fruit size of 66.52 mm was observed in case of T_9 (15 ppm NAA).

Similarly on 02-02-2006 maximum fruit size (72.80) was observed in case of T_5 (10 ppm GA_3) and minimum fruit size (67.06) was observed in non treated fruits as shown in Table 1. It can be inferred that preharvest application of growth regulators has no effect on fruit size but the quality of the fruit can be improved and harvesting can be delayed as described earlier.

Fruit weight (g): The data on fruit weight showed non-significant results for both the time of analysis. On 27-12-2005 highest weight per fruit of 154.80g was recorded in T2 (10 ppm 2, 4-D) followed by T_3 (20 ppm 2, 4-D) and T_5 (10 ppm GA_3) with 154.13 and 152.16g respectively, whereas, minimum fruit weight of 141.66g was found in case of control.

During 2nd analysis on 02-02-2006, highest fruit weight of 155.53g was found in T_7 (100 ppm GA_3) followed by T_2 (10 ppm 2,4-D) and T_3 (20 ppm 2,4-D) with 155.03 and 154.86g respectively while, lowest fruit weight of 141.95g was observed in case of T_1 Control. By critical observation of Table 1, it becomes clear that a slight increase in fruit weight was recorded, but it was non-significant, so it could be inferred that this slight increase in weight may be due to the application of the growth regulators. However, as the results are non-significant it is due to the reason that as the application of growth regulators is at preharvest stage, almost fruit has already gained the size and has almost completed the cell division and enlargement phase that is why the application of growth regulators at preharvest stage proved unsuccessful to increase per fruit weight. The results were found to be in agreement with that of Saraswathi *et al.*, (2003) who observed that growth regulators 2, 4-D and GA3 and their combinations significantly influenced the fruit weight.

Yield: Yield is a horticultural trait of immense importance. The data for yield was taken by counting the total number of fruits harvested per plant and also by weighing their corresponding weight per treatment.

Number of fruits per plant: The results regarding number of fruits per plant showed significant results. Maximum numbers of 708 fruits per plant were recorded in T3 (20 ppm 2, 4-D followed by T_9 (10 ppm NAA) and T_4 (30 ppm 2, 4-D) having 686.66 and 662.45 fruit per plant respectively. However the lowest numbers of fruits (420.59) were recorded in case of T_1 Control. In general Auxins (2,4-D and NAA) performed better to reduce fruit drop and to increase the final yield of the crop compared to GA_3 (Table 1).

Fruit weight per plant (Kg): The results showed significant differences among the various treatments (Table 1). Maximum fruit weight per plant (103.55kg) was found in T_3 (20 ppm 2, 4-D) closely followed by T_9 (10 ppm NAA) and T_4 (30 ppm 2, 4-D) with a fruit weight of 102.80 and 101.66 kg per plant respectively. Here again auxin (2,4-D and NAA) exceeded the GA_3 and proved their superiority to increase fruit weight per plant. The lowest fruit weight of 58.73kg per plant was found in case of T_1 (Control) where no growth regulators were applied. The application of growth regulators treatments significantly increased the fruit weight per plant as compared to control. Results regarding yield were found to be in agreement with that of Thomas & Lovatt (2004) and Davies & Zalman (2006) who reported that preharvest application of growth regulators like 2,4-D and GA_3 significantly increased the total number of fruits at the time of harvest and fruit weight per plant by reducing the preharvest fruit drop.

Juice percentage: Juice is an extremely important parameter for its industrial processing, being also related to size, which in turn, although determined by the genetic characteristics of each cultivar, but is affected by cultural practices such as application of plant growth regulators. The results showed significant differences for juice percentage among the various treatments. The perusal of Table 1 on 27-12-2005 shows that highest juice percentage (52.16%) was observed in T_6 (50 ppm GA_3) followed by T9 (15 ppm NAA), T_7 (100 ppm GA_3) and T_5 (10 ppm GA_3) with a juice percentage of 50.89, 50.30 and 50.08% respectively, these treatments were found statistically at par with each other. While the lowest juice percentage (41.87) was found in case of T_1 (Control). So it is evident that GA_3 treatments proved superior to increase juice percentage.

On the 2nd date of analysis (02-02-2006) maximum juice percentage (51.66) was found in case of T_7 (100 ppm GA_3) followed by T_5 (10 ppm GA_3) and T_6 (50 ppm GA_3) with a juice percentage of 49.99 and 49.90% while minimum juice percentage (43.74) was again found in T_1 (Control). So it becomes clear that GA_3 treatments proved excellent as for as juice percentage is concerned whereas 2,4-D and NAA also increased juice percentage as compared to non-treated fruits T_1 (Control). Results regarding juice percentage were found to be in consonance with that of Atawia & El-Desouky (1997) and Matthew *et al.*, (2002) who earlier reported that the application of growth regulators at flowering and preharvest significantly increased the juice percentage in various citrus species.

Chemical characteristics

Total soluble solids (TSS) (%): Total soluble solids measurement is considered to be an important parameter of quality of citrus fruits. Observations were recorded for both the times during analysis and processed for statistical analysis. During 1st analysis on 27-12-2005 maximum TSS (10.94%) was observed in T9 (15 ppm NAA) followed by T_7 (100 ppm GA_3) and T_8 (10 ppm NAA) with a TSS of 10.78 and 10.67% (Table 2) while, the minimum TSS (9.42%) was observed in case of T_1 (Control). It is clear from the Table 2 that all the treatments of growth regulators increased the TSS value. It is also important to note that TSS is an important parameter to know the time of harvesting; in case of Kinnow mandarin which is a late maturing cultivar optimum TSS (11.00) is required. Keeping in view we can observe that the harvesting time can be obtained 15 days earlier just by the preharvest application of 15 ppm NAA T_9 (15 ppm NAA).

Similarly during 2nd analysis of the fruit on 02-02-2006 the growth regulators treatments significantly increased the TSS compared to control. In this case maximum TSS (12.03%) was observed in case of T_4 (30 ppm 2, 4-D) followed by T_3 (20 ppm 2, 4-D) and T_9 (15 ppm NAA) with values of 11.93 and 11.79% respectively. Whereas, minimum TSS value (10.43) was again found in T_1 (Control). Results regarding TSS percentage were found to be in consonance with that of Atawia & El-Desouky (1997) and Huang & Huang (2005) who reported that by application of growth regulators like Auxin and Gibberellins we can significantly increase the total soluble contents of the fruit in citrus species.

Acidity percentage: The information procured for the determination of acidity in fruits sprayed with various growth regulators showed significant results for both the times of fruit analysis. All the growth regulators decreased the acidity percentage in Kinnow (Table 2). However, on 27-12-2005 maximum acidity (1.33%) was observed in case of T9 (15 ppm NAA) followed by T_7 (100 ppm GA_3) and T_1 (Control) with acidity of 1.28 and 1.26% respectively, while minimum acidity (1.04) was found in case of T_2 (10ppm 2,4-D). Normally citrus fruits are harvested when its acidity reaches to 1 or less than 1.00% and this is obtained at 15th of January in case of our local conditions. From the Table 2 we can observe that 1.04% acidity was obtained on 27 December 2005 just by foliar application of 10ppm 2,4-D (T_2 (10 ppm 2,4-D)). Similarly, when 2nd analysis of the fruit was done on 2 February 2006 the highest acidity (1.04%) was again observed in case of T_9 (15 ppm NAA) followed by T_{10} (20 ppm NAA) and T_1 (Control) with the values of 1.02 and 1.00% respectively; and minimum acidity (0.78) was found again in T_2 (10 ppm 2, 4-D). From the data presented in Table 2 it becomes obvious that for both the time of analysis the lowest acidity was found in the fruit that were sprayed by the 10ppm 2,4-D T_2 (10 ppm 2,4-D).

It is also important to note that as the concentration of 2,4-D increases the acidity is also increasing (Table 2) which suggest that for quality improvement 2,4-D should be applied at low concentrations. GA_3 application also proved helpful to reduce acidity as compared to NAA. Results related to acidity percentage of fruit were found to be in close agreement with that of Otmani *et al.,* 2004 and Xiao *et al.,* (2005) who reported that by the application of 2,4-D, GA_3 and NAA acidity percentage was significantly reduced.

Vitamin-C (mg/100g): Vitamin-C is a powerful antioxidant and is an important part of human feed. It helps to save the human from many serious diseases and scavenges the reactive oxygen species (ROS) produced in the body. Vitamin-C (Ascorbic acid) contents in fruits varies in concentration for various citrus spp.; vitamin-C is being affected by the environmental factors, time of fruit harvesting, plant vigour, age of plant and by application of growth regulators. So the vitamin-C was measured as quality parameter for the plants sprayed with various growth regulators. Observations revealed the significant results for treatments. The Table 2 shows that on 27-12-2005 maximum vitamin-C contents (25.67 mg/100g) were observed in case of T_8 (10 ppm NAA) closely followed by T_9 (15 ppm NAA) and T_6 (50 ppm GA_3) with values of 24.37 and 24.13 mg/100g while minimum vitamin-C contents (17.76) were observed in case of T_1 (Control). Similarly on 02-02-2006 maximum vitamin-C contents (45.30 mg/100g) were observed again in T_9 (15 ppm NAA) followed by T_4 (30 ppm 2, 4-D) and T_7 (100 ppm GA_3) with 44.44 and 41.88 mg/100g respectively.

It is obvious from the Table 2 that all the growth regulators treatments significantly increased the vitamin-C contents of Kinnow. 2,4-D and NAA treatments proved better compared to Gibbrellic acid treatments; as the concentration of Auxin (2,4-D or NAA) increases vitamin-C contents also increases. With the passage of time the concentration of Vitamin-C increases so the fruits which are harvested earlier have less amount of Vitamin-C compared to the fruits which are harvested later. Results regarding this parameter of study were found to be in agreement with that of Xiao *et al.,* (2005) who also observed that preharvest application of growth regulators increased vitamin-C contents of the citrus fruits.

Sugar contents (%): Sugars are an important parameter of quality measurement in citrus fruits as they are main and ready source of energy when used by human; so keeping this aspect the sugars of the fruit were analyzed at two different times of fruit harvesting and the effect of various treatments were studied. Sugars are divided into two groups i.e., non-reducing sugars and reducing sugars. First of all reducing sugars and total sugars were measured while non-reducing sugars were calculated by subtracting reducing sugars from total sugars.

Reducing sugars (%): The data regarding reducing sugars showed significant results for various treatments. On 27-12-2005 highest reducing sugar contents (3.44%) were observed in T_4 (30 ppm 2, 4-D) closely followed by T_3 (20 ppm 2, 4-D) with a value of 3.33% and was statistically at par with T_4 (30 ppm 2, 4-D). Whereas, minimum reducing sugar contents (2.64%) were found in case of T_{10} (20 ppm NAA). It is also important to note that all the GA_3 treatments proved inferior to increase sugar contents; they performed poorly and in GA_3 treatments reducing sugar contents were even less compared to control (Table 2). Similarly on 02-02-2006 maximum reducing sugar contents (4.22) were again found in T4 (30 ppm 2, 4-D) whereas, minimum reducing sugar contents (3.20%) were found in T_6 (50 ppm GA_3). It becomes obvious that 2,4-D treatments performed better to increase reducing sugar contents as compared to other growth regulators applied (Table 2). This time also GA_3 performed poorly; and more sugar contents were observed in control as compared to GA_3 treatments.

Non-reducing sugars (%): The non-reducing sugars were determined by subtracting reducing sugars from total sugars and were subjected to statistical analysis. The results showed significant differences for 1st date of analysis (27-12-2005) while non-significant results for 2nd date of analysis (02-02-2006).

On 27 December 2005 maximum non-reducing sugars (5.75) were found in T_4 (30 ppm 2, 4-D) followed by T5 (10 ppm GA_3) and T_3 (20 ppm 2, 4-D) with a sugar contents of 5.45 and 5.43% respectively, while, lowest non-reducing sugars (4.21) were observed in T_8 (10 ppm NAA). In case of non-reducing sugars 2,4-D treatments performed better compared to other whereas NAA was at the bottom. T_1 (Control) was found better among many other treatments; so suggesting that application of growth regulators has less prominent effect on non-reducing sugars in Kinnow.

On 2^{nd} date of analysis (02-02-2006) maximum non-reducing sugar contents (4.62) were found in T_1 (Control) and minimum non-reducing sugar contents (3.71) were recorded in T_9 (15 ppm NAA). It is also imperative to note that non-reducing sugar contents were non-significant on 2^{nd} date (02-02-2006) of analysis while significant on 1st date of analysis (27-12-2005) suggesting that the efficacy of plant growth regulators to alter the physiochemical properties is transient.

Total sugars (%): Total sugars were determined and the data were subjected to statistical analysis; the results showed significant results for 1^{st} date of analysis (27-12-2005) while non-significant results for 2^{nd} date of analysis (02-02-2006). On 1^{st} date of fruit analysis (27-12-2005) maximum total sugars (8.86) were found in T_4 (30 ppm 2, 4-D) followed by T_3 (20 ppm 2, 4-D), T_2 (10 ppm 2, 4-D) and T_5 (10 ppm GA_3) with total sugar contents of 8.76, 8.36 and 8.36% respectively, whereas, minimum sugar contents (6.94%) were recorded in T_{10} (20 ppm NAA).

Similarly on 2nd date of analysis (02-02-2006) maximum sugar contents (8.71%) were observed in T_4 (30 ppm 2,4-D) while minimum sugar contents (7.15%) were recorded in T_{10} (20 ppm NAA) as presented in Table 2. The results regarding sugar contents were found in consonance with that of Ingle *et al.* (2001) and Wang *et al*., (2004) who find that application of 2,4-D, GA_3 and some other growth regulators increased the sugar contents in various mandarin and sweet orange cultivars.

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