

Quality and Quantity Improvement of Citrus: Role of Plant Growth Regulators

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Abstract

Citrus is one of the most important fruit tree species in the world, as the fruits are a valuable source of nutrients, vitamins and other antioxidant compounds. The citrus productivity depends on various factors, among these the plant growth regulators holds a prime position. The use of plant growth regulators has become an important component in the field of citriculture because of the wide range of potential roles they play in increasing the productivity of crop per unit area. The plant growth regulating compounds actively regulate the growth and development by regulation of the endogenous processes and their exogenous applications have been exploited for modifying the growth response. Plant growth regulators have been used in citrus fruit production for influencing flowering, fruit set and fruit drop and play a major role in fruit growth and abscission. These regulators have also been used to influence fruit quality factors like peel quality and colour, fruit size, juice quality and to improve total soluble solids in different citrus species. This review may serve as a complete treatise on the possible roles of growth promoting substances on the physiological processes of citrus plant.

Highlights

- Impact of plant growth regulators on fruit quality and productivity of citrus

Keywords : citrus, plant growth regulators, flowering, fruit growth, fruit quality

The growth and development in citrus like all other fruit crops is governed by both the intrinsic as well as extrinsic factors. The intrinsic factors comprise of the genotype and the endogenous hormone balance, whereas, the extrinsic factors are the environmental conditions encountered by the plant. There are other factors, viz., planting density, irrigation and fertilization, which may affect the growth and development of the plant. Since the discovery of the plant growth regulators, they have been used to manipulate plant growth and development for the improvement of quality and quantity of the produce in order to enable the fruit growers to meet

to pressure of increasing demand for food of high quality.

In citrus industry, a large number of plant growth regulators have been evaluated for their commercial application. Plant growth regulating chemicals are used in citrus production in all the citrus producing areas. They are used for production technology and quality improvement. In production technology, emphasis is primarily on nursery production, crop regulation by manipulation of flowering, improvement of fruit set and fruit growth and manipulation of the time of harvest. The application of plant growth regulators for improvement of



internal fruit quality and postharvest fruit storage is of equal importance.

Plant growth regulators have been used in citrus fruit production for influencing flowering, fruit set and fruit drop (Berhow 2000). These regulators have also been used to influence fruit quality factors like peel quality and colour, fruit size, juice quality and to improve total soluble solids in different citrus species. Among PGRs, the auxins have direct effect on abscission by causing a delay of abscission resulting in improvement in fruit quality and yield in citrus. Naphthyl acetic acid, 2-4 Dichlorophenoxy acetic acid and Gibberellic acid have been tried for reduction of physiological drop (Ullah *et al.* 2014). Salicylic acid is considered a potent plant hormone because of its diverse regulatory roles in plant metabolism. It is well established that salicylic acid potentially generates a wide array of responses in plants and also affects the photosynthetic parameters which enhance yield (Mahdi *et al.* 2012).

The poor tree health also plays a significant role in enhancing of fruit drop due to depletion of nutrient supply to the demanding sinks. Citrus is however, a relatively high nutrient demanding crop (Wang *et al.* 2006) and deficiency or excess of nutrients can lead to inferior fruit quality. There is a need to boost up yield through proper nutrition and maintaining internal hormonal balance. Suitable combination of micronutrients and growth regulators may control excessive fruit drop for the improvement of fruit yield and quality. Although many efforts have been made to study (Baldwin 1993) the physiological and biochemical aspects of citrus, there is still an enormous unexplored potential in the study of regulation of metabolites associated with citrus physiology.

Citrus fruits use large amount of K as compared to other macronutrients (Alva and Tucker 1999) because K is involved in several basic physiological functions i.e. formation of sugars and starch, synthesis of proteins, cell division, growth and neutralization of organic acids (Liu *et al.* 2000). It improves fruit quality through enhancing fruit colour, size and juice flavor (Tiwari 2005). Reports indicate the deficiencies

of micronutrients like Zn, Cu, Fe and Mn in citrus orchards of India and among them Zn is more acute. Literature indicates that the application of Zn increases the fruit yield and quality (Rodriguez *et al.* 2005). The suitable combinations of macronutrients, micronutrients and growth regulators could control the excessive fruit drop and improve the citrus fruit yield and its quality (Doberman and Fairhurst 2000).

The literature pertaining the role of plant growth regulators during different periods of growth and development is reviewed under the following heads:

Role of Plant Growth Regulators

Flowering

Citrus trees, once past the juvenile phase, bloom every year. Annual flowering of adult trees is affected by several exogenous and endogenous factors. In citrus, cool temperature can induce flowering, as in most tropical and subtropical trees (Inoue 1990, Lenz 1967, Moss 1976, Nishikawa *et al.* 2007, Wilkie *et al.* 2008). In Satsuma mandarin, floral induction occurs in trees exposed to 15°C for more than 1.5 months (Inoue 1990b, Nishikawa *et al.* 2007). Trees generally remain in the vegetative growth phase until the trees are exposed to temperatures of less than 25°C (Inoue and Harada 1988). Under field conditions, the trees are exposed to cool temperature during fall, during which floral induction proceeds. The plant growth regulators stimulate the abscission in the flowers that causes heavy flower drop. According to Martinez *et al.* (2004) flowering of *Hernandina* gets reduced by 25% and of *Orogrande* by 60% when GA3 (20-50 mg/l) when given as a foliar spray (6 L per tree) to all the citrus trees. During bud development in citrus, the application of GA3 has shown to inhibit flower production (Guardiola *et al.* 1982), leading to greater ratio of terminal flowers in the leafy shoots thus higher development of fruits (Iglesias *et al.* 2007). These results were also shown by use of ethchlozate and GA3 for flower induction in citrus fruits but GA3 caused inhibitory effect (Takahara *et al.* 2001). According to Ben-Cheikh *et al.* (1997) gibberellins are the factors responsible for ovary transition. In vegetative organs, gibberellins activate the process of



cell division and cell enlargement (Talon *et al.* 1991) thus are also associated with initiation of growth (Talon and Zeevart 1992). Gibberellins reduce the flower production resulting in higher productivity of better quality fruits. It acts like a thinner agent but it also showed the ability to retain the flowers (Iglesias *et al.* 2007) whereas 2,4-D delay or stimulate the abscission. Talon and Zeevart (1992) reported that in citrus the reproductive processes are affected by plant growth regulators showing that regulatory mechanism being controlled by critical hormonal component.

The phenomenon of flowering in plants is under the control of many factors, viz., cultivar, genetic makeup, the environment and cultural practices. Most of the citrus cultivars flower profusely subject to optimum environmental conditions and there are more than 100,000 flowers per tree (Agusti *et al.* 1982). Depending on the cultivar, most of the flowers abscise leading to less than 1% fruit set (El-Otmani *et al.* 1992). The application of plant growth regulators is to regulate tree productivity either by reducing vegetative growth and enhancing flowering (during 'off-year') or by enhancing vegetative growth and reducing flower initiation and development ('on-year'). Monselise and Halevy (1964) reported that gibberellic acid could inhibit flowering in 'Shamouti' orange. Similar inhibitory effect having been reported by Davenport (1990). However, the triazole compounds that inhibit gibberellic acid biosynthesis have been reported to promote inflorescence production (Delgado *et al.* 1986 1986b; Harty and van Staden 1988). It is the time of application and concentration which determines the action of a particular growth promoter (Guardiola *et al.* 1982, Lord and Eckard 1987). In subtropical climates of the northern hemisphere floral induction occurs during the period from November through January. For sweet oranges and some mandarins induction occurs early during this period (Guardiola *et al.* 1982), whereas for 'Satsuma' mandarins it occurs later (Iwahori and Oohata 1981). The optimum concentration for promotion of flowering are GA3 (25 µg/ml) for 'Navelate' sweet orange with the application in mid-December and for 'Washington

Navel' with the appropriate time of application being mid-November in Spain (Guardiola *et al.* 1977). Optimum concentrations (100 mg/L) and time of application (late January) for 'Satsuma' mandarin in Japan is reported by Iwahori and Oohata (1981). Agusti *et al.* (1981) concluded that GA3 (10 mg/l) applied in late November could reduce flowering to 40% in 'Clementine' mandarin irrespective of the date of harvest (i.e., the presence of the crop was not inhibiting flowering). Low GA3 concentration reduced flowering by 37 and 70% when it was applied to 'Satsuma' mandarin trees in mid-December (Garcia-Luis *et al.* 1986) and late December (Guardiola *et al.* 1982), respectively. However, the duration of sensitivity is very short and treatments must be applied before the developing shoots are more than 1mm long (Guardiola *et al.* 1982), which corresponds to the stage of full sepal development of the apical flower (Lord and Eckard 1987).

Attempts to promote flowering using growth retardants that are reported to inhibit synthesis of gibberellins viz., CCC and paclobutrazol have not been able to provide conclusive results as reported by Harty and van Staden (1988) and Davenport (1990). Greenberg *et al.* (1993) reported that the application of paclobutrazol (foliar spray or soil application) shifts the balance of shoot types toward the pure leafless inflorescences, which is just the reverse of GA3, which pushes the balance toward vegetative, flowerless shoots.

There are reports of role of auxins in the process of flowering by Zeevart (1978). Application of 2,4-D during mid-November to mid-December to 'Navelate' sweet orange reduced flowering by approximately 30% (Guardiola *et al.* 1977). Higher concentrations of synthetic auxins did not provide any additional effect (Agusti 1980) and lower rates did not show inhibitory effect (Garcia-Luis *et al.* 1986). Abscisic acid may also be involved in the regulation of flowering (Young and Cooper 1969). The role of cytokinins in the regulation of flowering in citrus has received little attention, although a correlation between endogenous cytokinin levels and budbreak and growth has been reported (Davenport 1990). It



has been argued by Davenport (1990) that lack of flowering during (off-bloom) when trees are bearing the heavy 'on-year crop' for severely alternate bearing cultivars, is likely due to lack of cytokinins. The low level of cytokinins are due to lack of carbohydrates and other nutrients necessary for active root growth and for the production of cytokinins.

Gibberellins appear to be the only plant growth regulators that consistently inhibit flowering, but the use of gibberellic acid as a commercial regulators for reducing flower number to increase fruit size or in an expected 'on-year' to reduce alternate bearing is limited. Climatic conditions, i.e., temperature extremes that might prevail during flowering invocation and induction may result in poor fruit set or excessive fruit drop resulting in low yield.

Crop Regulation

Consumer preference worldwide is for large sized and healthy fruits (Gilfillan 1987, Miller and Hofman 1988). Fruit juice acidity (percent citric acid) and juice volume are very much affected by fruit size. Small fruit have a substantially higher per cent acid than larger fruit and total soluble solids per fruit is considerably lower in small fruit. Reducing the proportion of small fruit harvested increases grower return both for fresh fruit (because there are more fruit of commercially valuable export size) and for juice (because of increased juice and TSS). Small fruit size results due to many factors, viz., competition between fruit-lets (Hirose 1981). Usually, after an extremely heavy crop, alternate bearing can result in tree decline and even collapse in some extreme cases as 'Kinnow' mandarin (Jones *et al.* 1975). Competition for carbohydrates reserves among subsequent crops is probably one of the major cause but results are inconclusive (Monselise and Goldschmidt 1982). There is an inverse relationship reported between fruit size and flower number and fruit number per tree (Guardiola 1992). Consequently, flower and fruit thinning, both manually and chemically, have been used to improve fruit size (Zaragoza *et al.* 1992). Hand thinning requires a lot of labour and time. Whereas, chemical thinning has been desired

strategy for cultivars that bear profusely in order to avoid the effects of alternate bearing (Hirose 1981; Monselise *et al.* 1981; Gallasch 1984). A significant increase in fruit size occurs only if fruit thinning is considerable and performed sufficiently early in fruit development (Zaragoza *et al.* 1992).

The most widely tested growth regulators used for thinning are ethephon (Wheaton 1981, Gallasch 1988), NAA (Hirose 1981, Monselise *et al.* 1981, Wheaton 1981, Gallasch 1988, Guardiola *et al.* 1988). In order to avoid over thinning, treatments should be applied early in the morning or late in the afternoon and only during a period in which excessive temperatures (>30°C) are not common (Hirose 1981). In addition to an increase in fruit size, advantages of thinning also include prevention of tree collapse, which ensures tree survival. In this case, even a loss in immediate profit can be justified (Monselise *et al.* 1981).

Fruit-Set

After successful fertilization the early changes in flower are followed by fruit-set (Petho 1993). The early changes of the flower after a successful fertilization are the signs of fruit set. In some cases, the mere fact of pollination may also initiate the growth of the ovary. Without fertilization, the degeneration of the ovary is expected, which is followed by the death and abscission of the flower too. Flower drop is caused by the appearance of ethylene-produced auto catalytically.

The pollinated flower develop to a fruit, and the fertilized ovules grow to seeds due to an intense synthesis of growth substances. The intense cell division and growth of the tissues absorbs a lot of organic matter of the reserves competing with the vegetative organs, consequently, an interaction between the different parts of the organs of the tree is building up. Not only the fate of the growing fruit, its size and quality but also the physiological potential of the whole tree is influenced by relations of sources and sinks, which in turn may impair the maintenance of the fruits set. The growth substances induce the growth and thickening of the peduncle too (Krezdon 1973, Saleem *et al.* 2008, Jain *et al.* 2014)



Fruit Growth

Fruit size is a desirable character for the marketability of citrus. Small fruit size is very common in mandarins. The variation in fruit size results from differences in cell number or cell size or a combination of both. The potential for increasing fruit size by enhancing cell division early is a relatively undeveloped approach. Treatment with plant growth regulators such as gibberellins and cytokinins during or shortly after flower opening enhances early fruit growth (EI-Otmani *et al.* 1992 ; Guardiola *et al.* 1993) but does not always result in an increase in fruit weight at harvest. This early increase in fruit size is due mainly to a transient increase in cell division in the ovary wall. Early application of auxins increases final fruit size more consistently (Agusti *et al.* 1992; 1996, 1998; EI-Otmani *et al.* 1993; Aznar *et al.* 1995).

The rates of application of the plant growth regulator varies with cultivar, stage of fruit development and climatic region. It is important to ensure optimum tree nutrition and irrigation as well as to provide full tree coverage during application. The mode of action of these plant growth regulators has been summarized by Agusti *et al.* 1996, but it is important to note that they increase vesicle size, not number (Agusti *et al.* 1992, 1996; EI-Otmani *et al.* 1993), and that their effect on vesicle size is through an enhancement of cell enlargement, not cell division (EI-Otmani *et al.* 1993; Agusti *et al.* 1996). Developing fruit are important sinks for water and photo assimilates; their sink strength is increased as a result of auxin treatment, with all fruit tissues increasing in proportion. Auxin application increases fruit peduncle size through an increase in phloem cell size (Agusti *et al.* 1996) consistent with increased sink strength and carbohydrate movement into the fruit. Reduced fruit growth rate during the early stages of fruit development was positively correlated with fruit abscission, particularly when fruit were treated during the June drop period (Agusti *et al.* 1995).

Fruit Drop

According to Soost and Burnett (1961) the exogenous application of GA₃ improves the parthenocarpic fruit

set and growth of self-incompatible genotypes like Clementine that shows negligible parthenocarpic fruit set in the absence of cross-pollination El-Sese (2005) confirmed that the number of fruits per tree and the total yield increased with GA₃ as compared to control. The fruit set was significantly affected by the GA₃ treatment individually and also in combination with auxin and gave the maximum fruit set of 32.3 per cent (Saleem *et al.* 2005). The GA₃ and 2,4-D are used in abscission to control the overloading of fruit for better quality of fruits (Iglesias *et al.* 2007). The application of 2,4-D after fruit set results in acceleration of abscission (Lee 2003). The role of 2,4-D depends on external and internal factors. According to Iglesias *et al.* (2007), decrease in GA and increase the ABA level shows that both these plant growth substances control the initial fruit set and June drop along with other essential component. Randhawa *et al.* (1961) reported that 2,4-D reduced pre-harvest drop in Jaffa oranges. Bajwa *et al.* (1971) reported that in sweet orange cv. pineapple when sprayed with 2,4-D (20ug/ml) or NAA, both plant growth substance control the pre-harvest drop. According to Randhawa *et al.* (1961) application of 2,4-D (15 and 20µg/ml) and 2,4,5-T (5 and 10µg/ml) reduced the fruit drop in Lahore local and Nagpur mandarin.

The environmental conditions like high temperature and low humidity in soil and in air greatly influence the June drop (Levitt 1964; Davies *et al.* 1981). Knapp (1996) reported that the application of 2,4-D isopropyl ester acid at 60 to 70g /ha for during 6 to 8 weeks after bloom reduced the summer drop. Summer fruit drop usually occurs from month of mid-August to October till maturity.

The abscission of fruits is generally coupled with structural changes in the plant (Baird and Webster 1996), which means that several abscission zones are formed within the same inflorescence. It has been postulated that there is appearance of two or three abscission zones around the same fruit. Fruits are abscised because the cells get loose in the abscission zone. Pectin, hemicelluloses and cellulose are dissolved by the respective enzymes, and the mechanical stress detach the fruits. Sometimes, the



dead xylem elements keep the fruit hanging a while. As external agents, the wind may help the drop, but the dehydration of senescent cells causes tension and contributes to the process. The scar of the detached fruit is generally suberised or lignified.

The abscission zone is distinct from the rest of tissues not only in its anatomy but also in metabolic terms. Abscission zone excels by intense cell division, synthesis of proteins and RNA, high O₂ consumption as well as peroxidase-activity. Important role is attributed to the middle lamella and the attached primary cell wall, as being dissolved. The middle lamella is softened by the enzyme pectin-esterase, which demethylate the pectin making it soluble and the cells are easily separated. Another enzyme, cellulase is also activated together with galacturonase.

During the phase of abscission, the genes responsible for inducing the synthesis of hydrolytic enzymes dissolving polysaccharides of the cell wall mainly as cellulases and pectinases are activated. Those enzymes dissolve the middle lamella and also the cell walls and the cohesion of cells is weakened.

Following the flower drop, fruit drop ensuing before maturity is attributed to the collapse of the hormonal balance in the growing fruits, where the growth substances being active in favour of growth lost their influence against abscisic acid causing abscission. The role of ABA becomes prevalent when the young fruit-lets drop, and, when fruits are almost ripe. The difference between varieties prone to fruit drop corresponded to their ABA content too.

The abscission of young fruits seems to depend on auxin as a correlative dominance signal (Bangerth 1990). The investigations of Luckwill (1953) made early reports in searching the causal relations between auxin production of the developing fruit and fruit drop. They explained that IAA may speed up or alternatively inhibit the process of abscission.

Lack of nutrients, which does not depend solely on the meagre soil, but rather on the competition between the vegetative organs and the growing fruits of the plant. The fate of a young fruit is often impaired by the dominance of another fruit or vigorous shoots.

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Gibberellins are known for their ability to increase cell enlargement, thus enhancing fruit growth in certain species such as citrus (Eman *et al.* 2007, El-Sese 2005), guava (El-Sharkawy and Mehaisen 2005) and pear. In all species so far studied, gibberellins had the potential for increasing fruit size. Salicylic acid is considered to be a potent plant hormone (Raskin 1992) because of its diverse regulatory roles in plant growth regulator and its role is evident in fruit yield (Klessig and Malamy 1994). Application of plant growth regulators like gibberellins and salicylic acid alone or coupled with micronutrients may improve cropping potential.

Early reproductive processes in citrus are strongly affected by plant growth regulators indicating that the regulatory mechanisms that control fruit set and abscission of ovaries and fruitlets possesses a pivotal hormonal component (Talon *et al.* 1990). Gibberellins and cytokinins are generally considered to be positive regulators of fruit growth while auxins have been reported to act as stimulators of growth and also as abscission agents (El-Otmani and Oubahou 1996). Abscisic acid (ABA) and ethylene have been implicated in several ways in abscission. Gibberellins are thought to be pivotal effectors responsible for the ovary-fruit transition (Talon *et al.* 1992, Ben-Cheikh *et al.* 1997). They activate cell division and cell enlargement processes in vegetative organs (Talon *et al.* 1991) and therefore are generally associated with the initiation of growth (Talon and Zeevaart 1992). The intensity of abscission during the initial phases of growth is also related to the phenology of flowering.

Salicylic acid and other salicylates are known to affect various physiological and biochemical activities of plants and may play a key role in regulating their growth and productivity (Arberg 1981). Exogenous application of salicylic acid may also influence seed germination and fruit yield. Sivakumar *et al.* (2002) determined that the application of salicylic



acid increased the content of protein in grains of pearl millet. Kalarani *et al.* (2002) carried out a pot culture experiment to determine the effect of foliar application of different concentrations of salicylic acid on physiological and biochemical constituents as well as yield and quality of tomato and found that Salicylic acid at all concentrations showed its efficiency in inducing early flowering and increased fruit set percentage.

Besides, hormonal induction of fruit growth, nutrients may have regulatory functions on their own and/or through the maintenance of adequate hormonal levels (Gillaspy *et al.* 1993). The average number of flowers produced in a normal citrus tree is by far extremely high in comparison to the number of fruits that the same tree can support until ripening. Hence, many fruits are abscised during growth apparently due to competition for nutrients especially photo-assimilates. During the initial moments of phase I, citrus fruitlets function as carbohydrate utilization sinks but over the final stages of this period and during the transition from cell division to cell enlargement, developing fruits shift their metabolism and start to behave as storage sinks (Mehouachi *et al.* 1995). Defoliation during phase I reduces carbohydrate amounts, arrests fruitlet growth and promotes massive abscission (Mehouachi *et al.* 2000) whereas, defoliation after the June drop also arrests growth but does not induce abscission (Lenz 1967). The link between carbohydrates and fruit growth is currently supported by a wide body of evidence including several studies on source-sink imbalances, defoliation, girdling, shading, sucrose supplementation, de-fruiting and fruit thinning (Goldschmidt and Koch 1996, Iglesias *et al.* 2003, Syvertsen *et al.* 2003). First, the enhancement of carbohydrate availability was associated with an improvement of fruit set and yield of citrus trees (Goldschmidt 1999). Later, a strong relationship was demonstrated between carbohydrate levels available to fruitlets and the probability of abscission (Gomez-Cadenas *et al.* 2000, Iglesias *et al.* 2003). This phenomenon that has also been described for other tree species is also supported by studies on translocation of ^{14}C metabolites and CO_2 -enrichment

experiments (Moss *et al.* 1972, Downton *et al.* 1987). Hence, photosynthesis activity has been proved to be crucial since high carbohydrate requirements during fruit set increases photosynthetic rate (Iglesias *et al.* 2002). This suggestion also implies that a reduction in net CO_2 assimilation results in lower sugar production and fruit set. The sugar concentration in leaves might be the signal that regulates the feedback mechanism stimulating photosynthesis in response to fruit sugar demand. Thus, once carbon demands are fulfilled, carbohydrate accumulation may elicit end-product feedback control of photosynthesis. The positive effect of exogenous GAs on fruit set and growth may also partially operate through the induction of a stronger mobilization of ^{14}C metabolites to ovaries (Powell and Krezdorn 1977). The exogenous GAs have also been shown to stimulate growth and increase carbon supply in vegetative tissues (Mehouachi *et al.* 1996). Thus the sugars are deeply implicated in the regulation of fruitlet growth and that overall carbon deficiency induces fruit abscission.

Although the specific mechanism involved in the response of fruit growth to carbohydrates has not been studied at the molecular level, observations suggest that sugars may act not only as essential nutrient factors but also as signals triggering specific hormonal responses (Roitsch 1999). The observation linking carbohydrate and abscission was confirmed with the finding that carbon shortage during ovary and fruitlet drop increased ABA and ethylene and both are involved in the induction of early abscission (Gomez *et al.* 2000).

The alterations in the nutrient balance that are accompanied with increased fruitlet abscission during the June drop provoke an unambiguous tendency to both increase nitrogen content and to reduce carbon shortage. Abscission intensity may be correlated positively with carbohydrate shortage. The two main Conclusions that can be extracted are that the fruit fall that takes place during June is very likely due to the carbohydrate insufficiency caused by an increased carbon demand of a huge population of expanding fruitlets; and second,



carbon deficiency is again associated with ABA rise, ethylene release and massive fruitlet abscission. This idea that citrus fruit abscission is connected to carbohydrate availability was initially anticipated by Goldschmidt and Monselise (1977) who suggested that citrus might possess an internal self-regulatory mechanism that adjusts fruit load to the ability of the tree to supply metabolites. The above findings identify leaf sugar content, ABA and ethylene as major components of the self-regulatory adjusting mechanism of abscission. Recently, it has also been proposed that in addition to June drop, earliest ovary and fruitlet falls that occur through abscission zone, are also dependent upon nutritional factors such as sugars (Iglesias *et al.* 2006).

Literature indicates that the application of nutrients and plant growth regulators increase the fruit yield and quality, hence the suitable combination of macronutrients, micronutrients and growth regulators could control the excessive fruit drop and improve the citrus fruit yield and its quality.

Fruit Quality

Fruit quality is a concept that varies according to the final use of the fruit and at what point from orchard to consumer the fruit is evaluated. For the grower, any fruit that can be sold at a reasonably good price is of good quality. For the fruit packing industry, fruits that are of uniform size, free of blemishes offer good market. The harvested fresh fruit requires to be seedless (low seed number), high TSS:Acid ratio and excellent color, shape and firmness. Whereas, the requirement of the juice industry is fruits with high juice and sugar content. Parameters important to both the fresh fruit and juice industries have been shown to respond to plant growth regulators.

The juice content and organoleptic taste are important parameters and desirable for fresh fruit consumption of citrus. Organoleptic quality is the result of sugar and acid content, and the presence of volatiles in the juice. Nutritional quality includes sugar, acid, vitamins, etc. For fruit processing, internal quality is far more important than the external appearance of the fruit. Plant growth regulators that can be used

to manipulate these parameters in citrus are almost non-existent. Although not a plant growth regulator, lead arsenate was used for many years in Florida to improve fruit internal quality (Knapp 1996). This compound causes a reduction in total acidity and consequently, an increase in the sugar-to-acid ratio.

Granulation of fruit is a physiological condition in which the juice sacs become gelled with little extractable juice. It develops pre-harvest (Smoot *et al.* 1971) as well as post-harvest (Gilfillan and Stevenson 1977). A reduction of the disorder by GA₃ sprays has been reported, but this effect has not been reproduced consistently.

According to Kaur *et al.* (2000) fruit weight increased with increase in amount of 2,4-D in trees of Kinnow mandarin. The fruit weight and peel thickness has been increased with the application of Zn alone or in combination with GA₃ in 'Washington Navel' orange (Emen *et al.* 2007). The application of Zn and K increased fruit weight as compared with control. According to Sourour (2000) both Zn and EDTA increased the number and weight of fruits per tree. According to Chundawat and Randhawa (1972) GA increased the fruit size in Saharanpur special grapefruit trees. The application of 2,4-D at 20 µg/ml along with CuSO₄ at 0.25 or 0.50 percent increased the fruit size (Singh and Mishra 1986).

Chundawat and Randhawa (1972) reported that GA₃ and 2,4-D increased peel thickness in Saharanpur special variety of grapefruit. Also Dinar *et al.* (1977) observed that both gibberellic acid and 2,4-D increased peel thickness in Marsh grapefruit. Application of nutrients increased the juice content (Ram and Bose 2000). Maximum juice from fruits was observed from the trees sprayed with 1% urea and 0.8% Zinc sulphate (Malik *et al.* 2000). Trees treated with gibberillic acid yield juice more than 10% than the control ones (Davies *et al.* 2001). Such increase was economically beneficial to Florida citrus growers because processed fruit value increase with juice yield (Braddock 1999).

The application of Zn alone and with combination with Fe and Mn increased the TSS value as compared



to control. According to Monga and Josan (2000) the highest TSS was obtained from tree sprayed with Zn (0.3%) alone. Treatment with 2,4-D at 20 µg/ml and gibberllic acid at 20 µg/ml increased the TSS in grapefruit juice (Llanes *et al.* 1991).

Foliar application of Zn alone and along with Fe and Mn on Kinnow mandarin resulted in decrease acidity as compared to control (Monga and Josan 2000). According to Singh and Mishra (1986) 2,4-D increased the acidity level in Kinnow fruit. Age of tree and type of cultivar also influenced the acidity of oranges (Frometa and Echazabal, 1988).

Foliar application of Zn was observed to increase the ascorbic acid contents of juice in various citrus varieties (Dawood *et al.* 2001). Increased content of ascorbic acid was observed with application of Zn alone, Zn+Mn or Zn+B (Tariq *et al.* 2007). High ascorbic acid was obtained by sprays of Zinc and gibberellic acid (Eman *et al.* 2007). Chundawat and Randhawa (1973) reported that vitamin C content increased with spray of 2,4-D in Duncan cultivar of grapefruit. Immature citrus fruits have the highest amount of ascorbic acid whereas, ripened fruit have the least as reported by Nagy (1980). However, ascorbic acid content increased with ripening of fruits in apricot, peach and papayas but decreased in apples and mangoes (Lee and Kader 2000).

The TSS:Acid ratio of juice was increased with the application of SA, Zn and K alone and also by combination of both. Abd-Allah (2006) reported that the application of K in combination with micronutrients improved TSS:acid ratio while Zn+K and other different combinations was observed by Ashraf *et al.* (2012).

Among sugar derivatives; sugar nucleotides, sugar phosphates, glycosides and polyols are important. D-galacturonic acid, D-Glucuronic acid, L-ascorbic acid are sugar acids were found in citrus fruits. Sugar alcohols such as myo-inositol was found in oranges and grapefruits in the range of 88-170 mg/100g of juice. Lemons had 56-76 mg myo-inositol/100g of juice. The reducing sugars, non-reducing sugars and total sugars increase as the fruit start to ripe.

Fructose, alpha and beta -glucose and small amount of galactose were also found in Valencia orange juice. By using ¹⁴C labelled compounds, Sawamura and Osajima (1973) observed that translocation of sugars from leaf to fruit occurs in the form of glucose and fructose which are further changed into sucrose in the fruit. Kuraoka *et al.* (1976) found changes in sugar content of flavedo tissue of Satsuma mandarins grown in Japan. Ting and Altaway (1971) reported that the ratio of Fructose : Glucose : Sucrose as 1:1:2 in Valencia oranges.

Production

Optimum yield is a yield that is sustainable year after year and that utilizes the full potential of the available land along with the energy and nutritional resources of the tree. This yield is a function of tree planting density, canopy development, intensity of flowering, fruit set, fruit growth and number of fruits harvested at maturity. In young trees, a high proportion of photosynthate is allocated to vegetative development and growth. Usually, only a few flowers and fruits are produced during the 2 to 3 years after planting a nursery tree. Canopy development and growth continues and reaches an optimum between 5 and 12 years, depending on planting density, at which time yield is at its highest value (Boswell *et al.* 1970 1975). At this point a natural equilibrium between vegetative growth and reproductive development is established with variations due to environmental conditions and genotype of the cultivar, provided that nutrition and light interception are optimal.

The production per unit area is the key factor for the citriculturists. However, the productivity depends upon factors viz., light interception, carbon dioxide fixation, water availability, and mineral nutrient uptake. Besides, the productivity is also affected by competition among similar or different plant organs for photo assimilates and nutrients. Plant growth regulators have been used to improve productivity wherever feasible. Unfavourable environmental conditions may restrict many of the processes related to production from flower initiation (Moss 1969; Southwick and Davenport 1986; Lovatt *et*



al. 1988) to the development of inflorescences and flowers or may cause excessive drop of flower buds, open flowers, entire inflorescences, and developing fruitlets. Plant pathogens may cause excessive fruit drop and may deteriorate fruit quality (Lima *et al.* 1980). Imbalance of fertilizers that may supply excess of nitrogen lead to excessive vegetative growth and low flower number. On the other hand, nitrogen deficiency leads to low flower initiation and thus reduced yields (Davenport 1990).

Plant growth regulators are used to reduce the seasonal fluctuations in yield and also to maximize energy allocation to harvestable fruit in order to increase fruit number and fruit size rather than to enhance excessive vegetative growth. Plant growth regulators can be used to hasten or delay fruit coloring and fruit maturation so as to shorten or extend the harvest season to get the best economic productivity. (Jain *et al.* 2014)

Yield (Kg per tree) includes both fruit number and fruit size. Fruit number is a function of flower intensity and fruit set. Fruit size is a function of cell division and cell enlargement processes. The number of fruit that set and persist to harvest influences fruit size. Citrus fruit growth is a continuous process, fruit weight and fruit diameter is represented by a single sigmoidal growth curve (Guardiola and Lazaro 1987). The development of the ovary into a mature fruit proceeds through three stages: cell division, cell enlargement, and maturation. During Stage I, cell division predominates. Cell division is essential for the formation of the ovary during flower initiation and continues past petal fall to approximately mid-June to late July (Lovatt 1999). At this time cell division, which occurs in all the tissues, is completed except in the flavedo. Thus, during the period encompassing fruit set and June drop, fruit growth is dominated by cell division. Stage II is the period of maximum fruit growth occurring over the following 3 months for the early maturing cultivars (e.g., 'Satsuma' mandarin) and the next 7 to 10 months for the late maturing cultivars (e.g., 'Valencia' orange). This stage of fruit development is dominated by cell differentiation and cell enlargement. The juice

vesicles and locules increase in size with the uptake of water, and the flavedo continues cell division, whereas the cells of the albedo must expand and stretch to accommodate the growth of the locules. Dry matter and water accumulate in the vacuoles at a high rate (Guardiola 1992). Fruit that persist on the tree through the June drop period are not likely to abscise in the absence of pest damage, physiological disorders such as splitting, or stress such as water deficit and excess temperatures. Stage III is the period of fruit maturation. Fruit continue to increase in size predominately by cell expansion, but at a slower rate. Peel thickness may increase in some cultivars and puffiness may occur, particularly in the mandarin types.

According to Bengal *et al.* (1982), the combined application of NAA (25ug/ml) and urea (1%) increased the seed yield. Foliar application of NAA on paddy (*Oryza sativa* L.) under low level of N (0 and 60 kg/ha) gave the beneficial effects as reported by Grewal and Gill (1986). The beneficial effect was observed by increase in number of ear-bearing shoots/plant, grain weight, number of filled grains/panicle. The sufficient nitrogen supply was essential for increasing the yield of rye and barley by mono ethanolamine (Bergmann and Eckert 1990).

Conclusion

Plant growth regulators are commercially exploited in the field of horticulture because of the wide range of potential roles they play in increase of productivity per unit area. Their application in citrus industry during different phases of growth and development is currently in practice as is evident from the present literature. The advantage of plant growth regulators is their use at very low concentration because of which they do not lay any health hazards. However, it is important to understand the basic mechanisms underlying the citrus growth and development in order to manipulate the key physiological processes and make use of the plant growth regulators at the appropriate stage of development and at the optimum dose. Therefore it is desirable to pursue research on the mechanism of action of plant growth



regulators on the physiological, biochemical and genetic regulation of growth and development.

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