

CHAPTER 6

Plant-Growth Regulators

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Living organisms are complex structural and biochemical entities. Why a particular species differs from others and how a complex organism arises from a single cell is not clearly understood. It is clear that a high degree of organization of growth processes must exist to account for the orderly development of plants. A few organic compounds that influence certain biochemical and physiological reactions have been isolated from the wide array of organic and inorganic compounds found in plants. A growing body of evidence suggests such compounds are of general occurrence in plants, active at exceedingly low concentrations, and under genetic control. These compounds, which act as chemical directors of growth processes, are known as plant-growth substances or plant-growth regulators.

Although many early workers suggested the existence of chemical control mechanisms in plants, it was not until the twentieth century that observations made by Darwin (1896) led to the isolation of a chemical messenger by F. W. Went (1928). A detailed historical account of plant-regulator research since Darwin's time may be obtained from Skoog (1951), Tukey (1954), Audus (1959), Leopold (1964), and van Overbeek (1966). In recent years, considerable research has been devoted to both naturally occurring and synthetically produced plant-growth substances. Numerous compounds have been investigated, and significant progress has been made in both fundamental and applied research. These compounds, already important in agriculture, are destined to become increasingly valuable as more knowledge of their physiological influence is acquired.

Several naturally occurring plant-growth substances have been isolated from citrus tissues. Auxins were the first endogenous substances to receive attention. Gustafson (1939) found that extracts from flower bud ovaries of parthenocarpic varieties of oranges, lemons, and grapes contained higher auxin levels than ovaries from corresponding varieties that do not produce fruits parthenocarpically. Cameron, Cole, and Nauer (1960) reported a positive correlation between seed number and fruit size for the Valencia orange and trends toward positive correlations in four other citrus varieties. This and other similar evidence led to speculation that seeds directly or indirectly stimulate fruit growth through the action of plant-growth regulators. Monselise (1945) obtained auxin from the growing shoots of Shamouti orange. Stowe, Thimann, and Kefford (1956) reported that orange blossoms contain indole, indoleacetic acid (IAA), and an unknown growth inhibitor. Khalifah, Lewis, and Coggins (1963) found evidence for an auxin in young citrus fruits that does not appear to be an indole. This new auxin also has been isolated in fruits of date, fig, peach, and Natal plum, but no IAA was found in these fruits (Lewis, Khalifah, and Coggins, 1965). Fruits of guava and strawberry contained IAA but not the new auxin. This recent research raises the question of whether the new auxin and IAA have been confused with each other in the past.

Kawarada and Sumiki (1959) found gibberellin A_1 in the elongated water sprouts of *Citrus unshiu* Marc. Khalifah *et al.* (1965) reported evidence for the occurrence of three different gibberellin-like substances in orange and lemon fruits. Two of these compounds appear to be gibberellins A_1 and A_9 . Growth-stimulating activity also has been reported from lemon and orange oil and is believed to be due in part to the presence of lauric, linolenic, and linoleic acids (Haagen-Smit and Viglierchio, 1955). Germination- and growth-inhibiting coumarins have been isolated from citrus tissues (Khastagir, 1947).

In the citrus industry, plant-growth substances have proven effective in stimulating a number of desired responses, such as decreasing fruit drop losses, increasing fruit size, improving fruit set, and delaying fruit maturity. Differences in varieties and cultural practices make almost any physiological response useful somewhere in the industry, but to be successful a response must be dependable and reasonably free of undesirable effects. This chapter discusses many of the responses that have a history of practical success and others whose applications are in the preliminary to advanced stages. Plant-regulator research in the future can be expected to discover many more new uses for plant-growth substances.

FRUIT ABSCISSION

Citrus fruits have no well-defined physiological maturity stage. Harvest of oranges and grapefruit in California begins when the fruits are mature enough to be eaten and usually ends four to six months later. Abscission causes serious fruit loss, especially during the latter part of the harvest season.

The first reported citrus research on the use of plant-growth regulators to decrease preharvest abscission came from Florida (Gardner, 1941). Naphthaleneacetamide applied to Pineapple oranges early in the harvest season at a concentration of 100 parts per million (ppm) reduced fruit drop for twelve weeks or longer. A concentration of 10 ppm was ineffective, and even 100 ppm was not effective if applied after preharvest abscission had become a problem. The use of naphthaleneacetamide has not developed into a commercial practice.

In California, Stewart and Klotz (1947) applied 2,4-dichlorophenoxyacetic acid (2,4-D) to Valencia orange trees after the start of preharvest fruit abscission and obtained significant decreases in fruit losses. The literature since 1947 contains numerous reports of mature fruit abscission being reduced by 2,4-D and related compounds. Studies in California indicated that 2,4-D is also effective on navel oranges (Stewart, Klotz, and Hield, 1947), grapefruit (Stewart and Parker, 1947), and lemons (Stewart and Hield, 1950). Other reports could be cited, but the important point is that 2,4-D is being used successfully in California to reduce preharvest fruit abscission ([table 6-1](#)). Reports indicate that the same response generally has been obtained in Australia (McAlpin and Merrett, 1949), South Africa (DeVilliers, 1955), Japan (Iwasaki, Nishiura, and Shichijo, 1956), China (Hwang, 1958), and India (Randhawa, Sharma, and Jain, 1961). Results in Florida have been less successful than in California, but 2,4-D has proven effective on Pineapple seedling orange and Temple orange (Gardner, 1951). When applied to other varieties of citrus in Florida, 2,4-D has been ineffective.

Often it is desirable to apply 2,4-D with nutritional or pesticidal sprays. In general, 2,4-D is compatible with all of these sprays in common use in California (Hield and Erickson, 1962). However, Halse (1954) reported that Bordeaux mixture impaired the effectiveness of 2,4-D in preventing fruit abscission. When 2,4-D esters are combined with insecticidal oils, the effectiveness of 2,4-D is increased and the dosage of the regulator must be decreased to avoid plant injury. Fall applications of whitewash are frequent in the San Joaquin Valley of California as an insecticidal treatment. When 2,4-D is incorporated in whitewash, the concentration of the plant-growth regulator must be increased to obtain satisfactory control of abscission (Hield and Erickson, 1962).

Two other compounds, 2,4,5-trichlorophenoxyacetic acid (2,4,5-T) and 2,4,5-trichlorophenoxypropionic acid (2,4,5-TP) have also proven effective against abscission. In some instances, 2,4,5-T is more effective than 2,4-D (Stewart and Hield, 1950). After Sites (1954) reported that preharvest abscission of Pineapple orange in Florida was reduced by 2,4,5-TP, the compound was tried in California by the junior author in the hope that leaf malformations, which sometimes result from 2,4-D, would not occur. Under California conditions, higher concentrations of 2,4,5-TP proved necessary to obtain abscission control. Once the concentration was high enough to provide control comparable to that obtained with 2,4-D, leaf malformations resulted.

Most leaf distortions can be avoided by applying regulator compounds when young growing leaves are not present. Typical leaf distortions caused by 2,4-D are shown in [figure 6-1](#). In general, the high volatile ester formulations, such as isopropyl ester, have proven less injurious than low volatile ester formulations. Since 2,4-D applied to green-colored oranges delays the loss of green rind pigments, application of the compound usually is avoided in groves that will be harvested early in the season.

In addition to its effectiveness in control of preharvest abscission, 2,4-D reduces the amount of fruit stem dieback that occurs late in the harvest season (Stewart *et al.*, 1947; Hield and Erickson, 1962). This response, probably caused by the same physiological mechanism that delays abscission, results in a more turgid fruit.

Pesticide oil sprays occasionally enhance leaf and fruit drop. Oil-induced abscission can be reduced by including 2,4-D in the spray mixture (Stewart and Ebeling, 1946; Stewart, Riehl, and Erickson, 1952). Day and Erickson (1954) reported that 2,4-D, 2,4,5-T, and 2,3,4-trichlorophenoxyacetic acid are about equally effective in preventing ethylene-induced abscission of lemon leaves. Moderate control was obtained from 2,3,5-trichlorophenoxyacetic acid. Since the latter two compounds are not very active with respect to leaf malformation, it was suggested that they may be used for abscission control on plants which cannot tolerate 2,4-D and 2,4,5-T.

In both California and Florida, a vigorous research effort is being devoted to a search for compounds that will promote citrus fruit abscission as an aid to mechanical harvesting. Hendershott (1964) obtained evidence that inhibitors of sulphhydryl enzymes are more effective in inducing abscission layer formation than those which inhibit dehydrogenase systems or act as uncoupling agents. Wilson (1966) demonstrated that abscission stimulated by iodoacetic acid cannot be morphologically distinguished from normal abscission. The physiological mechanism by which iodoacetic acid produces abscission is not clear. Cooper and Henry

(1967) presented a preliminary report showing that field sprays of ascorbic acid can cause a loosening of mature citrus fruit without appreciable drop of leaves or small fruit. This treatment may have limited commercial application, since it causes pitting on the rind of mature fruit.

FRUIT SIZE

When small citrus fruits are treated with high concentrations of 2,4-D, many of them become misshapen (Stewart and Klotz, 1947). Treated navel oranges may develop thick rinds and protruding navels or become cylindrical in shape. Small seedlike structures, mainly enlarged seed coats, develop in navel oranges if high concentrations are applied to small fruits. In addition to this response in the seedless navel orange, similar morphological aberrations have resulted from 2,4-D applications to Valencia orange fruits. Applications of 2,4-D to grapefruit trees have resulted in cylindrical fruits with navel structures, thick, coarse rinds with excessively large oil glands, and fruits with dry, hard juice vesicles (Stewart and Parker, 1947). High concentrations of 2,4,5-T have caused lemon fruits to develop coarse rinds (Erickson and Brannaman, 1950a). The same type of response occurs from heavy applications of 2,4-D.

The desirable response from the application of 2,4-D to young fruits is an increase in fruit size. When appropriate concentrations are used (Hield and Erickson, 1962), undesirable side effects cease to occur, with the exception of moderate increases in rind thickness and roughness. The increase in fruit size can be used to economic advantage when a size problem exists if the orchard in question does not normally produce fruits with thick, coarse rinds. A size increase can be obtained in California with navel orange, Valencia orange, grapefruit, lemon, and lime. Some typical results after 2,4-D treatment are shown in [table 6-2](#). Experience with limes is limited (Erickson and Brannaman, 1950b), but small fruits are rarely a problem in lime culture. A better fruit size response is obtained with 2,4,5-T than with 2,4-D on lemons and grapefruit (Erickson and Haas, 1956; Hield and Stewart, 1956).

Application of 2,4-D and 2,4,5-T regulators to small fruits causes a delay in loss of green rind pigments, which may indicate that treated fruits are physiologically younger than those from unsprayed trees. This apparent delay in maturation may account for some of the size increase as well as reduced abscission of mature fruits. Additional factors that may contribute to increased size are a thicker fruit stem in proportion to fruit diameter, stimulated growth rate of certain tissues, and a thinning effect or reduced numbers of fruits per tree (Stewart, Klotz, and Hield, 1951; Stewart *et al.*, 1952). The effectiveness of 2,4-D as a "sizing" spray is related to stage of fruit growth. As fruits become larger, they are less responsive to 2,4-D. It should be stressed that 2,4-D does not replace the need for optimum cultural practices. For example, 2,4-D will not overcome the size problem resulting from an inadequate supply of moisture to trees (Erickson and Richards, 1955).

A significant reduction in splitting at the navel end of Washington navel oranges also has resulted from 2,4-D treatments applied when fruits are small. In addition, the onset of granulation has been delayed in Valencia oranges by applying 2,4-D to small fruits.

IMPROVING FRUIT SET

A means of obtaining improved fruit set in citrus would prove of enormous commercial benefit, and plant-growth regulators may eventually provide the means of bringing about such responses. No appreciable success has been achieved thus far in improving fruit set of citrus with these compounds, but research results have been promising on a wide range of other plants.

Although delayed abscission of flowers and young fruits has resulted from 2,4-D applications to orange trees (Stewart and Klotz, 1947; Stewart *et al.*, 1951), the end result was a decrease in number of fruits reaching maturity. Such a thinning effect has been observed many times in California and on Valencia oranges in South Africa. A notable exception has been reported in South Africa, where number of fruits retained by navel orange trees were increased by 2,4-D (DeVilliers, 1955). Data from a number of experiments leave no doubt that increased volume of navel oranges from 2,4-D-treated trees in South Africa is both the result of increased numbers of fruit and increased fruit size. The reason for the response is not known, although climatic and soil moisture conditions prevailing during the flowering and fruit set period for navels in South Africa appear to be more adverse than in California.

Naphthaleneacetic acid (NAA), indoleacetic acid (IAA), and indolebutyric acid (IBA) failed to increase fruit retention in Washington navel orange and Marsh grapefruit (Pomeroy and Aldrich, 1943). Many other compounds have been tested in preliminary experiments, and all except gibberellic acid (GA) have failed to give the desired results.

In California, application of GA to small Bearss lime fruits (3/8-inch or 9.5-mm diameter), Eureka lemon

flowers, and branches of Washington navel orange trees during flowering resulted in an increase in the number of fruits (Hield, Coggins, and Garber, 1958). No influence was detected on shape, size, color, or fruit quality. In navel oranges, an increase in small seedlike structures occurred which is similar to the effect caused by applications of 2,4-D to small fruits. In Japan, improved retention of navel orange fruits was reported when GA was applied by hand to individual organs during the flowering or young fruit period (Iwasaki *et al.*, 1962). Set of Clementine mandarin, a self-incompatible variety, was induced by GA application to flowers when pollen was excluded (Soost, 1958). Compared with Clementine fruits from pollinated flowers, treated fruits were smaller, more necked, elongated, and seedless. Increased fruiting was obtained when shoots and flowers of Orlando tangelo, Dream navel orange, and Valencia orange were dipped in GA solutions (Kretdorn and Cohen, 1962). With the Orlando tangelo, the number of fruits on GA-treated shoots was about equal to that obtained with Temple tangor crosses and superior to that obtained with open-pollinated shoots. Slight fruit malformations and splitting occurred with Dream navel and Valencia orange. Increased fruit retention was observed when Pineapple orange shoots with small fruits were dipped in GA (Kretdorn and Cohen, 1962). Green fruits, thicker peels, and some splitting were associated with this treatment.

Because of the need for treatments that will increase fruit set, detailed experiments have been conducted in California with GA on navel orange, Valencia orange, and Clementine mandarin. After navel orange trees were sprayed with relatively high concentrations of GA during the flowering period, drastic injury and reduced production occurred (Hield, Coggins, and Garber, 1965). Injury was expressed in leaf drop and death of new vegetative and flower-producing shoots. GA concentrations low enough not to cause injury produced no increase in fruit set. Although considerable leaf drop occurred, an increase in fruit retention was obtained when Valencia orange trees were sprayed with high levels of GA (Coggins, Hield, and Garber, 1960). Treatments were applied when the trees were past full bloom, with small fruit of up to 3/8-inch diameter. Increased fruit set and production resulted from GA applications to low-yielding Clementine mandarin trees, but production was not improved under conditions of adequate cross-pollination (Soost and Burnett, 1961). Fruit produced by GA-treated Clementine mandarin trees were small and coloring was delayed. Delayed coloring and certain juice quality effects suggested that the treatment resulted in delayed maturity.

In Florida, studies have been conducted in which GA was sprayed on entire trees (Kretdorn and Cohen, 1962). Late bloom sprays of GA on Orlando tangelo increased yield and number of fruits. Leaf drop, fruit splitting, and coarse, green fruits borne on thick, stiff stems resulted in some cases. Many fruits were extremely small. Treated fruits remained green longer, but were well colored at harvest. More recently, Kretdorn (1968, unpublished) showed a substantial increase in fruit set on Orlando tangelo with applications of 2.5 to 10 ppm of GA during bloom. Only a slight reduction in fruit size occurred, and no other adverse effects were noted. No increased fruit set was obtained when Dream navel and Valencia orange were sprayed with GA (Kretdorn and Cohen, 1962).

In most cases, the potential advantage of improved fruit set through use of GA has been overshadowed by undesirable responses. Nevertheless, the fact that GA can increase fruit set in California and Florida, where other plant-growth regulators have failed, indicates a potential for the future.

BIENNIAL BEARING AND THINNING

With many perennial plants, the production of alternate heavy and light crops is a problem. One undesirable feature of the biennial production pattern is the inferior size of fruits during the heavy crop year. Naphthaleneacetic acid (NAA) has been widely employed as a thinning agent in apples and to some extent in peaches, pears, and olives. Reduction of the number of fruits leads to an increase in average size and also reduces biennial bearing.

Certain citrus varieties are especially prone to production of alternate heavy and light crops. Under California conditions, the Wilking mandarin exhibits an extreme cycle of a heavy crop during the "on" year followed by essentially no flowers nor crop during the "off" year. During heavy crop years, fruits are exceedingly small, and the total crop load is sometimes so heavy that major tree branches are broken. NAA sprays to several citrus varieties during bloom showed no influence on fruit set (Pomeroy and Aldrich, 1943; Stewart *et al.*, 1952), but applications to Wilking mandarin at about the normal period of "June drop" produced a thinning effect and improved fruit size (Hield, Burns, and Coggins, 1962). Valencia orange, navel orange, and a number of mandarin varieties have been thinned with NAA applied near the normal period of "June drop." At present, we are unable to obtain the proper amount of thinning with the compound, but it appears to have no undesirable influence on external or internal fruit quality. To have an appreciable influence on the alternate crop year, it appears that the thinning effect must be of sufficient magnitude to

reduce the volume of fruit produced. With the resulting increase in fruit size, it is possible to greatly decrease the number of fruits without altering the volume produced by the tree. This is illustrated by the data on Wilking mandarin studies presented in [table 6-3](#).

Studies in Florida indicated that maleic hydrazide (MH) is an effective blossom-thinning agent for Dancy tangerines (Gardner, Reece, and Horanic, 1961). No thinning was obtained when the compound was applied after petal fall. Unlike the results obtained with NAA, no increase in fruit size was associated with thinning. A reduction in fruit size, suppressed development of juice vesicles, and increased rind thickness resulted from applications of MH to Washington navel orange trees (Erickson *et al.*, 1952). No such fruit quality effects were reported on Dancy tangerines. Whether this reflects differences in susceptibility, differences in penetration, or an interaction with environmental factors is not known. Hendershott (1962a) reported that the rate of absorption of MH by orange leaves is faster in a relative humidity of 90 to 95 per cent than at 60 to 70 per cent.

FRUIT MATURITY

Plant-growth regulators are a potential means of regulating various aspects of fruit maturity. The development of methods for bringing about early fruit maturity or delaying fruit maturation could prove of considerable advantage for numerous citrus varieties grown under a broad range of climatic conditions and subject to various marketing demands.

It has been demonstrated that GA delays loss of green rind pigments by navel orange (Coggins and Hield, 1958), Valencia orange (Coggins, Hield, and Garber, 1960), grapefruit (Coggins, Hield, and Burns, 1962), tangerine (Soost and Burnett, 1961), lemon (Coggins, Hield, and Boswell, 1960), lime (Burns *et al.*, 1964), and satsuma mandarin (Nishiura and Iba, 1964). High concentrations of GA applied to Lisbon lemon trees in the spring caused a significant delay in fruit maturity as well as a number of undesirable responses. Production during the treatment year was reduced, apparently as a result of increased fruit drop. Fruit drop and the percentage of harvested lemons without buttons were increased. Fruit stems showed an increase in diameter, a response also reported for 2,4-D. After a very sparse bloom in treated plots, production was drastically reduced the next year. The desirable feature of the treatment, a delay in yellowing, was marked: lemons from treated trees were greener as early as one month and for as long as seven months after treatment. Fruits treated with GA also appeared to develop a lemon-yellow color less rapidly in storage. Ultimate fruit size was not reduced by GA, but fruits of 3.6 to 4.7 cm in diameter at treatment time grew more slowly than untreated fruits. Growth rate of fruits that were larger or smaller at treatment time appeared to be normal.

Delayed yellowing has been induced by GA applied to lemon trees at any time of the year. The response reflects an over-all delay in maturity rather than a simple delay in rind maturity as appears to be the case with navel orange. Very few undesirable responses, if any, occur when relatively low concentrations of GA are employed. The authors have found that the over-all influence of an appropriate dosage applied in the fall appears to be beneficial and of economic value. In addition to the first-year effects on fruit color ([table 6-4](#) and [fig. 6-2](#)), fruit size ([table 6-5](#)), and seasonal harvest patterns ([table 6-6](#)), there is a change in harvest pattern in the second year ([table 6-7](#)). Probably this results from the influence of GA on flowering, which has been observed in numerous trials. The volume of fruit produced early in the season is reduced, but treated trees produce more fruit during summer when the market demand is high. If GA is applied in two successive years, the seasonal harvest pattern is altered even more, since both direct and indirect effects are operative ([table 6-7](#)).

The primary benefits of GA treatment are a more desirable seasonal harvest pattern in relation to market demands, a larger percentage of fruit with a long storage life, and a decrease in the number of small yellow lemons. These effects permit more flexibility in harvesting and marketing. Since the California lemon industry is based on the sale of fresh fruit and peak production normally occurs when demand is low, these findings have important economic implications. Gibberellic acid is registered (see [below](#)) and recommended for use on lemons in California (Coggins *et al.*, 1964). Similar responses have been obtained from GA treatment of limes (Bums *et al.*, 1964).

Erickson and Haas (1956) showed that some delay in fruit maturity results from application of 2,4,5-T to lemon trees, although it appears to be minor compared to that produced by GA.

Gibberellic acid treatments are recommended also for the navel orange in California. As the navel orange approaches legal maturity in California, the concentrations of chlorophyll pigments in the flavedo of the fruit decrease and carotenoid pigments increase. During the change in color from green to orange, the rind softens rapidly for a period and then continues softening at a slower rate throughout the harvest season (Coggins and Lewis, 1965). Since color development commences prior to the attainment of legal maturity, softening

actually begins before the start of the harvest season, continuing for as long as fruit remain on the tree. Under California cultural and marketing conditions, fruit may be harvested as late as eight months after onset of softening. Thus, there is ample opportunity for extremely soft rind tissue to develop, along with other senescence effects, contributing to a number of physiological rind disorders in the late harvest season which reduce shelf life and market value.

During the eight-month softening period in California, the capacity of rind tissue to utilize glucose decreases, sugars accumulate to approximately twice their initial concentration, and the ratio of potassium to calcium plus magnesium increases (Lewis *et al.*, 1967). These physiological changes suggest that mitochondrial and plasmalemma membrane systems become less functional during aging.

Recent histological studies by the senior author provide a structural basis for the soft condition that exists in senescent rind. A detailed description of rind tissue is presented in [Chapter 1](#). Interested readers also should refer to studies of navel and Valencia orange rinds reported by Scott and Baker (1947) and Bain (1958). These studies provide excellent descriptions of the structural changes that take place from anthesis to maturity, although the senescent stage is not considered in detail. In general, cells of the flavedo and albedo enlarge, become highly vacuolated, and change shape during development, maturation, and senescence ([fig. 6-3, A-F](#); magnifications are expressed on an area basis). Many intercellular spaces develop, and cell walls appear to weaken and break, particularly in the albedo. Tissue of the senescent rind appears to be structurally weak, and the relatively low amount of cytoplasm suggests that the tissue may possess low metabolic activity.

Preharvest treatment of individual navel orange fruits at the commencement of softening with 500 ppm of GA causes a considerable delay in the rind maturation and senescence processes ([fig. 6-3, G-H](#)). Although the response to high concentration treatments of individual fruits is greater than that obtained with entire tree sprays of 5 to 20 ppm of GA, the same types of anatomical changes probably account for the presence of firmer rinds on fruits treated with recommended rates.

The events leading to senescence in navel orange rind appear to include pigment changes, softening of the tissue, increases in carbohydrates, some changes in carbohydrate metabolism, and changes in cation concentrations. All of these factors clearly are modified by GA, resulting in a delay in senescence.

Rind disorders that appear to be associated with aging include rind staining, water spot, increased susceptibility to decay, puffy rinds, and accumulation of a rind exudate. Each of these disorders can be reduced by GA treatments.

The reduced susceptibility of GA-treated fruits to rind staining (Eaks and Jones, 1959; Coggins *et al.*, 1963) is probably related to the retardation of the softening process. Rind staining is a physiological disorder associated with mechanical abrasion during harvesting, handling, and packing. Treatment with GA to reduce rind staining also delays loss of green pigments. The delay in loss of pigment can be a serious disadvantage, if conditions make it necessary to harvest while the fruits still retain chlorophyll, since consumers interpret this characteristic as a sign of immaturity. Treatment should be carried out after a marketable color has developed.

When navel oranges age on the tree, the rind surfaces occasionally become sticky. The sticky material cannot be removed by washing and packing operations, and thus constitutes a problem that detracts from quality. The condition has been substantially reduced by preharvest treatments with GA (Coggins *et al.*, 1965).

Various oil emulsion formulations sprayed on the foliage of navel orange trees in late summer or fall to control scales, mites, and other citrus pests increase the susceptibility of fruit to water spot. This disorder is brought about by surface deposits of water entering the rind and accumulating in its tissues. The rind spots serve as ready points of entry for decay organisms. Even those fruits that escape decay are of no value on the fresh fruit market. Treatment of fruit with GA appears to substantially reduce water spot (Riehl, Coggins, and Carman, 1965).

Although delayed aging of grapefruit and Valencia orange rind tissues is a useful response, treatment of these fruits with GA results in two rind color problems. Gibberellic acid applied to green fruits delays loss of green pigments; applied to fruit of mature color, it enhances regreening.

FRUIT STORAGE

The keeping quality of fruit is a primary concern of the citrus industry. In general, the maximum interval between harvest and consumption of oranges and grapefruit is two months. Lemons may be kept in lengthy storage before being placed in cartons for shipment. Length of storage of lemons ranges from a few weeks for yellow-colored fruit to five or six months for dark-green fruit. Extensive research is being conducted on means of prolonging storage and reducing storage disorders.

Prior to the late 1940's, *Alternaria* decay was one of the most serious fungus diseases of stored lemons. The first evidence that *Alternaria* decay could be controlled by modification of fruit physiology came from preharvest treatments with 2,4-D and 2,4,5-T. In both preharvest and postharvest treatments, 2,4,5-T has proven more effective in control of the disease than 2,4-D, and ester formulations have been more effective than amine salt formulations. [Table 6-8](#) presents data comparing the effects of 2,4-D and 2,4,5-T on storage life of lemons. Since no residue tolerance has been established for 2,4,5-T, however, this discussion is centered around the use of 2,4-D.

Although 2,4-D is relatively ineffective in inhibiting the growth of *Alternaria in vitro* (Erickson, DeWolfe, and Brannaman, 1958), treatment of fruit with this compound prior to storage reduces the fungus to a minor role in storage decay (Stewart, 1949; Stewart, Palmer, and Hield, 1952; Erickson, 1952; Iwasaki, Nishiura, and Shichijo, 1956). The primary reason for success appears to be that treated lemons are more resistant to the entry of fungus. Once the fungus has gained entrance, it grows as well in treated as in nontreated fruits (DeWolfe, Erickson, and Brannaman, 1959). *Alternaria* spores are present under the button (receptacle and calyx) of most lemons (Bartholomew, 1926). Once the button dies, invasion can readily occur. Treatment with 2,4-D delays the development of the abscission layer, the button remains in a living condition ([table 6-8](#)), and entry of the fungus is reduced. The usual practice is to apply 2,4-D as a final step in the washing procedure for lemons. Where wax is applied prior to storage, 2,4-D is added with the wax.

Treatment with 2,4-D also delays aging and reduces external decay (which is probably a result of retardation of the aging process). Delayed aging is characterized by firmer fruits and a delay in development of the lemon-yellow color. If the dosage is too high, the delay in coloring can become a problem.

A reduction in black buttons, reduced *Alternaria* decay, and delayed aging also has resulted from the use of 2,4-D on oranges and grapefruits (Stewart, 1949). Delayed yellowing of Persian limes (Hatton, 1958) and improved storage life of Clementine mandarin (Gutter, 1956) has been effected by postharvest treatments with 2,4,5-T.

FLOWERING

There is little doubt that flowering of plants is under chemical control, since qualitative and quantitative changes of plant-growth substances occur during flower induction and development (Harada and Nitsch, 1959). However, which changes are the cause and which are the result of flowering has not been established. Among the major environmental factors known to be critical for flowering of most plants are temperature, soil moisture, and photoperiod. The precise effects and interactions of these and other factors on flowering are not known in detail. In the case of citrus, it is clear that flowering usually follows the breaking of dormancy induced by an appreciable period of cool temperatures or drought. Photoperiod does not appear to significantly influence the flowering of citrus (Furr, Cooper, and Reece, 1947).

Methods to accelerate and delay flowering could be of considerable advantage in citrus breeding and commercial production. Breeding programs and early production, especially in close plantings, would benefit from the ability to cause young trees to flower and set fruit. Very young juvenile citrus seedlings, especially grapefruit, often produce a single terminal flower, and then the plant normally does not flower again until it has passed through the juvenile phase of growth (Furr *et al.*, 1947). Flower induction of young grapefruit seedlings appears to be favored by certain low-temperature situations (Hield, Coggins, and Lewis, 1966), although these same conditions do not appear to induce flowering in older seedlings.

Experiments directed toward the control of the flowering process have shown promise for the future. Kessler, Bak, and Cohen (1959) found that xanthine, uracil, and 2,3,5-triodobenzoic acid promoted the flowering of young grapefruit seedlings, but did not affect four-year-old trees. Monselise and Halevy (1964) obtained flower induction on branches of young lemon trees from applications of benzothiazole-2-oxyacetate. Some slight effect was obtained from (2-chloroethyl)trimethylammonium chloride and *N*-dimethylamino succinamic acid. A pronounced decrease in vegetative growth and a marked increase in numbers of flowers produced the following spring resulted from the application of *N*-dimethylamino succinamic acid to apple, pear, and sweet cherry trees (Batjer, Williams, and Martin, 1964).

Monselise and Halevy (1964) inhibited flowering of orange trees with high concentrations of GA. Delayed flowering of lemons also has been observed after GA treatments. To bring harvest period into phase with demand, delayed flowering would prove particularly advantageous with lemons and limes.

Both delayed flowering and inhibition of flowering after GA treatment have been reported for a number of tree crops (Hull and Lewis, 1959; Crane, Primer, and Campbell, 1960; Griggs and Iwakiri, 1961). A combination of 2,4-D and NAA has delayed the flowering of cherry trees (Tukey and Hamner, 1949). Such results usually have been accompanied by undesirable vegetative effects.

VEGETATIVE EFFECTS

Some information is available on the influence of plant-growth substances on the vegetative growth of citrus. One vegetative growth response, the use of maleic hydrazide (MH) to enhance dormancy and reduce susceptibility to frost, is already being used commercially to some extent in Florida.

Haas and Brusca (1954*a*) improved the germination of Koethen sweet orange seeds by soaking them in dilute solutions of 2,4-D and by applying the solution to seed beds. The same authors (1954*b*) studied the effect of soil applications of 2,4-D on growth of eight citrus varieties. Certain varieties were stimulated to greater vertical growth at low concentrations of 2,4-D, whereas higher levels increased foliage density rather than height.

Increased seedling growth has been obtained with GA (Marth, Audia, and Mitchell, 1956; Lange, 1957; Kuykendall, 1958; Randhawa and Singh, 1959; Monselise and Halevy, 1962; Nishiura and Iba, 1964), although the same treatment was found by some of the investigators to decrease root growth (Kuykendall, 1958; Monselise and Halevy, 1962; Nishiura and Iba, 1964). Marth *et al.* (1956) reported an initial increase in shoot growth rate after treatment with GA, but no effect on height of plants was evident four months after treatment.

In Texas, Cooper and Peynado (1958) treated young grapefruit trees with GA, initiating shoot growth during periods between the spring, summer, and winter growth flushes. As with untreated trees, growth on treated trees occurred in a series of flushes and total growth was not improved by GA. Some treated shoots developed long thorns and abnormally narrow leaves. Essentially the same results have been obtained with young navel trees in California.

A common cultural practice with lemons and some other citrus trees is removal of part of the tops of the foliage canopy. Frequent top mowing is a practice in many lemon groves. Hield, Coggins, and Boswell (1963) demonstrated that application of MH to young regrowth in the top of lemon trees will suppress subsequent growth (table 6-9), but have little effect on the rest of the tree. Growth inhibitors such as MH may eventually prove useful in suppressing top growth, restricting lateral growth in close plantings, and maintaining a desirable tree shape for mechanical harvesting.

Dormant citrus is more frost resistant than actively growing trees (Cooper, 1959). Prolonged dormancy has been obtained by winter application of MH to Valencia orange and grapefruit trees in California (Erickson *et al.*, 1952). Although MH has been shown to be generally ineffective for inducing dormancy of trees in Texas (Cooper, Peynado, and Otey, 1955; Cooper, 1959), it has shown promise in Florida (Stewart and Leonard, 1960). Treatment of nonbearing Florida trees induced dormancy and increased frost resistance. The length of dormancy varied from one month to as long as six months after sprays of 1,000 ppm of MH were applied in two consecutive years (Hendershott, 1962*b*). In California, where cooler winters prevail, studies indicated that MH is of uncertain value for increasing the frost tolerance of young trees (Bums, Garber, and Hield, 1962). The fact that GA can be employed to break natural dormancy in citrus (Cooper and Peynado, 1958) raises the question of whether GA can be used to break MH-induced dormancy. A number of other compounds have failed to induce dormancy in citrus (Cooper, 1959).

Plant-growth substances have played a major role in enhancing the rooting of cuttings from a wide range of plants. As early as 1935, Cooper (1935) reported that lanolin containing IAA greatly increased the rooting of lemon cuttings. Treatments not only caused increased production of roots on leaf-bearing cuttings, but also initiated root formation on leafless cuttings. No roots formed on the untreated leafless cuttings. Cooper (1938) reported that treated lemon cuttings produced a band of meristematic cells between the phloem and xylem in two days. Within six days, some of these cells had differentiated into root primordia.

Cooper (1940) found that citrus appeared to root best from leaf-bearing, mature terminal growth. IAA and indolebutyric acid (IBA) treatments seemed to be equally effective in enhancing the rooting of many citrus varieties, but treatment with talc dust containing the compounds was not as effective as soaking the base of cuttings for twenty-four hours in water solutions of the growth regulators.

Although IAA and IBA have proven equally effective in promoting rooting of many citrus varieties, NAA was reported superior to IAA for the rooting of sweet lime (Jauhari and Rahman, 1958). Kulkarni (1957) showed that indolepropionic acid could be used to improve the rooting of lemon cuttings. Erickson and Bitters (1953) investigated the effect of NAA, IBA, 2,4-D, and 2,4,5-T on the rooting of one representative each of five Rutaceae genera other than *Citrus*. No single compound that enhanced rooting was found best for all of the genera studied.

Improved bud grafting in grapefruit has been reported with IAA and IBA. Removal of wood from the bud shield combined with IBA gave the best results (Maiti, Singh, and Singh, 1959).

RESIDUES OF PLANT-GROWTH REGULATORS

The possibility of residues of plant-growth regulators remaining in citrus fruit at harvest presents a problem that will become increasingly important as commercial applications are developed for these compounds. In the United States, it is illegal for fruits to enter into interstate commerce unless a product used in treatment has been registered with the U.S. Department of Agriculture and the Food and Drug Administration has established a safe residue tolerance for the compound.

Before any registration of the product can be approved, there must be evidence of a cultural problem of sufficient magnitude to justify the use of an agricultural chemical to combat it. Such a chemical must provide some control over the problem and be relatively safe for the crop in question as well as the production of subsequent crops. The chemical employed in treatments must leave no toxic residue in the crop, or any residue must be of such low magnitude as to present no health hazard.

With the exception of 2,4-D and gibberellic acid, little data is available concerning residues of plant-growth regulators in citrus tissues. By means of bioassay tests, Stewart *et al.* (1952) found rind surface residues of 2,4,5-T in citrus fruits two and one half months after postharvest application of triethanolamine salt. Their studies revealed no residues from postharvest applications of the isopropyl or butyl esters of 2,4,5-T. Erickson (1955) calculated that the deposit left by postharvest applications of 2,4-D to lemons averaged between 1.8 and 2.8 ppm of the fruit weight. More recently, Erickson and Hield (1962) described a method of analysis for 2,4-D in citrus fruits and reported residue data obtained. They found that oranges from trees sprayed with 20 ppm of 2,4-D contained an average of 0.1 ppm of the compound one day after treatment and that postharvest applications of 500 ppm to lemons gave an average of 1.1 ppm two days later. One week after treatment, the oranges contained less than 0.1 ppm of 2,4-D. Most of the 2,4-D was recovered as acid, although some unknown ester-like residue was also present. Since the isopropyl ester of 2,4-D was applied, there was an unresolved question as to whether this particular molecule was present as part of the residue. Evidence is now available that all of the isopropyl ester entering the fruit is hydrolyzed and is not found as residue. Either 2,4-D acid or the isopropyl ester can serve equally well for the 2,4-D portion of the unknown ester-like compound that is synthesized by the plant tissues (Erickson, Brannaman, and Coggins, 1963).

Two plant-growth regulators are presently registered for use in treatment of citrus in the United States. Preharvest sprays of the isopropyl ester of 2,4-D are registered for oranges, grapefruit, and lemons. Postharvest sprays of 2,4-D on lemons also are registered. Gibberellic acid is registered for preharvest use on fruiting lemon and navel orange trees. Because no significant residues were detected when gibberellic acid was employed in preharvest treatments for the purpose of delaying fruit maturity and rind senescence, registration for its use in California was obtained on a no-residue basis. The registration stipulated that lemon fruits may not be harvested within thirty days and navel oranges within ten days after application.

[LITERATURE CITED](#)