INCREASING CITRUS FRUIT SIZE WITH SYNTHETIC AUXINS

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The price of fruit is largely determined by quality factors whose appreciation is at the mercy of customer's whims. For the fresh fruit market flavor and appearance are still basic determinants for the acceptance of the fruit and the popularity of the cultivars. In addition, the importance of fruit size as a parameter of quality has increased markedly in recent times. This is reflected in the changes in the legal regulations which have risen recently in minimum diameter to accept a fruit as marketable in the European markets. Further, the consumer's preference for big fruit determines huge differences in market price to a point that the income from the smaller, albeit marketable, fruit is often lower than the actual costs of production and commercialization. Fruit size has become as important as total yield in the determination of the profitability of the citrus plantations. This applies not only to the small-fruited mandarins but also to species of a larger fruit size such as lemons, oranges and grapefruits.

Fruit size is affected by many factors and for a given cultivar may range between wide limits. In a survey carried-out with more than 500 trees of the Navelate sweet orange it was found that the mean fruit weight ranged from 280 g (a very large fruit) to less than 125 g (Guardiola, 1988). Some of the factors affecting fruit size are beyond the control of the grower (climate effects) or may not be manipulated (soil type and rootstock). On the other hand, fruit size is inversely related to fruit number and crop load, but this effect accounts for less than 50% of the total variability in size. Flower number, a physiological parameter which may be manipulated, and several cultural practices -pruning, girdling, fertilization and irrigation-, have marked effects on fruit size. However, in many cases a premium is obtained increasing fruit size beyond the limit which may be obtained through the optimization of these parameters. The application of synthetic auxins is performed to achieve this goal.

The Determination of Fruit Size

Fruitlet growth, and final fruit size, results from the accumulation of dry matter and water. It is determined by the **sink strength** of the fruit and the **supply of metabolites**. The sink strength of the fruit measures its potential capacity to accumulate assimilates. It is largely affected by the genetic potential of the cultivar, but is markedly affected by environmental conditions (temperature) and by flower quality, a complex parameter which depends on flower number, the type of inflorescence in which the flower is borne and location in the tree (Guardiola, 1992). Metabolite supply depends on their availability in the tree and on intersink competition.

Both parameters are partly related. Carbohydrate demand increases the rate of photosynthesis, but this increase in not unlimited and a minimum leaf area per fruit is necessary to obtain a maximum fruit size. Conversely, short periods of limitation in supply may affect irreversibly the sink strength of the fruitlet and reduce its growth rate at later stages of development.

Fruitlet growth rate may be limited both by the sink strength of the fruit and by the supply of metabolites, and the limiting factor may change during the different stages of fruit development. The tight relationship between final crop yield and fruit count per tree indicates that demand of assimilates by the fruit is the main limiting factor in the determination of crop yield. On the other hand, individual fruit weight is inversely related to fruit count by tree (Goldschmidt and Monselise, 1977; Guardiola, 1988). From the kinetic analysis of fruit growth (Van-Rensburg et al., 1996) and the correlations in fruit size during several stages of fruit development (Guardiola, 1988; Guardiola et al.,1988) it seems that in most cases a period of supply-limited growth rate occurs at the end of the physiological fruit drop (June drop). Final fruit size is largely determined at this time.

Both conditions are amenable of manipulation through the application of appropriate synthetic auxins. When fruitlet growth rate is increased reducing intersink competition through the removal of a part of the developing fruitlets the auxins are being used as **thinners**. On the other hand, when we intend to increase the sink strength, and hence fruit size, of the fruit without affecting fruit count, the auxins are being used as **fruit growth enhancers**.

Auxin Effects on Fruit Development

The application of synthetic auxins to developing citrus fruits results in several direct effects on fruit development which affect fruitlet behavior both in a direct way and through the changes in assimilate partitioning in the tree. The main aspects of the auxin effects are shown in the flow chart diagram depicted in Figure 1, in which both the direct and the indirect effects caused by the application of auxins are presented.

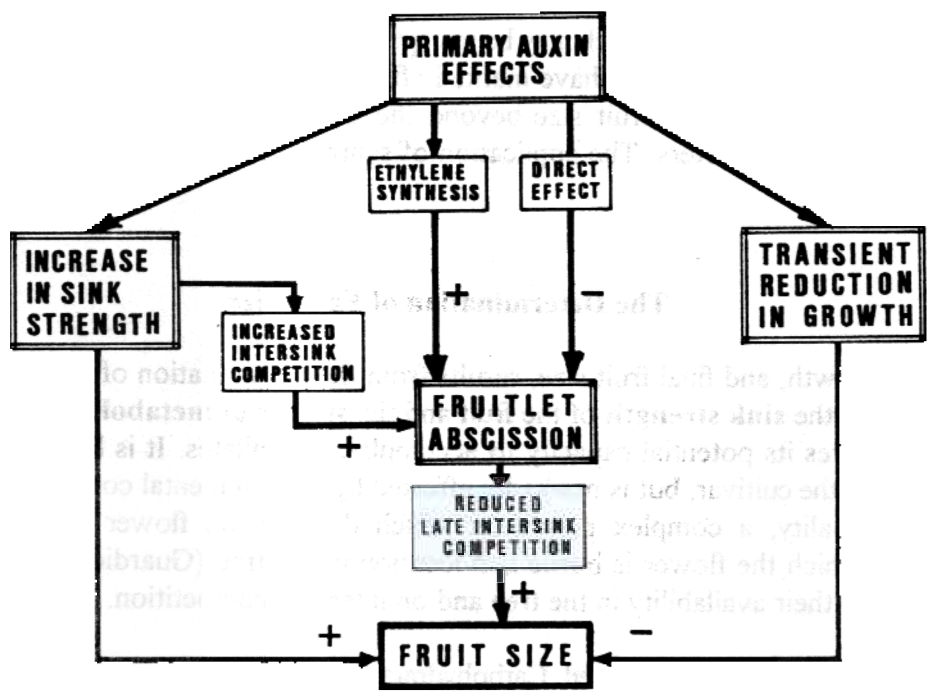

Figure 1. Diagram showing the primary effects of synthetic auxins on fruit set and growth and the influence on final fruit size. From Guardiola (1988).

In general terms, the application of an auxin has four primary effects on fruitlet development:

a) A transient reduction in fruitlet growth rate. As discussed elsewhere (Guardiola, 1996) this effect is a general response to the application of auxins, particularly when performed during the earlier stages of fruitlet development (Guardiola et al., 1993). This effect may result in a final reduction in fruit size.

b) A direct effect on fruitlet abscission, which may potentially result in a delay of fruitlet abscission and/or an increase in set. This effect is only shown when ethylene synthesis is low or prevented (see below).

c) An increase in fruitlet abscission which is mediated through the auxin-induced ethylene synthesis by fruit. This additional abscission results in a reduction in late intersink competition and an increase in final fruit size.

d) An increase in the sink strength of the developing fruitlets, an effect which is sometimes measurable several days/weeks after the transient reduction in growth reported in (a). This increase in sink strength results in an increase in final fruit size, but also may induce an increase in fruitlet abscission unrelated to the auxin-induced ethylene synthesis. This later effect is apparent when the increase in fruitlet growth rate occurs before the end of the physiological fruitlet abscission, and results in a further increase in final fruit size.

There are marked differences among the available auxins to elicit the above described effects. Therefore, the relative importance of each one of these effects depends on the nature and concentration of the auxin applied, but also on the stage of development of the fruitlet at the time of auxin application, the citrus cultivar, and the status of the tree, an aspect linked probably to carbohydrate availability. A judicious selection of the conditions may enhance the importance of the desired effect, and therefore we may define the auxin application referring to the main effect sought.

It must be stressed, however, that a complete separation of effects is not always feasible. Further, the conditions of the applications must be determined for each cultivar, as in some cases marked differences in responsiveness have been found.

Auxins as fruit thinners. Since fruit count is inversely related to fruit size, this parameter may be increased through the reduction in the number of developing fruits (thinning). Fruit thinning always results in a reduction in crop yield, but when performed with auxins this reduction is smaller than predicted from the relationships between fruit count and yield. One reason is that, when appropriately timed, the auxins thin selectively the smaller fruits from the tree. Further, thinning results in an increase in the size of the remaining fruits, which compensates in part for the reduction in yield due to the reduction in fruit count.

Thinning by auxins results from the auxin-induced ethylene synthesis. To be effective it must be performed before the end of the June drop (Figure 2). Beyond that moment the fruit becomes largely insensitive and can be hardly thinned. Also, within some limits an early thinning is more effective to increase fruit size than a late thinning. This is a distinct advantage of auxin-induced versus hand thinning. Hand thinning can only be performed, and at a considerably higher cost, after June drop.

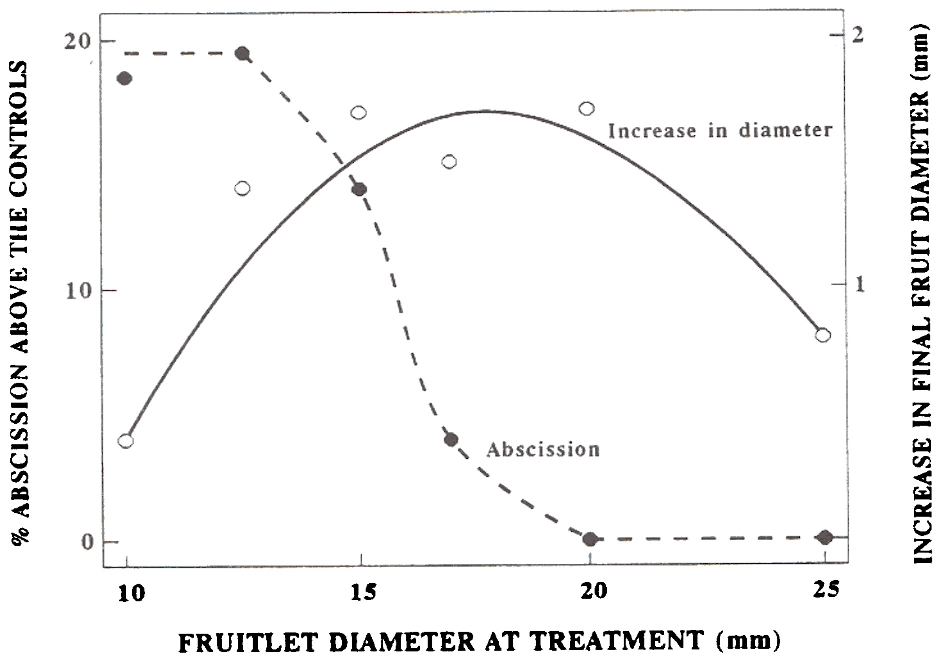

Figure 2. The influence of the stage of development, as defined by the fruitlet diameter, in the growth-enhancing effect and in the abscission induced by a NAA (25 ppm) application. From Guardiola (1996).

Thinning is justified on overloaded trees which produce a significant amount of unmarketable fruit and risk to enter into an alternate bearing habit. In regular bearer trees with a lower crop load the benefits obtained through thinning may be offset by the loss in yield. Further, some of the reported increases in fruit size may not be due to the reduction in competition brought about by thinning but to the direct effect of the auxin on the sink strength of the fruit, and similar effects on fruit size may be obtained without any thinning. This situation is demonstrated in the experiment reported in Table 1 comparing the effect on fruit size in Satsuma mandarin of a thinning NAA application (applied on June 16 during the physiological fruit drop) and that of a later application (performed on July 11) which does not thin fruit (see below). The amount of large fruit produced was similar in both cases despite the 34% thinning of fruit caused by the early NAA application.

Incidentally, this table shows one of the most common pitfalls found in many reports on thinning, which merely show the mean fruit weight. The much higher mean fruit weight for the thinned (84 g) than for the unthinned (71 g) trees does not reflect an increase in fruit size but merely the selective thinning of the smaller fruits, a response which itself does not increase the crop value. Reports should show the sizing of the fruit best expressed in Kg per tree rather than on per cent frequency. In this way, the economic return of the application may be easily calculated.

Table 1. The effect of the timing of NAA applications on thinning, fruit size and crop value in Satsuma mandarin. (In brackets expressed as a percentage of the control values). (From Ortolá et al., 1991).

Parameter (per tree)	Treatments		
	Untreated	NAA (June 16)	NAA (July 11)
Fruit number	885	582 (66)	874 (99)
Mean fruit weight (g)	67	84 (125)	71 (106)
Crop yield (Kg)	56	48 (86)	61 (109)
Small size fruit (Kg) (< 55 mm)	30	15 (50)	30 (100)
Large size fruit (Kg) (> 55 mm)	25	33 (132)	31 (124)

The use of auxins as fruit growth enhancers. The direct effect of some auxins on the sink strength of the developing fruitlets was demonstrated in the early 50's by Stewart and coworkers, but the attempts to exploit this response to increase fruit size in the absence of thinning is quite recent (Guardiola, 1981; Guardiola and Lázaro, 1987; Vanniere et al., 1987).

To this aim, the auxin application is performed after the physiological fruitlet drop, at a time the fruit is not sensitive to ethylene-induced abscission. The increase in fruitlet growth rate results in an increase in final fruit size when fruit load is not too high. When fruit load is excessive, fruitlet growth is limited by metabolite supply and the response to these auxin applications is weak (García-Luis, 1985). Further, as there is no reduction in fruit count crop yield should be increased.

In practice, an increase in yield is rarely found. The sensitivity of the fruitlets to applied auxins is lost rapidly after the end of drop, and the applications are usually performed at slightly earlier dates, during the last days of June drop, which results in some thinning. Typical results obtained with this technique are shown in Table 2 for satsuma and clementine mandarins. There is a small reduction in

Table 2. The effect of an application of 2,4,5-T (10 ppm) at the end of June drop on yield and fruit size in Fino Clementine and Owari Satsuma. (From Guardiola, 1996).

Cultivar and treatments	Fruit count (thousands tree^{-1})	Mean fruit weight (g)	Crop yield (Kg tree^{-1})
Fino Clementine			
Untreated	2.04	55.7	113
2,4,5-T treated	1.71*	61.4**	109^{NS}
SE	0.07	0.5	4.2
Owari Satsuma			
Untreated	1.99	70.4	138
2,4,5-T treated	1.69^{NS}	75.7**	127^{NS}
SE	0.12	1.3	8.2

NS, * and **. Non-significant and significant at P = 0.95 and 0.99, respectively, in the paired comparison to the untreated controls.

fruit count which in many cases is below the statistical significance, which is compensated by the increase in fruit size, and crop yield is not affected.

These initial observations performed on small-fruited mandarins have been extended recently to other citrus cultivars and the conditions for the application of this technique have been determined for many of them. As mentioned above the response obtained depends critically on fruit load. Also, there are marked differences among auxins to elicit a growth response from the fruitlets. Further, considerable expertise is needed to time the auxin applications. A late application is largely ineffective while too early ones may cause an undesirable thinning (Figure 2). Under optimal conditions, the results on fruit size are comparable to those obtained through thinning.

The response to the applications at flowering. An increase in fruit size resulting from the application of the synthetic auxin 2-4,D at flowering was reported for oranges and grapefruits by Stewart and coworkers in the 50's, but this research was not pursued and the response was considered to be too erratic for practical use. We have pursued this research in my laboratory and found that at lower application rates than initially used the application of 2-4,D at flowering results in a reliable thinning of several mandarins and hybrids (Duarte et al., 1996). Further work confirmed a similar response for oranges and grapefruit. At variance with the thinning effect reported above, this thinning does not reduce crop yield, and the reduction in fruit count is compensated by the increase in fruit size (Table 3).

Table 3. The effect of the time of a 2,4-D (20 ppm) application on yield and fruit size in Esbal clementine. (From Duarte et al., 1996).

Treatment and date of application	Fruit count (thousands tree^{-1}	Mean fruit weight (g)	Crop yield (Kg tree^{-1})	
			Total	Marketable
Untreated (control)	1.83	46	84	52
2,4-D at flowering	1.30	61	80	69
2,4-D 2 weeks AFB	1.31	58	76	65
2,4-D 6 weeks AFB	1.59	55	88	72
SE	0.09	1.6	5.4	4.6
Significance (F value)	0.01	0.01	NS	0.05

The mechanism of this thinning effect is different to the ethylene-mediated thinning described above. At the concentration used (17 to 20 ppm) the application of 2-4,D does not induce ethylene synthesis. There is a selective effect on fruitlet growth rate, which is increased in some but not all the developing fruitlets, while fruitlet abscission is initially delayed. At the end of June drop, the rate of abscission becomes markedly higher in the 2-4,D treated trees through the selective abscission of the smaller fruitlets, so final set is finally lower.

As compared to the other systems of fruit size manipulation reported, the application at flowering has some distinct advantages. Perhaps the most relevant one is that the time of application is not critical and may be performed at any time from flowering up to at least 6 weeks after full bloom, not to mention that 2-4,D is the cheapest of all the available auxins. As a limitation it should be remarked that for an unknown reason some cultivars seem to respond poorly to this application at flower opening.

Concluding Remarks

Increasing fruit size through auxin applications has been for a long time an open possibility for citrus growers. This technique was not popular in the near past as the results were considered unreliable or, at least, considerable expertise was deemed necessary to succeed with their use. With the increasing influence of fruit size on market prices and the better understanding of the mechanism of action of the auxins leading to consisting and reliable results the time has come to exploit the advantages obtained from their use.

As presented above there are three different basic mechanisms of auxin action, and each one has advantages and pitfalls (Table 4). The most appropriate technique shall be determined by cultivar sensitivity and the general status of the tree, and in many cases similar results are obtained with different techniques and the choice may be left to the grower preference. In any case there is not a best technique or auxin, and auxin applications can not substitute for basic failures in cultural practice.

Table 4. Advantages and pitfalls for the different strategies for the use of synthetic auxins to increase fruit size in citrus. (From Guardiola, 1996).

1. **THINNING PART OF THE DEVELOPING FRUITLETS**
   - There is always a reduction in yield.
   - Response depends on temperature and fruit load.
   + Advisable when non-marketable small fruits are expected.
   + It may correct alternate bearing

2. **INCREASING THE SINK STRENGTH OF THE FRUITLETS**
   - Considerable expertise is needed to time the applications.
   - Response may be weak on overloaded trees.
   + Total yield either is not affected or is slightly reduced.

3. **APPLICATION OF AUXINS AT/SHORTLY AFTER ANTHESIS**
   - Not all cultivars are equally responsive.
   + Easy to apply and to time the applications.
   + No significant effect on total yield.
   + Less adverse effects on fruit quality.

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