FRUIT DEVELOPMENT AND SENESCENCE

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Introduction

The mature citrus fruit represents the end of a complex set of events starting with the formation of reproductive structures we call flowers. Likewise, the formation of flowers is preceded by complex and incompletely understood physiological processes and morphological changes. The purpose of this lecture is to briefly describe (not explain) some of the morphological and horticultural changes between the time citrus flowers open and fruits mature. This lecture draws heavily on the 4 papers cited, on the author's experience and knowledge of appropriate literature, and on the author's research.

Morphological Changes

Between full-bloom and maturation, cell division, cell differentiation, and cell growth convert a small citrus ovary into a fruit of considerable economic value. Generally, large-fruited citrus varieties start out with relatively large flowers which have relatively large ovaries. In the case of Valencia orange, the transition from an ovary at full-bloom to a mature fruit represents something on the order of a 4,000-fold increase in size. That is, an ovary of approximately 0.04 ml with 8 to 12 carpels develops into a mature fruit of 160 ml containing 4,000 or so juice sacs in 8 to 12 segments.

Excellent fruit development studies have been done by Bain (1958) and Holtzhausen (1969) on Valencia and navel orange, respectively. Also, in 1981, Holtzhausen referred to a useful unpublished (M.Sc. thesis) study made by Button on grapefruit. Because these studies are in close agreement, I have assumed that their findings are generally applicable to citrus.

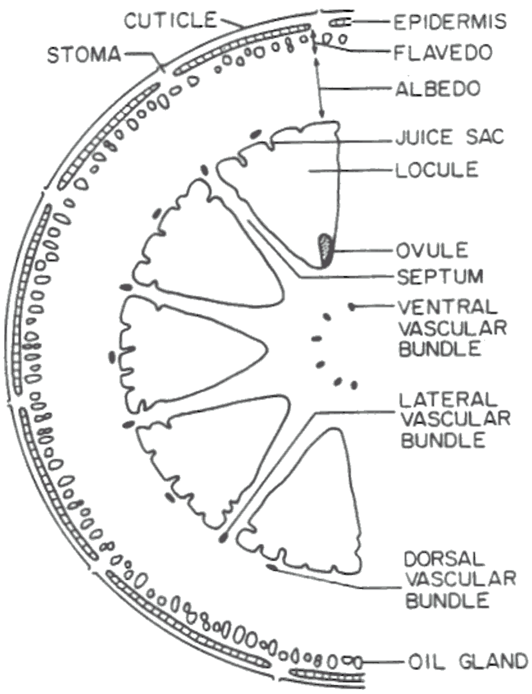
Figure 1 is a diagrammatic representation of a citrus ovary at full-bloom. A brief review of the location of various tissues will make the following discussion more meaningful.

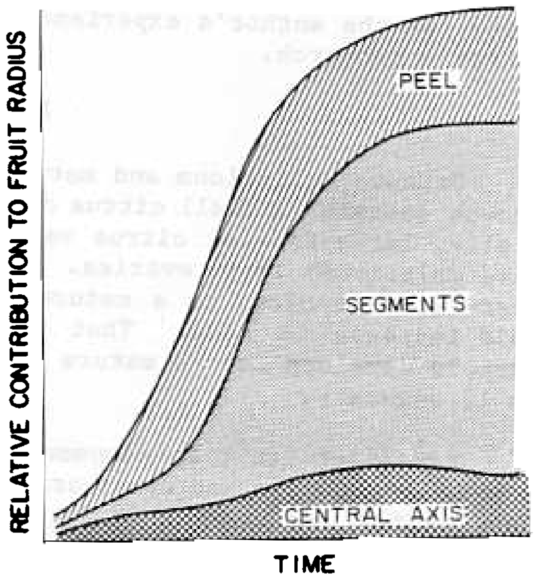
Stone fruits, fig, raspberry, blueberry, grape, and olive have a double sigmoid growth curve. Apple, pear, date, pineapple, banana, avocado, strawberry, tomato, melon, and citrus fruits have a single sigmoid curve as shown in Figure 2 for grapefruit.

Whether the growth pattern of a fruit fits a single or a double sigmoid curve cannot be used to decide whether the fruit is of the climacteric or nonclimacteric type. For example, a sharp rise in respiration is associated with the ripening of an avocado. No such rise is seen at any maturation stage of citrus fruits. Avocado and citrus have a single sigmoid growth curve. Avocado is a climacteric fruit and citrus fruits are nonclimacteric.

<u>Stage I</u>.--Bain described 3 stages of development for the Valencia orange. Stage I is a period of slow volume growth but of intense cell division throughout all tissues. This period requires approximately 9 weeks. Subsequently, cell division is found only in the outer layers of the peel and in juice sacs. Holtzhausen found essentially the same for the navel orange, including a time span of 9 weeks. Because the navel orange matures in considerably less time than the Valencia orange, it is obvious that the navel orange spends less time at Stages II and III than does the Valencia. However, apart from time differences, the developmental events are similar for these cultivars.

In Stage I, fruit growth is due to cell division and cell enlargement. Oil glands present at full-bloom enlarge and new ones appear. Juice sacs are present at full-bloom and continue to be formed throughout Stage I. Growth of juice sacs during Stage I is primarily by cell division, which occurs mainly at the head of developing sacs. However, most of the volume growth that occurs in Stage I is due to the peel. For example, Bain found that the peel made up 95% of the fruit volume at the end of Stage I.





1. (left panel). Diagrammatic cross-section of a citrus fruit (adapted from Holtzhausen, 1969).

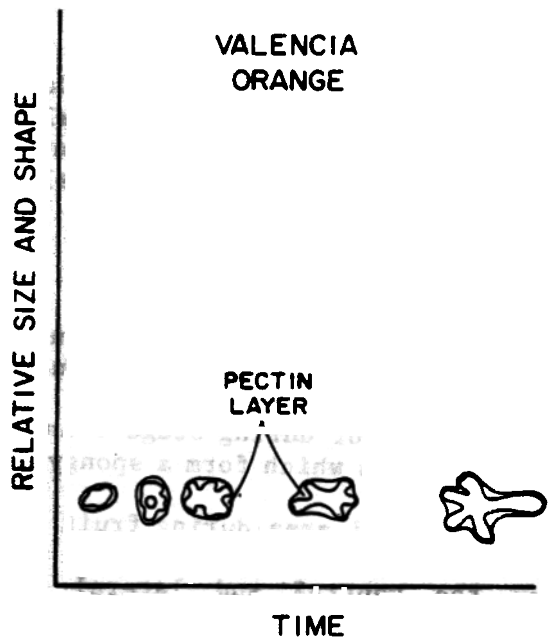
2. (right panel). Development of the grapefruit (adapted from Button as reported by Holtzhausen, 1981).

<u>Stage II</u>.—The demarcation between stages is not abrupt, but Stage II is characterized by very rapid fruit growth. During this period, cell enlargement and cell differentiation predominate. Except for the epidermis, the outer layers of the flavedo, and the tips of juice sacs, cell division has concluded by the beginning of Stage II.

During the early parts of Stage II, the peel grows in thickness, mainly by enlargement of albedo cells. Subsequently, the peel becomes thinner in concert with rapid growth of the pulp segments. During the time the peel becomes thinner, the albedo cells continue to enlarge. The peel becomes thinner because the albedo cells enlarge primarily in the tangential direction via growth of arms (Fig. 3). At this stage of development, albedo cells have the general shape of spiders positioned such that they join one another at the tips of their legs. This forms a spongy tissue. Due to further growth of the arms of these cells, considerable increase in circumference along with a decrease in rind thickness occurs. This growth pattern results in fewer cell layers than existed in the peel at the end of Stage I.

The same kind of spongy tissue that develops in the albedo also develops in the central axis and in septum tissue. In absolute terms, there is considerable growth in all these tissues during Stage II. However, in absolute as well as relative terms, the increase in size during Stage II is due mainly to growth of the pulp segments. For example, at the end of Stage I, Bain reported that the central axis and pulp accounted for 5% of the volume of the developing Valencia orange. At the end of Stage II, she found that the central axis and pulp accounted for 58% of the volume and 67% of the fresh weight of the fruit. Most of this is due to pulp segments—juice sacs become larger and juice content increases in the enlarging cells of the juice sacs.

The relative contribution of the central axis, endocarp (pulp segments) and peel to the cross-sectional area of the navel orange throughout development is shown in Figure 4. Even though cross-sectional area figures are less dramatic than volume changes, it is clear that the endocarp is a minor constituent at full-bloom and becomes the major constituent as fruit growth proceeds.



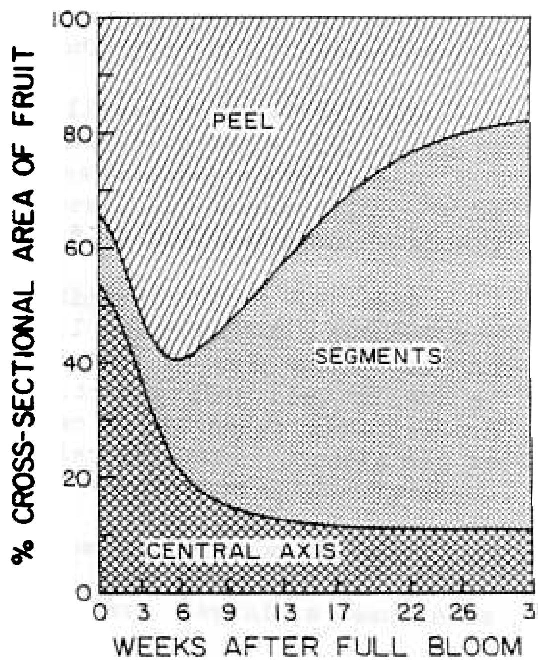


Fig. 3. (left panel). Development of albedo cells - Stage II (adapted from 1958).

Fig. 4. (right panel). Development of the navel orange fruit (adapted Holtzhausen, 1969).

Stage III.—This is the maturation period. Although volume growth continues for essentially as long as the fruit remains on the tree, the rate of growth is considerably lower than in Stage II. Fissures develop in the central axis and in the albedo of many cultivars. In some cultivars, the albedo becomes a minor constituent, and in these cultivars, the peel can be removed with great ease.

In subtropical climates, chlorophyll pigments disappear from the flavedo and carotenoid pigments are revealed. In many cultivars, carotenoids increase substantially and/or are converted into highly colored pigments during and after the loss of chlorophyll from the flavedo. Depending on the type of citrus fruit being considered, regreening occurs during Stage III.

Sugars and water continue to accumulate in juice sacs and the concentration of organic acids decreases.

Summary.--Now that you have a general idea of morphological changes that occur during development, let's briefly look at the summation of changes that occur in the various parts of the fruit. We will start with the outer-most part and proceed inward.

The cuticle is very thin at full-bloom and becomes somewhat thicker during growth and development. Wax components of the cuticle increase substantially in quantity and complexity. Stomata appear several weeks after full-bloom and become somewhat plugged during maturation and senescence. The epidermis is one cell layer deep and cell size increases little, if any, during fruit growth. The tremendous increase in circumference is brought about primarily by continued cell division.

Cell division and cell enlargement occur in the outer layers of the flavedo up to maturity. Growth of the inner layer of the flavedo and growth of the albedo is due to cell division and enlargement through Stage I, and to cell enlargement thereafter. Oil glands are present at full-bloom and new ones develop continuously. Oil glands enlarge throughout fruit growth and development.

Some juice sac primordia are present at full-bloom on dorsal and lateral walls of the locules. During Stage I, the number and size of primordia increase. Cell division occurs in the tips of juice sacs into the early part of Stage II. Subsequent growth is due to cell enlargement. The bulk of the tissue between the segments and in the central axis consists of cells that follow a developmental pattern similar to those of the albedo. That is, cell division and enlargement occur during Stage I and subsequent growth is due to the development of multi-armed cells which form a spongy tissue.

Vascular bundles increase in length and cross-sectional area during fruit development. The dorsal bundle, which probably transports the photosynthate stored in juice sacs, has a larger cross-sectional area than the ventral and lateral bundles.

Final fruit size is a product of cell division and cell enlargement. Cell division predominates in Stage I, cell enlargement is the main feature of Stage II, and maturation events characterize Stage III.

Horticultural Changes

Brix and Acid. Juice sacs are the primary site for the accumulation of soluble solids. The absolute quantity of soluble solids (primarily sugars and organic acids) increases tremendously during growth and development, but toward the end of Stage II, the amount of acid relative to the remainder of the soluble solids decreases. Once the soluble solids-to-acid ratio reaches specific levels in oranges, grapefruit, tangerines, and mandarins, the fruit are said to be mature and of edible quality. Subsequently, the concentration of acid continues to decrease. This causes the ratio to increase. Eventually the solids-to-acid ratio becomes so high that the juice is insipid and the fruit is judged to be overmature, or senescent.

Pigments.—Chlorophyll and some carotenoid pigments are present in flavedo tissue at full-bloom and throughout Stage II in all cultivars. In many cultivars, especially in subtropical climates, essentially all of the chlorophyll disappears from the flavedo sometime during Stage III. In many cultivars, such as white grapefruit, the loss of chlorophyll simply reveals the carotenoid pigments. In other cultivars, additional and different carotenoids accumulate in the flavedo, giving the fruit its characteristic mature color. Carotenoids also accumulate in juice sacs. These pigments are not water soluble. In some cultivars, for example the blood orange, water-soluble pigments, called anthocyanins, also accumulate in juice sacs and in cells of the flavedo. As mentioned earlier, regreening can occur in the flavedo. Regreening is favored by high nitrogen and high potassium nutrition and by warm weather.

Rind Integrity.—At the end of Stage II, the rind is physically strong. During Stage III, the rind softens rapidly and then continues to soften at a slower rate for essentially as long as the fruit is kept on the tree. Whether rapid softening occurs early or somewhat later in Stage III varies with cultivars. Softening seems to be related to fissures which develop in the albedo, and to the development of weaker connections between cells, possibly due to changes in pectic constituents. Also, flavedo and albedo cell membranes become more permeable during this stage of development. It is at this stage of development that citrus fruits become quite susceptible to physical damage and to decay organisms.

Susceptibility to Decay Organisms.—Toward the end of Stage II and early in Stage III, most citrus fruits are reasonably resistant to decay organisms. For example, if spores of *Penicillium digitatum* or *Geotrichum candidum* are placed into lemon rind tissue at this stage of development, decay will not result. At a later stage, these organisms are major problems in the storage of lemons. *Geotrichum* in particular is of no consequence until fruit become more susceptible due to age.

In nature, susceptibility to decay organisms depends on entry and subsequent growth of the organism. Both seem to be more likely as the fruit matures and approaches senescence. This seems to be due to the organism having less trouble entering the rind, either by its own action or through injuries, and on the rind tissue providing a better growth medium for the organism as maturity and senescence progress.

Exogenous Growth Regulators

2,4-D.—This growth regulator is widely used in citrus to reduce the incidence of mature fruit drop. Also, it is widely used postharvest on lemons to delay aging of the fruit and to delay abscission of the button. Whether used preharvest or postharvest, its primary action is to delay the development of the abscission layer.

When used preharvest, the delayed development of the abscission layer permits a longer harvest season with only modest losses from fruit drop. Also, the delayed development of the abscission layer keeps the phloem and xylem connnections in better condition for a longer period of time. This results in a slight delay in fruit senescence. When used postharvest, the delayed abscission and the resulting delayed senescence of the button, keeps the port of entry for the *Alternaria* fungus closed. Decay from this fungus is thereby controlled without the use of a fungicide or a fungistat. The advantage of such an approach is that the fungus is not exposed to a fungicide or fungistat to which it can become resistant.

GA_3.—Under certain conditions, this growth regulator is applied preharvest during Stage II or Stage III to delay fruit maturation or senescence. For example, it is used on grapefruit in South Africa and Florida, on Valencia orange in Israel and California, on Minneola tangelo in Australia and California, and on navel orange in South Africa and California. GA_3 is used routinely on most of the California navel orange acreage scheduled for late harvest. Typically, GA_3 is applied two weeks before color break and 2,4-D is applied approximately 2 months later. The objective is to minimize fruit drop with 2,4-D and to delay senescence with GA_3. Such a combination permits California to continue harvest of navel orange into early July when production and marketing factors make this desirable.

The primary benefit from GA_3 is delayed rind senescence. This reduces the severity of a number of preharvest and postharvest rind disorders, reduces susceptibility to decay, and prolongs storage, shipping, and shelf life. It also causes a modest delay in the decline of acid level in the juice and delays the development of "over-ripe" flavors. On the other hand, once GA_3 is applied, any subsequent plan to harvest early in the season is doomed due to the substantial delay in rind color development caused by GA_3. Obviously, the primary value of GA_3 is for fresh-fruit market situations.

By design, this lecture has placed emphasis on morphological changes during growth and development of citrus fruits and less emphasis on horticultural changes and on the impact of exogenous growth regulators on development and senescence. I hope the emphasis on morphological changes provides the background information you need for the subsequent lectures in this series.

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