Some Aspects of Pollination and Fertilization in Subtropical Fruit Species

C. A. Schroeder
Dept. of Biology
University of California, Los Angeles

Reuben Hofshi
Del Rey Avocado Company
Fallbrook, CA

Many factors are responsible for the ultimate yield in tree fruit crops. Cultural practices such as irrigation and mineral fertilization most certainly influence the ultimate crop load and potential fruit size. Trees have to be well nourished and watered to maximize production. Light is critical for flower initiation and development and the production of assimilates, therefore canopy manipulation to maximize light interception is of greatest importance. Crop loads can be managed to minimize alternate bearing by fruit removal at appropriate times and by judicial pruning. However, without flowers there would be no fruit formation hence this article considers some of the factors which influence the processes of pollination with particular emphasis on avocado pollination.

With few exceptions, subtropical fruit crops reproduce sexually and depend on two interrelated processes: pollination and fertilization:

Pollination: The arrival of one or more pollen grains onto the receptive stigma.

Fertilization (sexual): The germination of the pollen on the stigma, growth of a pollen tube through the style and the union of the sperm and egg to form an embryo.

Flowering and pollination are important aspects of fruit production in most subtropical fruit crops. Many fruits such as 'Washington Navel' orange, 'Bearss' lime, 'Thompson' grapefruit, bananas and pineapples are seedless as the flowers are without viable pollen, hence pollination and seed formation are of little or no importance. Such fruits develop as the result of internal stimuli, a process termed parthenocarpy. The fruit set and yields in such crops, however, often can be influenced by control of moisture and mineral nutrition applications to the trees at appropriate times during the year. A very unique tropical fruit, the mangosteen (*Garcinia mangostana*), has no pollen producing flowers yet will develop fruits with viable seed. The seed with viable embryos develops without pollination from female tissue within the ovule as apomictic or asexual embryos. Still other fruits such as the 'Sultanina' grape are initiated through the pollination process with subsequent embryo formation, but the embryo aborts at an early stage. The fruit continues to develop but is seedless upon reaching maturity.

It is quite possible to induce fruit development in flowers in many species by application of various types of auxins and growth promoters at the appropriate time. Auxin induced fruits are seedless and sometimes quite acceptable for commercial production. Other hormone treated fruits as the Calimyrna fig develop attractive size but lack good quality. The cherimoya can be induced to set fruit by hormone sprays. These sprays need to be applied several times during fruit development, therefore, limiting its commercial feasibility. Avocado can be induced by hormonal sprays to develop seedless fruits, “cucs”, but the resulting fruits are small and only have limited market appeal.

Most fruit species produce perfect flowers with female (pistil) and male (anther) structures in the same flower. Dioecious species bear female flowers and male flowers but on different plants. A good example of this is the date palm. Combinations of perfect (flowers contain both male and female components) and unisex flowers occur in polygamous species such as kaki persimmon and carob. It is also possible to have separate male and female flowers on the same plant. Examples of this type of flowering behavior (monoecious) are walnuts and pecans.

Single seeded fruits such as avocado, mango and litchi generally produce flowers in very great abundance yet, few fruits are set and reach maturity even on mature trees. Avocado trees may have 1.5 million flowers but will set and mature only a few dozen to a few hundred fruits. Plant breeders, who have hand pollinated avocado flowers, will attest to the low returns from carefully hand pollinated flowers. The 9 stamens in the avocado flower produce hundreds of pollen grains, yet only one pollen grain is required to set a fruit. The single stamen of the mango flower produces 200 to 300 pollen grains, one of which can set a fruit. Similarly in wind pollinated species such as walnut and pistachio, the expenditure of energy in excessive production of flowers and pollen and the relatively small numbers of fruit resulting from such efforts has no reasonable biological explanation.

Fruits of the kaki or oriental persimmon cultivars will set adequate to excessive crops of seedless fruit under California conditions without pollination. Most of the common and available kaki persimmon cultivars in California have only pistillate (female) flowers which produce the fruit. When specific pollen producing kaki cultivars are provided nearby, these persimmons can produce an abundance of kaki seed which are sought by nurserymen for use as rootstocks. Pollination of the kaki persimmon apparently is provided by the honey bee.

Many *Citrus* cultivars benefit from self or cross pollination which is expedited in most instances by the honey bee. Self or close pollination is adequate for most *Citrus* cultivars but a few selections such as ‘Clementine’ and ‘Fairchild’ mandarins require specific cross pollination. Many species of *Citrus* exhibit polyembrony and produce 2 types of embryos in a given seed. The product of sexual fusion of the pollen gamete and the egg gives rise to the hybrid, sexual or gametic embryo. Upon formation of the gametic embryo there arises from the tissue surrounding the egg apparatus, one or often several apomictic embryos as a result of an internal stimulus associated with sexual fusion. These apomictic (nucellar or asexual) embryos are derived from the female tissue of the ovule hence provide the exact genetic composition of the female parent. While the seed contains a sexual embryo and possibly several other apomictic embryos derived only from female tissue, it is important to identify each of these. Frequently this can be done by growing each of the embryos of a seed as separate plants to a stage where mature leaf characteristics can identify the plant from the sexual embryo as it will often differ in leaf character from the other plants derived from the given seed. A more precise identification of the individual seedlings can be made by using molecular markers.

The cherimoya is a fleshy fruit with many seeds. It is considered as a special type of fruit, a syncarpium, as the several pistils are fused together and united into a single mass along a central receptacle. The flower is somewhat inconspicuous and green in color but is large, sometimes two or more inches in length. The 3 fleshy petals surround a central mass of spirally arranged pistils with a ring of many anthers at the base. In common with the avocado, the cherimoya flower has both male and female phases within the same flower and exhibits a dichogamous behavior. The flower opens in the morning in the female phase by separating the petals at their tips to partially expose the glistening stigmas of the many receptive pistils. The female phase lasts all that day and the next day until sometime between 3 and 7 p.m. of the second day when the stigmas become dry. At this point their receptivity to pollen is reduced, but some receptivity may exist for a short period of time. Also at this point, the flowers switch to the male phase. The petals of the flower are now open wide and the anthers have begun to shed their pollen. Since the receptivity of the stigmas is absent or very low at the time pollen is shed, self pollination rarely occurs, but when it does self-pollinated fruit are often misshapen due to uneven distribution of pollen. Honey bees will collect cherimoya pollen from the exposed stamens in the afternoon, but cannot enter the unopened or slightly opened flowers in the female stage. Hence, bees are of no value as pollinators in cherimoya. Several species of beetles have been shown to expedite the pollen transfer in the cherimoya's native habitat. Because of the strong floral dichogamy in California, and the low numbers of appropriate beetles, growers can only be assured of adequate fruit set if hand pollination is practiced. It is best to collect pollen anthers in the late afternoon and apply it to flowers which are newly opened at that time with their petals slightly separated. Pollen can be held overnight to be applied by a small brush to flowers in the receptive stage the next morning, but there is less pollen capable of pollination and fertilization the following morning, so this method can be less successful. Hand pollination is an economically feasible practice to obtain high yields of excellent quality fruit in California and other areas where the cherimoya is grown. Hand pollination provides many advantages to the grower such as the positioning of the fruit on the tree which can enhance the ease of harvest and handling. Another advantage is the growers can control the time of fruit set, hence, the time of harvest which allows spreading the duration of the harvest season. Finally, one can control fruit quality particularly as fruit size can be modified by amount of pollen used to set the fruit. Some control of the nature of the surface texture (smoothness) can also be influenced by the choice of the pollen (male) parent.

One of the oldest horticultural practices, pollination in the date palm, was known in biblical times as it is depicted on Assyrian bas-relief and as Theophrastus described it about 326 B.C. Date flowers are borne in great clusters as male flowers in spathes in the crown of one palm and as female flowers on other palms. While wind pollination and occasional visits by honey bees, other insects and perhaps some birds will result in a few fruits, it is necessary to transfer pollen by hand to obtain a satisfactory crop for human use. The grower collects several flower stalks of staminate (male) flowers which are shedding pollen and places these among the flower stalks of the receptive pistillate (female) inflorescences. If the pollen is mature before the pistillate flowers are receptive it can be stored at low temperature under a dried condition for many months, sometimes until the following flowering season. Specific pollens can enhance the fruit quality of certain date cultivars by causing the fruit to mature as much as 2 or 3 weeks earlier. Harvesting of early maturing date fruit can avoid damage from early season rains which can cause fruit damage.

In nearly all fruit species, the general health of the plant especially the status of the mineral nutrition and the availability of adequate moisture throughout the year will influence the several sequential developmental steps in flowering, pollination and fruit set. The fruit buds of many species are differentiated and formed sometimes several months before the flower can be visually detected. Tree nutrition prior to bud appearance can affect the condition and development of the buds. After the buds develop into mature flowers the factors of specific pollination vectors become of importance. Among the several actual and possible pollinators the honey bee is perhaps of greatest importance for most tree crop plants in California.
While the discussion which follows focuses in avocados, the principals discussed applies to other tree crops, including certain *Citrus* cultivars. Effective pollination is critical to one important subtropical crop grown in California, the avocado. The avocado is often planted in environments that are different from its native highlands of Guatemala and southern Mexico. The pollination process of the avocado is influenced by many factors and it is critical to productivity, particularly in marginal environments. Climatic conditions of temperature and humidity are the main factors that affect both pollination and fertilization. The degree of influence is unique to specific cultivars and the growing environment (Table 1).

Factors Influencing the Pollination Processes

Temperature

Temperature is the most important factor influencing floral behavior and pollination in avocado. The avocado flower at maturity exhibits a condition of dicohgamy (protogyny) when the flower opens in a receptive female stage then closes overnight and opens the next day in the male stage. At this time, the pollen is shed and the stigma, under most environments, is no longer functional. When the daily average temperature falls below certain thresholds, there is a disruption in the onset of flower opening. The period between the male and female phases is prolonged and the flowers remain open longer (Sedgely, Gazit, Ish-Am). At low temperatures (below 17 C (63 F) day and 12 C (54 F) night) the female-male flowering sequence can be completely reversed, extended through the night or the female stage can even be skipped altogether (Sedgely). Varying degrees of male-female flowering overlap is common. This overlap is conducive to close-pollination (within the same cultivar), and may help explain the good production encountered in single variety blocks.

Low soil temperatures, below 13 C (55 F) can affect water uptake by the roots (Whiley). A flowering tree under low soil temperature at night followed by a bright sunny day could go through severe water deficit and flower desiccation due to impaired root function (Dr. U. Kafkafi, personal communication).

Relative Humidity

Hot and dry weather will desiccate stigmas, flowers and entire inflorescences of subtropical fruit trees. Eisikowitch and Melamud suggested that female phase avocado flowers will fail to open when ambient relative humidity drops below 45%.

Pollinators and Cross Pollination

The avocado stigma within most avocado varieties and under most environmental conditions appears not to be receptive when the pollen in the given flower is shed. Pollen transfer therefore must rely on some external vector. The avocado flower is visited by many insects such as honey bees, flies, non *Apis* bees and wasps. The honey bee has been shown to be the insect responsible for at least 80-95% of avocado pollination (Ish-Am) and for the majority of pollen carried per hour (Vithanage). The use of the honey bee in avocado, however, does present some practical problems. It is only the nectar collecting honey bee and the nectar-pollen collector that can effect pollination. The strictly pollen forager has no reason to visit the flower in its pistilate (female) stage (Ish-Am). The honey bee prefers *Citrus* and many wildflower crops to avocado flowers. Moreover, avocado pollen grains are large and spherical, and the honey bee must utilize sugar, enzymes and water to form the pollen loads. Pollen loads of mustard, a primary competitor for bee visitation during avocado bloom, are built faster, and are more than double in size (Ish-Am). Observations in Israel indicate that the honey bee will abandon an avocado orchard during the flowering period of nearby citrus trees (Ish-Am, Dr. Y. Adato, personal communication). Increasing bee density has been recommended as means to compensate for the abandonment due to competition.

The effectiveness of the honey bee is influenced by temperature. The honey bee is reluctant to fly out during cold and overcast days and reduces its activity during very warm days. The worker bee will only go to a location related to it by the dance of scout bees and will not forage randomly on its own. The bees relate to flowering events using a biological clock. When temperatures fluctuate, honey bees that were visiting a tree on a given day may not encounter open flowers at the same time on the next day. The empty stomached foragers may be recruited instead to forage in more promising pastures, abandoning the avocado (G. Sherman, personal communication).

Honey bees visit only one to three trees in close proximity (Stout and Ish-Am). In environments where cross-pollination has been shown to contribute to increased avocado yield and fruit retention, it is necessary to plant large numbers of opposite floral type trees at close proximity, theoretically every other tree for best results (Bergh; Gazit; Goldering et al; Ish-Am) (Table 2). Unfortunately, different cultivars in the same block, some of which have low commercial value, complicate orchard management and harvest. Growers conventionally plant pollinizers in rows and expect bees to cross between rows for cross-pollination. Only a limited percentage of bees cross between rows and this percentage is increased with increase of wind velocity. Higher bee densities will escalate the number of bees crossing between rows but not their percentage (Ish-Am). There is a possibility that terrestrial bumble bees may eventually help fill some of the need for distance cross-pollination left vacant by the honey bee foraging behavior (Ish-Am). Efforts are underway to identify the native pollinators of the avocado in its native habitat in Mexico and Guatemala. (Gazit and Ish-Am, personal communication).

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C. A. Schroeder *- Professor of Botany- Emeritus UCLA. Teaching and research, 1943-1983. Special interest in tropical and subtropical fruit crop physiology, structure, pollination needs and fruit development. Traveled to major subtropical countries for advisory assignments, research, and lectures. Published many articles on Citrus, avocado, persimmon, cherimoya, and other fruits. Recipient of California Avocado Society "Merit of Honor and Leaf Cluster Award."*

Reuben Hofshi *has been involved in many aspects of the California Avocado Industry since 1975. He is currently a member of the Production Research Committee.*

Table 1. Factors influencing pollination of avocado flowers.

FACTOR		EFFECT	REFERENCE
Air Temperature	33 C Day/28 C Night (91 F Day/82 F Night)	Less Flowers and Shorter Flowering Period	Sedgley and Annells
	25 C Day/20 C Night (77 F Day/68 F Night)	Best for Pollination	Sedgley and Annells
	17 C Day/12C Night (63 F Day/54 F Night)	Increased the Length of the Floral Cycle	Sedgley and Annells
Soil Temperature	Below 13C (55 F)	Poor Water Uptake Flower Desiccate	Whiley, Kafkafi
Cross-Pollination	Opposite Type Pollinizer	Decreased Effectiveness with Distance	Bergh, Ish-Am, Stout
Bee Density	More than 25 per Tree	Effective Visitation	Ish-Am
Effective Pollination	6-20 Pollen Grains per Stigma	Rapid Pollen Tube Growth	Ish-Am, Shoval
Flower Competition	Citrus and Wildflower	Decrease in Bee Activity Especially in Early to Bloom Varieties	Ish-Am, Stout

Table 2. Avocado Flower Types for Commercially Available Cultivars.[z]

Flower Type	Cultivar
A	Gwen Hass Lamb Hass Pinkerton Reed
B	Bacon Ettinger Fuerte Nabal Sir Prize Zutano

[z] See article by Robbertse et al., pg. 17, for description and timing of avocado flower stages.