Pollination:

Theory & Practice

The CCA Cherimoya Handbook

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Introduction

"The reason for a flower is to manufacture seeds" is a quote from a poem that succinctly explains the biological function of a flower (Heller 1983). Flowers are the site of sexual reproduction in the flowering plants.

Pollination, or the transfer of pollen from the stamen (male organ of the flower) to the stigma (terminal portion of the female organ) is the first step in sexual reproduction resulting in the production of seeds and their container, the fruit.

This chapter focuses on theoretical and applied aspects of the pollination of cherimoya (*Annona cherimola Mill.*), sweetsop (*Annona squamosa L.*), and atemoya, a hybrid between cherimoya and sweetsop. I will also refer to some information that pertains to all members of the Annona family (*Annonaceae*) of the flowering plants.

Background on flower structure and both seasonal and within flower timing of the female and male portions of the flower will be discussed to set the stage for a discussion of the theory and practice of insect and hand pollination.

Flower Structure

Typical Flower Structure

Most flowers consist of four whorls of organs, each with specific functions. At the base of the flower are the sepals, which enclose the young flower bud and are usually green (Fig. 1 Diagram of a typical flower).

Inside and above the sepals are the petals, which are usually brightly colored to attract pollinators. Within the whorl of petals are the reproductive organs, the stamens or male organs and the carpels or female organs.

Each stamen is composed of a stalk called a filament and a terminal sac or anther where pollen is produced. After pollination, pollen will germinate to produce a pollen tube that will deliver the enclosed sperm cells to the female reproductive cells deep within the carpel.

A carpel is composed of a stigma, the receptive surface where pollen adheres, the narrow style which leads to the ovary and the ovary. Enclosed within the ovary are the ovules which will become the seeds after fertilization. Each ovule has an embryo sac which consists of a number of cells including the egg cell and the central cell.

Fertilization in flowering plants has two fusion events which lead to the production of a seed. Nuclei (the cellular site of the majority of genetic material) of the two sperm cells within the pollen tube each separately fuse with nuclei of the egg cell and the central cell to produce the embryo of the seed (fertilized egg cell) and the endosperm (fertilized central cell) which will nourish the seed during its development and the seed coat surrounding the embryo and endosperm.

Pollination and subsequent hormonal changes associated with seed development cause the ovary to develop and ripen into a fruit.

Cherimoya Flower Structure

Cherimoya flowers are similar in structure and function to the generalized flower described above, yet they have some unique features (Fig. 2., Diagram of a Cherimoya flower).

Figure 1. Diagram of cross section of a typical flower.

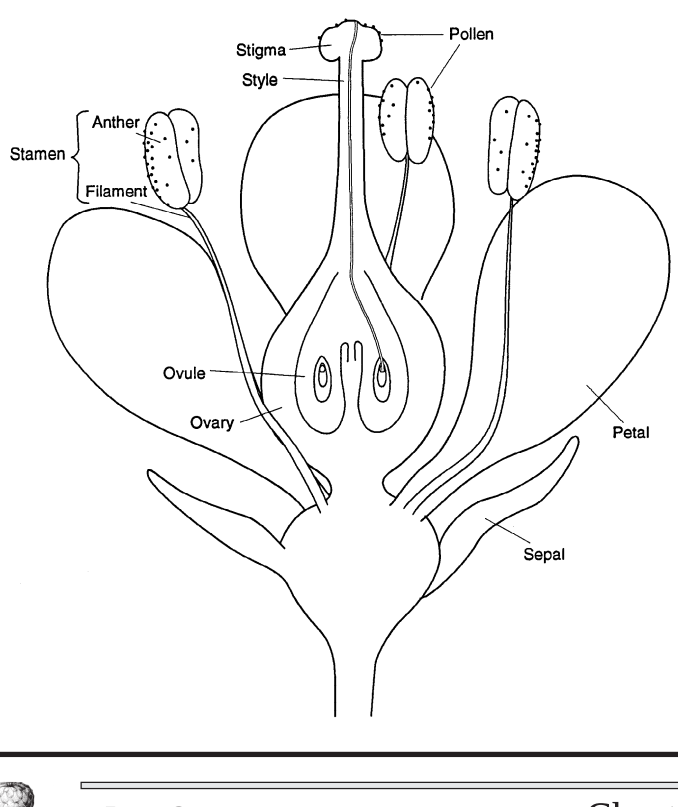
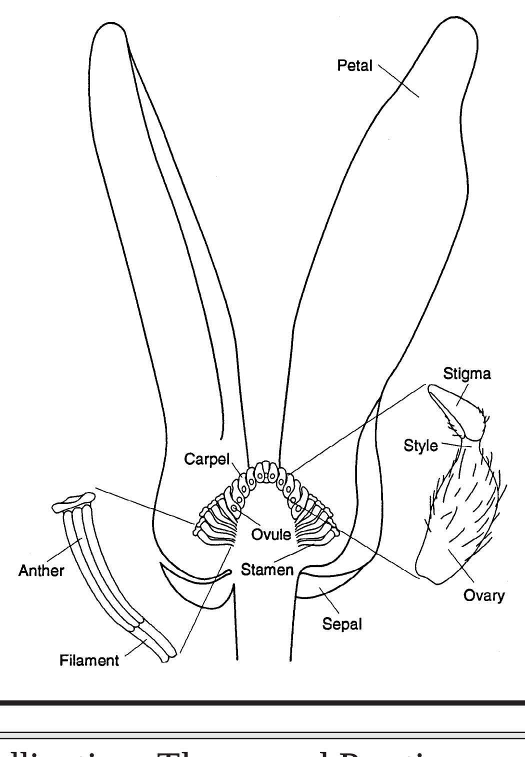


Figure 2. Diagram of cross section of a typical cherimoya flower, enlarged stamen and carpel.



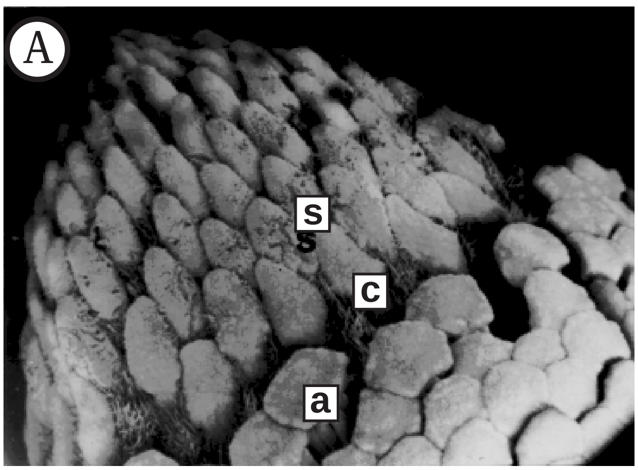
Cherimoya flowers are oriented so that the most inside of the four whorls of structures, or the carpels, are pointing downward. In other words, the flowers hang upside down on the tree.

A flower from a branch has been removed and turned so the cone-shaped female structure is pointing upward to easily identify the its structures.

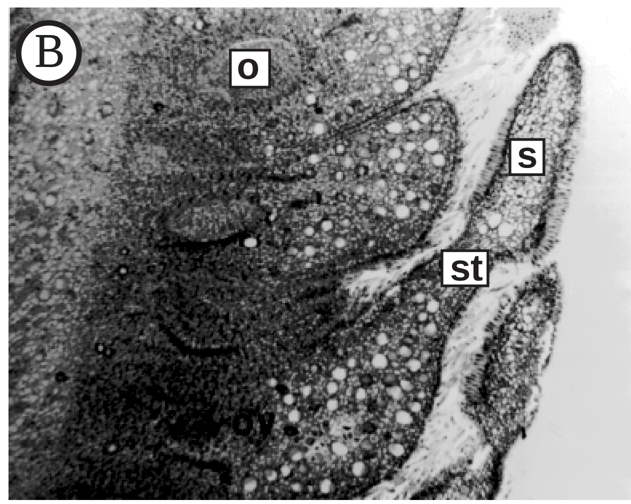
Each cherimoya flower has three small triangular green sepals at the base surrounding six petals arranged in two whorls. The outer three petals are light green in color, oblong in shape, and velvety in texture. The base of the outer petals are keel shaped. The inner three petals are also light green and very small; they may be absent in various cultivars of cherimoya (Wester 1910).

Figure 3. Scanning electron and light micrographs of multiple carpels and stamens showing the surface view. The many individual carpels form the cone shaped female structure.

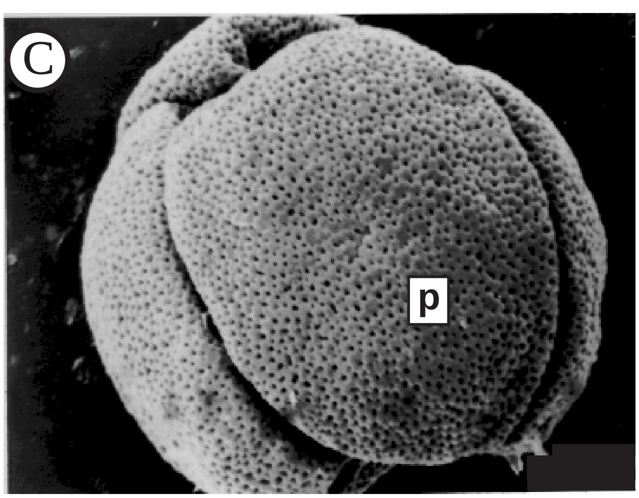
Only the stigmas (s) of the carpels (c) visible from the surface. The whorl of stamens below the many carpels are viewed on end with the "cap" of each anther (a) visible. 20 X magnification.



Light micrograph of a longitudinal section through one whole carpel and partially through three other carpels.



Scanning electron micrograph of pollen grains (p) united into a group of four grain called a tetrad. 150 X magnification.



Light micrograph of a longitudinal section through one whole stamen and partially through four others. Note the numerous pollen grains (p) within the anther (a) portion of the stamen. 55 X magnification.

Note the stigma (s) also seen in face view in figure A and the style (st) and ovary (o) of the one whole carpel and an ovule in the top partial carpel. 55 X magnification.

Inside the petals of a mature closed flower are many white stamens spirally arranged around the base of many carpels (Fig. 3A). Each stamen contains many pollen grains united in groups of fours called tetrads (Fig. 3C and 3D).

The many individual carpels are arranged as a spiral on the surface of an elongated and tapered structure called the receptacle which together form the cone shaped female structure which points downward when the flower hangs on the tree (Fig. 3A and 3B) (Wester 1910).

Each carpel contains a separate stigma, style, and ovary (Fig. 3B). Each carpel contains a single ovule, although infrequently two ovules develop within a single carpel (Schroeder 1951).

Upon fertilization, the fruit of the cherimoya, a syncarpium, is formed by fusion of the carpels into a solid mass (Schroeder 1951).

A cherimoya fruit will consist of between one and 200 carpels depending upon the number of successful fertilization events since the wall of the carpel will expand only in those carpels which set seed (Figure 4) (Schroeder 1951).

Flowers of the sweet sop, *Annona squamosa L.* are very similar in structure to that of the cherimoya. The most pronounced difference in flower structure between these Annona species is the length of the petals. Sweetsop flowers have shorter petals than cherimoya flowers (Wester 1910).

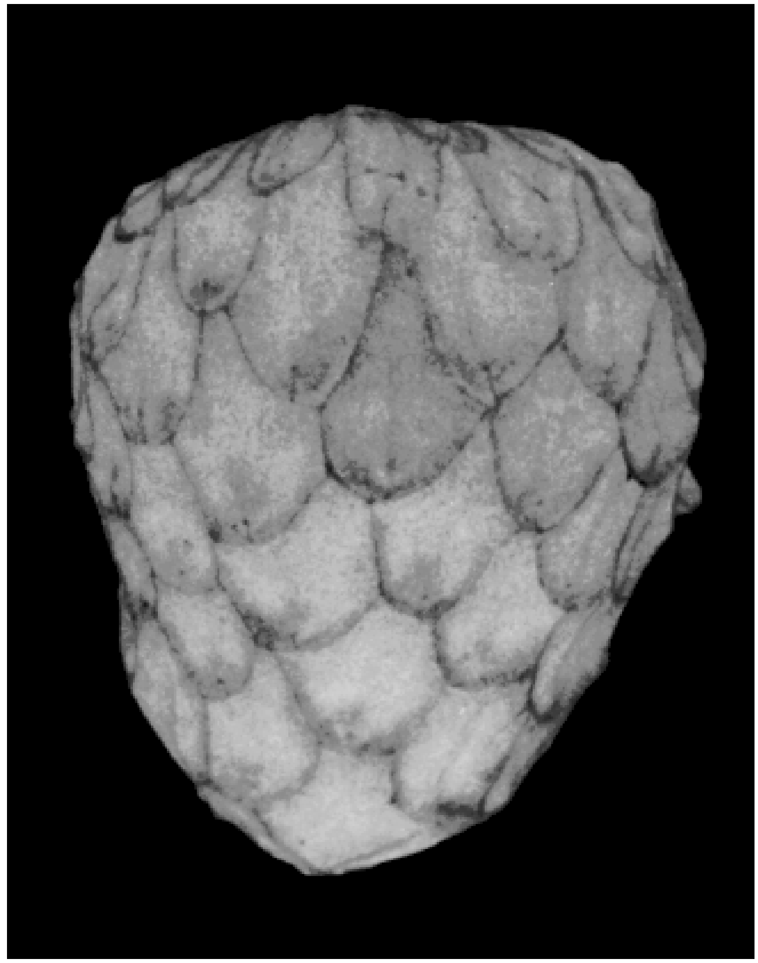


Figure 4. Cherimoya fruit whose carpels have uniformly expanded as a result of successful pollination.

The Timing of Flowering In Individual Annona Flowers

Flowering Season

The flowering season of cherimoya flowers varies in different parts of the world. In Sangareddy, India cherimoyas have two blossoming periods with a profuse flush during the summer (June to October) and a second one in the rainy season (March to mid May).

Only one crop is produced each year from the summer bloom, which matures between November and January (Venkataratnam 1958). In New Zealand flowering of cherimoyas occurs from December through January depending upon the cultivar (Richardson and Anderson 1990).

In Southern California, the flowering period lasts from early to mid June until late August or early September. For all growing regions of cherimoyas, flowering is associated with the vegetative flushing pattern (George and Nissen 1988; George and Campbell 1991), but environmental conditions also effect flowering.

High vapor pressure (1.2 k Pa) and soil moisture stress (-2.0 Mpa) particularly at high temperatures (28 degrees C) significantly reduces flowering and fruit set (George and Nissen 1988).

Although the flowering period extends over several months in all parts of the world, the quality of flowering varies during this period.

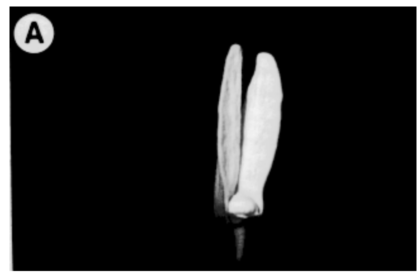
Researchers in Chile have found that the first flowers to open gave poorer fruit set with hand pollination than the ones that open later in the season. Flowers that opened earliest generally released pollen grains in an immature stage of pollen development (Saavedra 1977).

These immature pollen grains had thick walls abundant starch and did not germination (Saavedra 1977). Pollen of flowers that opened later in the season had no starch grains, greater cellular activity, and a higher proportion of germination (Saavedra 1977). In addition, there are cultivar differences in the seasonal blooming behavior of cherimoya flowers.

A study by Yonemoto and Yamashita (1990) conducted in Japan with three cultivars of cherimoya ('El Bumpo', 'Pierce', and 'Sabor') found that the number of open flowers varied considerable over the season.

Two of the cultivars had peak periods of flowering, 'El Bumpo' and 'Sabor', but the third, 'Pierce', showed fairly consistent numbers of flowers opening over a 5 week period in the middle of the flowering season (Yonemoto and Yamashita 1990).

Figure 5. Three cherimoya flowers, each with one petal removed to facilitate viewing the multiple carpels (top cone shaped structure) and multiple anthers below during three stages of flowering:



(A) female phase day 1

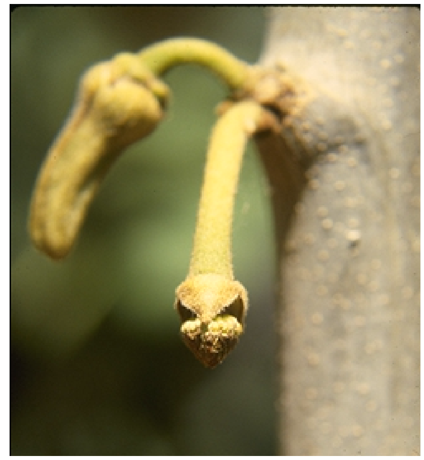
(B) female phase day 2

(C) male phase flower day 2. 10 X magnification

Figure 5. The development of a cherimoya — flowers to fruit.

Vegetative and flower buds appear after the leaf petiole falls naturally or is removed by physical/chemical methods.

A fleshy flower petal has been removed to show the rows of male and female flower parts.



Flower petals have fallen. If pollination has occured, the fruit will begin to develop, otherwise the flower will dry up and fall off.

Multiple flower buds can occur at a site. The flowers mature over a period of several weeks.

The flower has been pollinated and the fruit is beginning to develop. The next few weeks are critical as the fruit can still abort.

After several weeks, the fruit has increased in size and should grow into a mature fruit. Flowers may continue to develop at the same site for several weeks. The excess flowers can be gathered as a source of pollen.

Insect Pollinators

Natural Pollination

Cherimoya, sweetsop and most of the 2300 species in the *Annonaceae* plant family have flowers which are pollinated by beetles (Gottsberger 1988). These plants have floral characteristics common to beetle pollinated plants.

The flowers are closed flowers with characteristic odors, feeding tissue (specialized tissue which are eaten by beetles) and the flowers have plant parts that protect the reproductive parts (Gottsberger 1988).

Members of the Annona family with large flowers are mostly pollinated by large Scarb beetles, but those with small flowers like cherimoyas, sweetsop and atemoya attract small beetles like those in *Nitidulidae* or *Staphylinidae* families (Gottsberger 1989).

A study of the role of nitidulid beetles in natural pollination of atemoyas in Israel found that four species of nitidulids (*Carpophilus hemipterus L., Uroporus humeralis F, C. mutalatus Er.*, and *Haptoncus luteolus Er*) were common visitors throughout the flowering season (Gazit et. al. 1982).

Additional experiments were conducted to determine if these beetles were just visitors or actually pollinators. Flowers containing nitidulid beetles and selected flowers without beetles were marked at four commercial orchards then later reexamined for evidence of fruit set.

Based on this experiment and an additional one where the four species of nitidulid beetles were trapped in insect proof plastic nets on flowering twigs, they concluded that fruit set depends on the presence of nitidulid beetles, but that there was no difference between the four species of beetles in their efficiency to pollinate atemoyas (Gazit et al. 1982).

In Florida nitidulids were the most common (51.4%) visitors to atemoya flowers of the 7 orders and 13 families of insects found outside or inside petals. Of the 9 species of nititulid beetles found to visit atemoya flowers, *C. fumatus* was the most common (Nagel et. al. 1989).

To test whether the nitidulid beetles were important for pollination, atemoya trees were caged and treated with malathion before the experiment to remove potential insect pollinators, two cages were supplied weekly with nitidulids and two were sprayed weekly with malathion as a control.

Tagged flowers were monitored for fruit set and those caged trees supplied with nitidulids had significantly higher fruit set than caged trees without nitidulids, indicating that these species of beetles do contribute to atemoya pollination (Nagel et. al. 1989).

Another study in Israel found that there was an increase in the number of nitidulid beetles in atemoya flowers as the flowering season progressed and that the nitidulid beetles were most active during the early morning and late afternoon when female phase flowers started to switch to the male phase (Podoler et al. 1984).

Nititudlid beetles were found to be associated with fruit set of atemoyas and cherimoyas in Australia, Chili, and the United States (Florida) (George et. al 1988; Laport and Dent 1992; Nagel et. al. 1989). In Australia, fruit set has been shown to increase linearly with increasing numbers of nitidulid beetles *(Carpophilus hemipterus L.)* per flower (George et al. 1988).

In addition they found that the number of nitidulid beetles per flower was greater with higher soil temperatures, but that rainfall greater than 5 mm per day reduced the number of nitidulid beetles per flower (George et al 1988).

Last spring, growers in the Carpinteria region of California noticed many fruit on regions of cherimoya trees where flowers were not hand pollinated and after closer examination a number of small beetles were found in flowers.

These beetles were keyed out and tentatively classified in the *Staphilinidae* family within the genus *Eusphalerum*. Beetles in this family have been previously found in cherimoya and atemoya flowers in Chile and the United States (Florida) (Laport and Dent 1992; Nagel et. al. 1987).

In many parts of the world where cherimoyas, atemoyas, and other commercial *Annonas* are grown, all fruit harvested are fruit set without hand pollination either by self pollination or beetle pollination.

In California, most spontaneous cherimoya fruit set is probably due to rare overlap of male and female phases within individual flower resulting in self pollination of an individual flower (Ellstrand et. al. 1991). Many of these fruits are misshapen due to insufficient pollination.

Only those carpels that are pollinated and set seed will expand, so insufficient amount of pollen or uneven distribution of pollen over the stigma surfaces of the many carpels of the flower cause the fruit to be misshapen.

In regions where cherimoyas, sweetsops and atemoyas are grown, the lack of or extremely low population levels of *Nititulid* beetles or other pollinators to transfer the pollen from male phase to female phase flowers causes a reduction in fruit yield (Richardson and Anderson 1990).

Experiments were conducted in Australia, U.S. (Florida), Chile, and Israel using rotting fruit placed at the base of cherimoya and atemoya trees to encourage nititulids to visit flowers and increase the amount of natural pollination and subsequent fruit set. These experiments have been met with limited success. Fruit set percentage was shown to be greater than no pollination, but still not as great as hand pollination.

However, in Spain, insects in the genus *Orius*, a known pollinator of cherimoyas, existed in large populations in corn. A series of experiments were conducted to test whether interplanting corn with cherimoyas would promote insect pollination by beetles in the genus *Orius* (Hermoso et. al. 1993).

They found that the population of *Orius* sp. in cherimoya flowers increased when the interplanted corn had the corn ears removed. Moreover there was a good correlation between the number of pollen tetrads on cherimoya stigmas and fruit set when corn plants were grown at a distance from cherimoyas (Hermoso et. al. 1993).

Yet cherimoya pollination by *Orius* beetles is dependent upon environmental conditions since they found that the development of *Orius* is greatly affected by high winds reduceing the number of insects per flower and the amount of fruit set (Soria et al. 1993).

In addition, they demonstrated that interplanting corn with cherimoyas can double yields if grown in areas that are not windy (Farre personal communication).

Although in many parts of the world fruit set can occur in the absence of hand pollination, hand pollination is still the most reliable method for obtaining uniformly sized fruit set (Richardson and Anderson 1990).

Hand Pollination

Commercial Cherimoya Growers Hand Pollinate Their Crops

Hand pollination of cherimoya and atemoya flowers is now a common practice in many commercial production regions in the world (Kahn et. al. 1991; Richardson and Anderson 1990). Hand pollination has a number of advantages in addition to increased yield of uniformly shaped fruit.

Fruit maturation and consequently harvest date is dictated by the date of pollination so pollination by hand allows growers to spread the harvest period according to market requirements, manipulate crop load of individual trees and position of fruit within the canopy of the tree (Richardson and Anderson 1990).

Schroeder was the first person to develop a viable method for hand pollination. This method was adopted and later modified by growers and researchers in the U.S., Chile, New Zealand, Israel, Spain and Australia (Hopping 1982; Richardson and Anderson 1990; Kahn et. al. 1991; many others).

The basic method described by C.A. Schroeder involves collecting flowers which are about to shed their pollen (3 to 6 p.m.), then separating the pollen from other flower parts and storing it in a vial until the next morning when it is applied to the female receptive surface of open flowers with a small paint brush (Schroeder 1942, 1971, 1988).

This method is effective, but a number of recent studies on aspects of hand pollination suggest modifications and ways in which you can be flexible about aspects of hand pollination. Kahn has divided these studies into three sections:

1. Collection and storage of pollen from male phase flowers.
2. When and how to hand pollinate female phase flowers
3. Methods of applying pollen.

When to Hand pollinate female phase flowers

Since flowers are in the female phase from the time they are full sized mature flowers (approximately 24 hours prior to opening) until male phase starts approximately 36 hours after petals open,

There is a large window of time in which hand pollination can be done. However, some periods of time are clearly better than others.

The flowers appear to be most receptive during the morning of the first day of the female phase soon after the petals have begun to separate. However, since pollen is not shed in the morning, pollen with high viability is not available in the morning unless it has been stored overnight.

A study by George and Campbell (1991) indicated that it is best to pollinate flowers which are just beginning to open in the early morning and late afternoon with relative humidity above 70% (George and Campbell 1991).

Fruit set is reduced if flowers are pollinated during the hottest period of the day. (George and Campbell 1991). Hand pollination experiments conducted at the Kerikeri Horticultural Research Station in New Zealand using the cultivar Reretai compared morning pollinations of newly opened flowers (Day 1) pollinated with pollen stored over night to evening pollinations (Day 1) with freshly shed pollen from different flowers in male phase.

Their results indicated that evening pollinations yielded significantly greater percent fruit set and more perfectly shaped fruit (Richardson 1990). However, there is no reason one can not produce perfectly shaped fruits by pollinating in the morning with pollen and anthers from either female phase or male phase flowers that have been stored overnight.

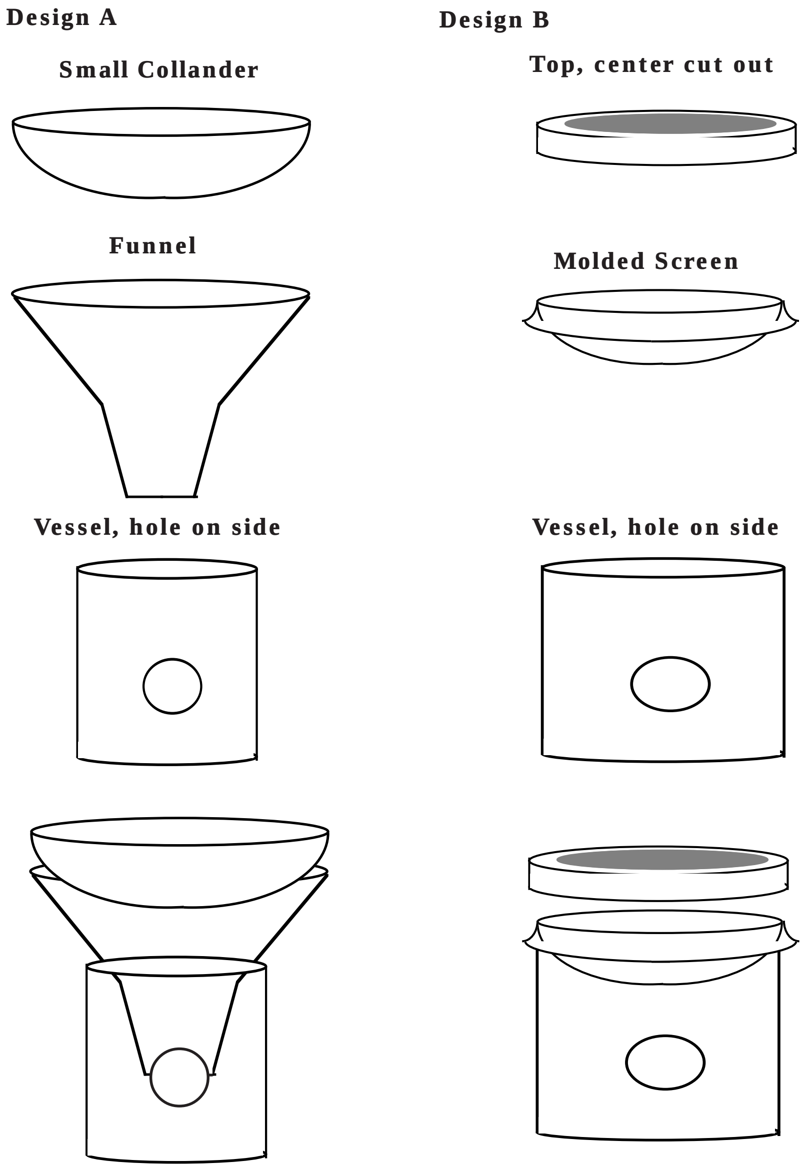
Pollen storage does reduce the number of pollen grains that are able to germinate and the best conditions for storage in Southern California are not known, but one can adjust for these uncertainties by using more pollen (and or anthers) to hand pollinate than one would when using fresh pollen.

Methods of applying pollen

There are a number of methods used to apply pollen on to the stigmas of the female portion of the flower. The two most common methods are the use of small paint brushes or a pollen "puffer" or "gun". A commonly used brush for hand pollination of cherimoyas is a 1/8 inch diameter artists brush with camel's hair bristles.

Two versions of the Condor Cup, a pollen collection device.

Exploded view of two versions of the Condor Cup.
Design A benefits from a wider top than bottom.
Design B is easier to make.



The Collection Device

The Condor Cup is held by the lower two fingers of one hand against the heel of the hand. The fore and middle fingers are used to pull petals off.

The Condor Cup takes 1-2 seconds to collect a flower's pollen, and less from flower clusters. A brush takes 6-7 seconds and more practice, and a lot of pollen gets blown away.

The Condor Cup loses little pollen since the flowers are collected right on the screen. Fingers swirl the petals from a few flowers to release their pollen. It also permits collection of more pollen, since many flowers are within the reach of a single outstretched hand, but are not within the reach of two hands.

We have not examined the effect of petal removal on efficiency of set, but feel that it might reduce self-pollination. However, faster pollination using the Condor Cup more than offsets any lower number of fruit set as a result of pollen collection using this method.

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Some growers who hand pollinate with small brushes choose to cut the bristles in half to make them stiffer. If one chooses to use a small brush, it is important to carefully separate the three petals with one hand and gently brush the pollen over the stigmas of the cone shaped female portion of the flower using a circular motion, then close the petals on the brush and slowly remove it from the flower so that as much pollen as possible is retained on the female receptive surfaces (Anderson and Richardson 1990).

It is also important to mark the stem of the flower with a water proof marker as a means of monitoring hand pollination and to prevent harvesting pollinated flowers as a pollen source (Richardson and Anderson 1990).

Pollen "guns" or "puffers" are also commonly used to hand pollinate *Annona* flowers. Two commonly used types of pollen "guns" in Southern California are called "superfrut" (Distributed by J.E.L. 033-312536 Quillota Chile) and "Max" (Kurita Tabaco Ikubyoo Ikufu Company 1636 Horigome-cho, Sano-shi, Tochigi, Japan. Phone: 0283-22-3101). In order to compare the efficiency and effectiveness of hand pollination using a puffer or brush method,

Richardson and Anderson (1990) tested three methods of hand pollination. They compared pure pollen or pollen/*lycopodium* mixture using a brush and the pollen/*lycopodium* mixture using a hand puffer. *Lycopodium* powder is the spores of a primitive nonvascular plant in the genus *Lycopodium*. *Lycopodium* acts as an inert dilutent for pollen. They found that the puffer application of pollen/*lycopodium* mixture produced significantly higher fruit set than the other two methods, but fruit shape did not differ significantly between the three methods.

Using the brush applications alone, they found that the pollen/lycopodium spore mixture showed improved fruit set over pure pollen. The puffer application was also more efficient than the brush method producing 5 fold increase in number of flowers pollinated per unit time.

The puffer requires pollen to be mixed with a dry media. Sainte Marie (1987) also compared the use of brushes and pollen "guns" for hand pollination as well as the effect of different pollen dilutents such as *lycopodium* spores or PVC (polyvinyl chloride) powder ("Geon 213" made by BF Goodrich Co. Geon Vinyl Division, Cleveland, Ohio 44131) on the percentage of fruit set and fruit shape.

The pollen "gun" produced better shaped fruit and heavier fruit when combined with a pollen dilutents than the brush method. The brush method of hand pollination is only recommended when pollen is scarce (Sainte Marie 1987).

The pollen dilution found to be most effective in combination with a pollen "puffer" is 2/3 pollen and anthers and 1/3 pollen dilutent such as PVC powder or Lycopodium spores. At Cal Tropics in southern California cherimoya flowers are hand pollinated with a pollen "puffer" containing pollen and anthers mixed with an equal volume of PVC (polyvinyl chloride) powder. The anther mixture is placed in a pollen gun for hand pollinations the same day (Tony Brown personal communication).

Therefore, there are a number of options one can select from when choosing when and how to effectively hand pollinate cherimoya and other related annona flowers. Some may be more effective for some people than others, so it is important to keep records on how and when pollinating to evaluate how effective the methods are in respect to fruit set.

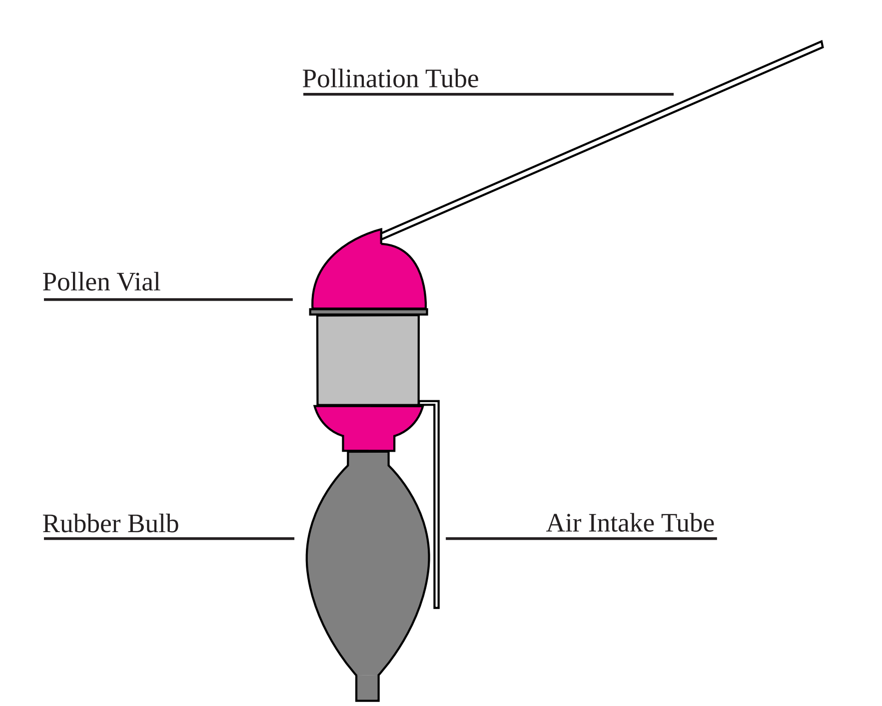
Summary

Plants in the Annona family of plants have fascinating and intricate floral biology due to the presence of protogynous dichogamy, floral synchrony and beetle pollination.

Currently, in most parts of the world, the absence or low number of beetles in species capable of pollinating cherimoya, sweetsop and atemoya flowers and the presence of protogynous dichogamy causing male and female functions to be separated in time necessitates hand pollination as the most reliable method for obtaining uniformly sized fruit.

Inorder to effectively hand pollinate flowers of cherimoya and related species, knowledge of floral structure and the timing and visual characteristics of the female and male phases of flowering are essential. This chapter details the theoretical and applied aspects of pollination of cherimoyas and related species.

Pollen Puffer



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Giving Nature A Hand

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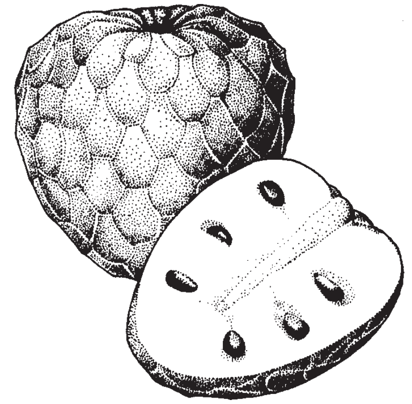


The cherimoya is a superb fruit with a major problem — It refuses to set good fruit naturally.

Hand pollination of cherimoya (Annona cherimola) is essential to set a good crop of fruit. Depending on the cultivar grown, cherimoya trees will flower from December through to February (In California trees flower from June through to August).

A five year old tree may produce several hundred flowers throughout this period. Under natural conditions less than 5% of flowers set fruit, however, if the hand pollination technique outlined below is employed, 60 - 90% fruit set can be achieved.

Often, naturally set fruit are grossly misshapen and therefore unsalable. Hand pollination ensures that the majority of fruit produced are of good shape and size, and consequently a markedly improved economic return.

A basic understanding of cherimoya flower anatomy and physiology is useful if hand pollination is to be efficiently employed. The flower consists of three long fleshy petals which enclose a small cone-shaped structure (the stigmas or female flower parts) surrounded by a white collar (a complex of stamens or male parts) as shown in Figure 1.

Figure 1

The male and female portions of the flower are both functional, but do not mature at the same time. As the flower opens the female stigmas are sticky and receptive but pollen is not shed from the stamen until about 36 hours later.

By this time the stigmas have dried out and can no longer be pollinated. It is highly unlikely that pollination will occur within a single flower. Successful pollination under natural conditions will depend on pollen moving from one flower to another.

However, pollen transfer between flowers is also extremely poor, as the flowers are not attractive to pollinating insects and access for insects to the receptive stigmas is restricted due to the flower shape (Figure 1).

The structure of the flower and nature of pollen grains also precludes pollination of flowers by wind-borne pollen.

Hand pollination is really the only way to achieve successful cherimoya fruit production.

Hand pollination was first proposed by Art Schroeder in 1941, and has been successfully employed by cherimoya growers around the world, including New Zealand.

The equipment needed for hand pollination is both simple and inexpensive. A paper bag, tissue paper, a small camel hair artists brush, a small lidded vial and a black permanent marker pen.

Flowering takes place over a three-week period and pollination of receptive flowers should be carried out every 3 or 4 days. With a little practice, one person can pollinate about 150 flowers per hour, so the task is not as arduous as it first seems.

Further research is now underway to determine more efficient and economic methods of hand pollination, including the use of hand-held puffers and pollination timing.

Step-by-step Directions For Hand pollinating cherimoya flowers.

Step 1

Flower Structure: The flower consists of three long fleshy petals which enclose a small cone-shaped structure (the stigmas or female flower parts) that is surrounded by a white collar (a complex of stamens or male parts).

Step 2 Pollen Collection: Flowers with petals partially open, but still firmly intact, should be picked in the late afternoon around 4 p.m. and immediately placed in the paper bag.

These flowers are at the end of the female stage. Flowers should be harvested from limbs either close to the ground or at the top of the tree, where fruit production is undesirable because of excessive blemish. Care should be taken to avoid

Step 3 Over-mature Flowers: Flowers that are fully open, so that the petals no longer cover the female cone should not be harvested.

At this stage flowers have been in the male stage for 24 hours and pollen has been released or is no longer viable.

Step 4 Flower Storage: Once sufficient flowers have been collected (20 will provide enough pollen to pollinate 60 flowers), a moist (not wet) tissue is placed in the bag. The bag should be kept open, and placed at room temperature overnight.

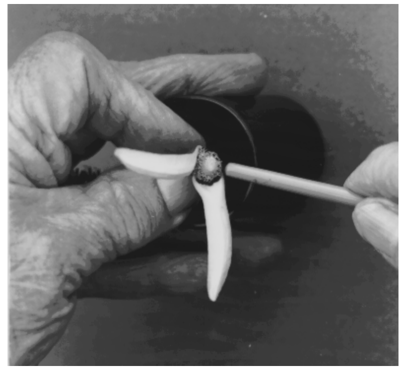
This creates a sufficiently humid environment within the bag for pollen maturation and release, as well as preventing the pollen from drying out.

Step 5 Pollen Extraction: The following morning, preferably around 7 a.m. the flowers should be removed from the bag. Use the tip of a paint brush handle to scrape the pollen sacs from the ring above the stigma cone. If the pollen sacs are difficult to remove and appear rubbery, then the pollen is too immature and should be discarded.

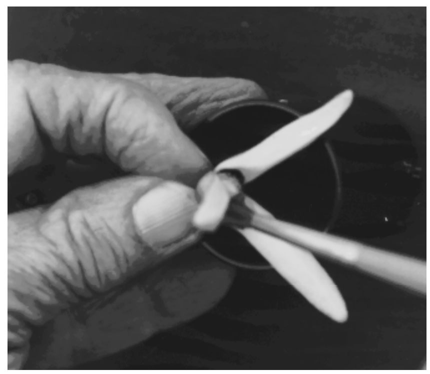
Step 6 Pollen Storage: The pollen sacs and grains should be placed in a small airtight vial.

Step 7 Flower Selection: Only slightly open flowers in the female stage of their development should be selected for pollination.

Step 8 Pollination: Dip the paint brush into the pollen and then close the vial. Carefully separate the three petals with one hand (being careful that the flower is not damaged) and then, using a circular motion, brush pollen over the tip of the female cone.



Step 9



Brush Removal: Close the petals on the brush and slowly remove it from the flower so that as much pollen as possible is retained within the flower.

Step 10 Marking Flowers: Following pollination of a flower, it is a good idea to place a small, easily visible mark on the flower stem, using a black permanent marker pen. This only takes a little extra time, but reduces the chance of flowers that have been pollinated being harvested for pollen. It also allows growers to follow the development of pollinated flowers through to the harvest to prove that the time involved was well spent.