PROBLEMS AND TECHNIQUES ASSOCIATED WITH NATURAL AND CONTROLLED POLLINATION OF BLACK CHERRY (*Prunus serotina* Ehrh.)

Donovan Forbes[1]

In the Tennessee Valley Authority black cherry tree improvement program, first work has been concentrated on seed source studies, selecting plus phenotypes, and establishing both clonal and seedling seed orchards. But the problems associated with natural and controlled pollination have been recognized and work is under way toward their solution. Problem areas include natural selfing; natural pollination; and requirements for controlled pollination, including flowers and flowering, pollen collection and storage, bagging, emasculation, and fruit collection.

Natural selfing - Natural selfing needs early exposure here since its presence will considerably alter the breeder's approach. My experience is limited to two populations, one in north central Florida an the other in east Tennessee. In the former, several thousand flowers were isolated prior to opening to learn whether selfing was a characteris of this population. Only three natural selfs--flowers allowed to self within a bag--set fruit. These dried and dropped inside bags before maturity. In east Tennessee, however, 21.5 percent of natural selfs on six trees set fruit. Some fruit was removed while green and examined. Thirty percent of these appeared normal. The others were either empty or had shriveled seed coats. All remaining selfs were debagged to prevent heat effects but dropped unexpectedly about two weeks before adjacent natural crosses and were lost. This season I again tested the east Tennessee trees and had hoped to produce some seedling selfs. All four trees tested did produce selfs. They progressed well for about two weeks but dropped very early inside bags. It is possible that all selfs drop early as a result of complications of selfing. However, there are two other reasons that probably would have caused the drop. First, an insect, probably either a cherry fruit fly (*Rhagoletis* sp.) or the plum curculio (*Conotrachelus nenuphar*) attacked all study trees and caused about 80 to 90 percent of all fruits to drop. Second, bagged leaves and fruits dried noticeably during the first hot days in May. Most dropped fruits inspected showed insect or heat damage or both. But many unbagged insect-pollinated fruits dropped before the onset of hot days and were no insect infested. This drop is similar to the early drop in horticultural varieties and could have been due to a nutritional deficiency.

With those trees that do self, only one situation is potentially dangerous to a breeding program. It arises when seed development through germination is indistinguishable from crosses. This will not be a great problem provided self and cross seedlings are visibly different at some fairly early stage of development. If seedling selfs are not different, I would exclude the mother tree from a crossing program to prevent injecting a self-compatible allele into the breeding population.

From a positive standpoint, the presence of self-compatibility opens another door for the cherry breeder, the possibility of pure-line breeding I won't discuss the possibilities, but they may be especially important i selfs grow well and are fairly easy to maintain.

[1]Tennessee Valley Authority, Norris, Tennessee.

Flowers, flowering, and natural pollination - The following description of flowers, flowering, and natural pollination originated from observations on the two populations described earlier. These may vary slightly from those in other areas.

Flower racemes occurred with near equal frequency from top to bottom and on all sides of open-grown trees. The small perfect flowers were about 1/4 inch in diameter when fully opened. The single pistil was surrounded by 15-21 stamens. Normally the 50-70 flowers borne on a single raceme opened over a 2-5 day period. Opening proceeded from base to end of raceme, but some lower flowers frequently opened later. Imminent opening was indicated by swollen or slightly lifted petals. In all cases a flower would open within two hours after this condition was reached. About 1/4 to 1/3 of flowers were in a receptive condition once flower opening was under way on a given raceme. A large tree would usually flower over a 12 to 2-week period except during a very hot or very cold spring when the interval was shortened on lengthened. Flower opening generally progressed upward from base to top of the tree, a characteristic most easily observed in large trees.

Natural pollination was primarily dependent upon weather conditions and insect pollinators, principally the honeybee. Observations over three growing seasons made clear the need for warm days to achieve a heavy fruit set. Warm days not only stimulated flowering but also promoted insect activity. Notice that I said warm, not hot. Best fruit set occurred during a season with cool nights and warm days, no higher than 80^{0}F. A sudden warming that brought on flowering followed by several cold days just after flower opening practically eliminated fruit set because of reduced or complete lack of insect activity. Prolonged high temperatures had less effect because there was some insect activity, but flowers opened so fast that many must have gone unpollinated resulting again in a reduced set. Heavy and continual rain over the flowering period had much the same effect as warmth followed by cold.

Controlled pollination - Large-scale controlled pollination in black cherry is severely handicapped by several factors: First, pollen is of the sticky type and is not easily adaptable to high speed methods as is the wind-carried type; second, flowers are small and delicate and not subject to easy and successful emasculation; third, flower opening proceeds in general from base to tip of raceme, which allows only 1/4 to 1/3 of a raceme to be pollinated at one time; and fourth, fruit may be lost to bird pilferage and from natural abscission at maturity unless bagged until harvest. Most important plus factors are abundance, regularity, and precociousness of flowering. Seedlings flower as early as grafts, some after only one year. Flower crop size varies from year to year, but there always seems to be a fair one provided there is no frost kill.

Two general pollination methods I have successfully used with black cherry are: (1) application of pollen collected on free anthers and (2) use of an entire raceme of freshly opened flowers as an applicator. Under the first method, flowering twigs are brought in for pollen collection, placed in water, and isolated. Opened flowers are then removed and the remaining ones allowed to open. When about 1/3 or more are open, a raceme is removed and placed on a screen with others, then rubbed lightly with

another screen (figure 1). Anthers break off and drop through the screen into a collection vessel. These free anthers can be air-dried until they dehisce or can be refrigerated in a dessicator. They can be successfully stored a number of ways soon to be described in a publication on work done at TVA's forest physiology laboratory. Best viability retention was found to be at -30° C, while at 4° C a moderate reduction occurred. Sealed vials made excellent storage containers.

In the first method--using pollen collected on free anthers--either previously opened or unopened flowers can be used, but the process varies from one to the other. When bagged but already-opened flowers are pollinated, pollen applicators should not be returned to pollen containers unless new containers are used for each mother tree. This avoids mixing pollens. When unopened flowers are used, pollen applicators can be reused and returned to pollen containers, because stamens are not yet unfolded and anthers have not dehisced.

With opened flowers I have successfully pollinated using the following procedure: The bag is removed and pollen applied to all receptive stigmas. Flowers that are past their peak receptivity will have a dark spot on the style and show a visible drying of flower parts. A camel's-hair brush or any other small applicator that will hold pollen is effective. I regularly use a 1/4 x 4-inch dowel covered on one end with parafilm. This plastic holds pollen well and clings to the dowel. Either the parafilm is changed when another pollen charge is needed or pollen containers changed when moving to another mother tree. All pollinated flowers are rebagged.

With unopened flowers, bags are not put on until after pollination. First work on a tree is raceme selection. I try to choose a stout raceme-bearing limb that is as upright as possible; this is especially important if water-proof bags are used, because they may droop and become partially filled with water. All opened flowers are then removed. Remaining buds will be of two types, those that are about to open--an evidenced by swollen or slightly lifted petals--and those that are tightly closed. On those flowers that are about to open, petals are removed by grasping them with tweezers. With a little practice all petals can be removed at one time. This leaves the stigma exposed and all anthers neatly tucked away.

Anthers starting to lift are no problem; they will not begin dehiscing until an hour or more after full elongation. Each exposed stigma is touched with a pollen-laden applicator then all flowers bagged. Flowers pollinated this way may not be immediately receptive, but <u>will be</u> very soon after pollen is applied.

In the second pollination method, pollen is collected by bagging unopened flower-bearing racemes then removing the entire raceme-bearing stem after flower opening. The bag is retained and the freshly cut stem placed in water; milk bottles in racks make good containers. Several tagged limbs can be carried in each rack and used when required. Individual racemes are removed from a bag by slitting it, excising a raceme and removing it, then resealing the bag with masking tape.

Flowers to be used as females are also bagged prior to opening. After flower opening has reached its peak, the bag is removed and male parent flowers are gently rubbed over the female flowers. Rebagging follows.

Emasculation - Emasculation is a questionable practice with black cherry for two reasons: First, flowers are small and delicate, making anther removal difficult--often the style is broken or cut--and, second, success leaves a weak, unprotected style that may be either broken or lost during the bagging process or afterward. I have ruled out its use except in a greenhouse situation with plants known to self.

Bagging - Bagging is not a simple process on black cherry primarily because most flower-bearing limbs are slender and weak and any excessive heat buildup inside bags will kill developing fruit. As a result, bagged limbs often must be supported, and cool bag material should be used. I have used synthetic sausage casing and Teryline but have found only the latter acceptable for more than a short period because of its low heat buildup and its water permeability. Sausage casing is acceptable if bags are removed shortly after pollination. What is really needed is a very light net-like bag that will stretch with twig expansion but still will exclude insects. Until this type bag is available, I suggest debagging two or three days after pollination and taking the risk of fruit loss.

For support, bagged limbs can be tied to a higher limb, provided it is attached to the same larger limb as the one bagged. This will reduce incidence of bag pulloff. The best system I have used so far involved first tying a 1/4-inch dowell alongside the bag then running a string from bag tip to a higher limb and back down to the dowel's base. The triangle produced is strong and practically eliminated bag pulloff.

Fruit Collection - Under natural conditions fruit ripens unevenly, even on the same raceme; this is expected because flowers open unevenly. But fruit ripening varies even more so than does flower opening. All developing fruit is first green, then goes from reddish-green to dark red, then black. In an earlier study, I found that dark red and black fruits produced seeds that germinated best, but that some seed from green fruits and many from light red ones also germinated. There will be some variation in controlled-pollinated fruits also, but all those pollinated together should be harvested when the first ones mature.

Insect-pollinated fruit should be collected on drop cloths. It can either be collected as it matures or all at one time. The first method will work in an orchard, but the second is preferred for widely scattered forest trees. The first guarantees mature fruit, but with the possibility of considerable loss. The second risks reduced germination, but makes up for this in time saved and complete collection of fruit.

SUMMARY

Several problems make controlled pollination in black cherry risky and reduce the odds on natural pollination to considerably less than a sure thing. We can't very well fight the weather and insect idiosyncrasies that control natural pollination, but there are some fairly good controlled pollination procedures that will produce acceptable results regularly. A

change here and there in materials and technique should guarantee predictably good results.

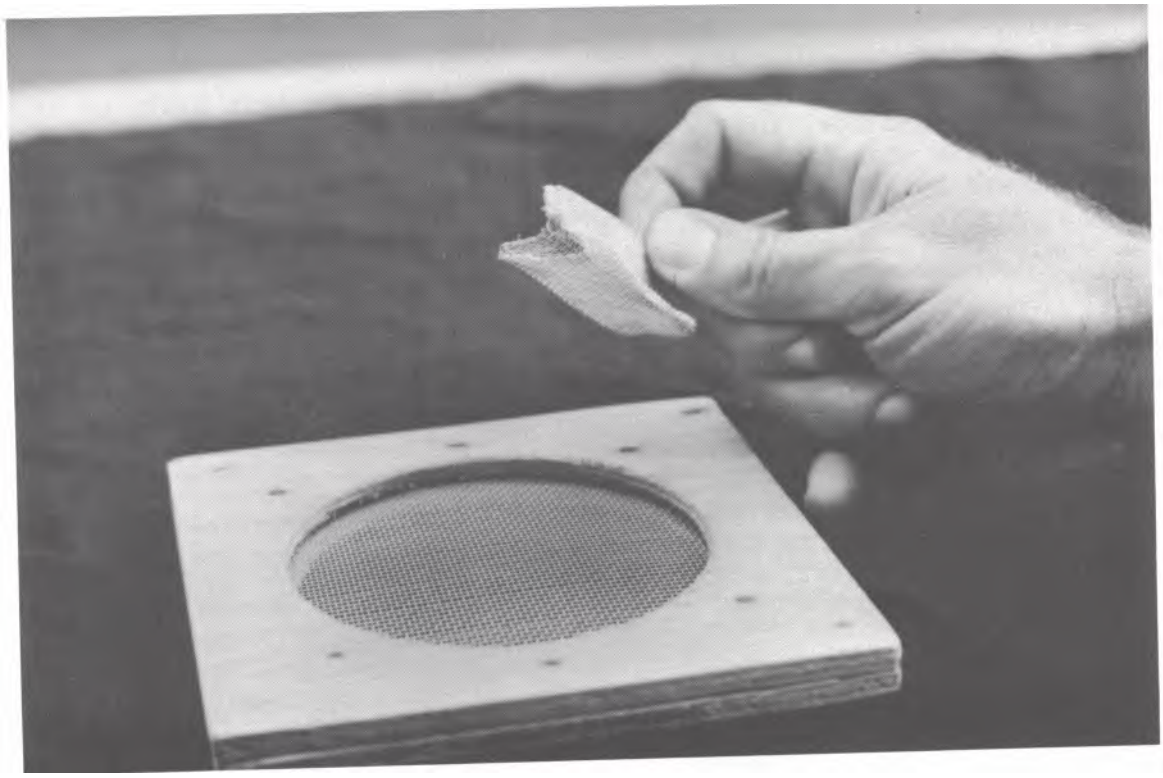

Figure 1. Screen technique used to collect anthers of black cherry.