INBREEDING EXPERIMENTS IN SUGAR MAPLE (ACER SACCHARUM MARSH) - - EARLY RESULTS

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Controlled pollination in sugar maple (Acer saccharum Marsh) may provide a method of developing new trees that have higher growth rates, improved form and quality, sweeter sap, and greater resistance to disease and insect attacks.

Basic to any pollination work is a thorough knowledge of the flowering and fruiting behavior of the species with which the breeder plans to work. For sugar maple, an unpublished exploratory study by the author in 1958, involving both field observations and a search of the literature on the species reproductive processes, pointed up the need for research on its flowering and fruiting behavior. That exploratory work also was helpful in outlining the objectives and procedures to be used in a second and more intensive investigation.

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This paper is a report on one phase the more intensive and more comprehensive second study. namely, on the incidence of self-fertility and the characteristics of selfed progenies in the early seedling stages.

LITERATURE REVIEW

Though the literature reporting on inbreeding studies among the angiosperms is extensive, relatively little of it deals with tree species. Concerning maples specifically, Wright (9), in a summary of tree breeding experiments at the Northeastern Forest Experiment Station, reported 4 percent filled seed from 49 selfed flowers of A. saccharum, 1, 9 percent from 100 A. rubrum L. flowers, and 4.8 percent from 231 flowers of A. platanoides L.

Piatnitsky (8) reported a high degree of self-fertility in A pseudoplatanus L, 65 percent of the fruit containing filled seed. In A campestre L. he noted that 27. 5 percent filled seed were set to self- pollination.

Among inbreeding experiments with other hardwood species, Nielsen and Schaffalitsky (7) noted that some trees of Fagus sylvatica L. were self-compatible to a limited extent.. Blinkenberg et al (2) made similar observations in later experiments with the same species. Tree-to-tree variation in self-compatibility was observed by Cram (4) in a study of the Russian pea tree (Caragana arborescens Lam). Guard (5) found that self-pollinated flowers produced one-third the seed set of cross-pollinated flowers in Liriodendron tulipifera L. Carpenter and Guard (3), working with the same species, reported that filled seed set to self- pollinations for 3 trees were 0. 0, 1. 4, and 5.6 percent of the fruit examined.

MATERIALS AND METHODS

Ten native, mature flowering trees were selected for use in this study Six of these were in the vicinity of Burlington, Vermont, and four were in Williamstown, Massachusetts. The two areas are about 140 miles apart. In blooming habits, two of the trees in each area were protandrous, and the others were protogynous.

Pollen Collection and Handling

Several flower-bearing branches, approximately 3 feet long, were taken from each tree during the first and second weeks in March at the Williamstown and Burlington locations, respectively. These were forced into bloom by placing them in buckets of water in a greenhouse under warm and humid conditions (75° F and 80 percent relative humidity)

After the flower buds had burst and the anthers had begun to yellow, the branches were reduced to small twigs and placed in beakers in such a manner that the pollen sacs dehisced on papers spread beneath. The pollen sacs have a tendency to dry up without shedding much pollen if allowed to remain on larger branches. Beakers were isolated as necessary to prevent contamination of different pollen lots during shedding and collection. The pollen was stored in glass vials at 30 percent relative humidity and 35° F.

Flower Isolation and Pollination

To isolate the female flowers from foreign pollen, transparent cellulose sausage casings were placed over the branches while the flower buds were still closed The mouth of each bag was bushed with non-absorbent cotton and secured with a piece of twine. For the selfing experiment none of the flowers were emasculated.

Blooming occurred 10 to 12 days earlier at Williamstown than at Burlington According to observations at Burlington female flowers of protandrous trees bloomed 5 to 6 days later than female flowers of protogynous trees.

Artificial self-pollinations were made when the female flowers were in full bloom by removing the isolation bag, applying pollen to each stigma with a small brush, then replacing the bag. To prevent contamination were swabbed and the brush was washed in alcohol between each pollen change. The pollen was not laboratory-tested, but seed sets in cross-pollinations (table 1) with the same pollen attested to its viability.

No protandrous trees were pollinated at Williamstown because the lower branches of these trees, on which the bags had been placed, produced only male flowers; female flowers appeared only higher in the crowns. Since protandrous trees in the Burlington area during 4 years observation have produced flowers of both sexes at all crown levels the absence of female flowers at the lower levels on the Williamstown trees was not anticipated at the time of bagging.

One experimental tree in the Burlington area was killed by heat from a nearby barn fire. Several branches of a second tree in the same area could not be used because a group of curious picnickers removed the labels.

The bags were removed 10 to 12 days after pollination, at which time the stigmas were completely shrivelled. During the first week of August, the fruits were enclosed in nylon mesh bags to minimize loss from wind and squirrels. Fruits were collected during the last week in September and the first week in October, allowed to dry for 3 to 5 days at room temperature, and then placed under refrigeration.

After about a week under refrigeration the fruits were checked for seed development. This was a relatively simple test, done by applying pressure with the thumb and index finger to the point where the samaras were attached, thus forcing the sutures to open wide enough to show whether or not a seed was present Filled fruits then were stratified in moist sphagnum moss in polyethylene bags at 35° F.

After about 2 months stratification the seed began to germinate. Twice each week all germinated seed was removed from the refrigerator and planted in plant bands in the greenhouse.

The number of branches bagged was in excess of requirements for the planned experiments. These bags were left intact during the pollination period. Here auton -omous selfing occurred through the overlap in blooming time of male and female flowers, The fruits set to this autonomous selfing were collected at maturity stratified, and planted in the greenhouse in the same manner as those from the hand pollinations.

RESULTS AND DISCUSSION

Self fertility

The sugar maple trees used in this study showed various degrees of self-fertility. Seed set in terms of flowers pollinated ranged from 8.1 percent to 27.1 percent. This was considerably less than the seed set to cross-pollination (table 1).

Variation between trees in seed set to self-pollination was found to be statistically significant by a chi-square test. Cram (4) reported a similar degree of variation in his selfing studies with Caragana arborescens. Our seed-set was substantially higher than that obtained by Wright (9) in his self-pollination work with sugar maple.

Table 1.--Seed set in 1960 to controlled self- and cross-pollinations

Tree No.	Flowers pollinated	Self-pollinated seed set	Cross-pollinated seed set
	Number	Percent	Percent
M-1	108	11.1	----
M-4	148	8.1	27.5(364-5)[1/]
M-6	68	20.6	43.3(402-5)
M-7	78	10 3	23.2(82-2)
M-13	96	19.8	40.5(496-5)
H-509	100	13.0	23.8(286-3)
H-573	118	27.1	36.4(350-4)

1/Figures in parentheses indicate number of flowers pollinated and number of male parents used. Example: M-4, 364 flowers pollinated by pollen from 5 different trees.

Characteristics of selfed progenies

The seedling characteristics noted below apply to selfed progenies from both hand pollinations and autonomous pollinations. One of the most striking characteristics of the selfed progenies was extremely wide variability in their appearance: they ranged from small, stunted individuals with miniature leaves to exceptionally vigorous plants At the weak extreme, many seeds produced only a radicle during germination, and then developed no further.

Albinism appeared in the progenies of 2 of the 7 trees that were selfed. The two progenies, one of 35 seedlings and one of 29 seedlings, each included 4 albinos. These abnormal plants all died within 8 days after germination.

The number of selfed seedlings in each progeny was relatively small, and no definite conclusions as to the mode of chlorophyll inheritance are possible at this point. However, the low ratios of white to green seedlings in the 2 progenies suggest that albinism is due to a single gene difference such as that described in maize by Lindstrom (6), and is inherited as a simple recessive.

A few seedlings with yellow cotyledons and pink cotyledons also appeared in the selfed progenies. These behaved in much the same manner as the recessive "yellow cotyledon" described in cucumber by Aalders (1). These were non-lethal deviations, and the cotyledons gradually became green within a week or 10 days.

Another unusual character was observed in both cross-pollinated and selfed progenies--namely, initially procumbent stems, which later became erect. This character was tentatively termed "lazy" because of its similarity to manifestations of the "lazy" gene in maize. Though strongly suggestive of a recessive, this character will require further investigation with larger progenies in order to clearly determine the genetic basis for its appearance.

The characters described above appeared independently of one another, which probably means that their gene loci are on separate chromosomes.

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DISSCUSSION[1]

VALENTINE - Do you have information on the relationship of the trees in the plantation? Do they come from a common seed source or would it be pretty well mixed?

FOWLER - We do not know anything about the relationship of the trees within the plantation.

KRIEBEL - I was wondering, Bill, whether your flowering situation is similar to ours in Ohio. I assume that your female flowers are morphologically hermaphroditic in that they have anthers of reduced size. Have you ever found any of these that have anthers of the same size as the male flowers, or any which were physiologically perfect - female flowers which also shed pollen or had anthers of a size which might approach normal?

GABRIEL - No I've never seen that Recently Professor Gershoy and I collaborated on a paper on pollen germination in sugar maple. Al spent quite a bit of time in the greenhouse observing the male flowers on these hermaphrodites, and he said that he saw one case of pollen shed.

SANTAMOUR - Don, would you consider that there is anything in the past geological history of red pine that would favor self compatibility?

FOWLER - Over the past few years I have had the opportunity to visit many small isolated red pine stands which are located north of the general range of this species,. Most of the red pine in this region is restricted to small islands, peninsulas or east or northeast lake shores. Practically all the stands visited were established after a fire had burned through an old red pine stand. Because of the location of the stands, the fires must have been ground fires, which left a few scarred trees to regenerate the area. In some of the stands visited, it is evident that a very few trees regenerated the entire area.

Under conditions such as these, where only a few trees play a role in regenerating an entire stand, I am quite sure that trees, which are able to produce good viable seedlings following selfing, would be favored over trees dependent upon cross fertilization.

It is likely that during times of glacial advance and retreat, conditions similar to those found in the north today occurred at one time or another over most of the species range.

SANTAMOUR - Those things come about more or less naturally, then?

FOWLER - Yes. I believe so.

[1] Transcripts of the discussions were sent to each of the participants for editing with the specific request not to change the contents of their remarks.