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A HOMOZYGOUS INTERSPECIFIC F_2 HYBRID OF *GOSSYPIUM BARBADENSE* × *GOSSYPIUM HIRSUTUM* VIA THE SEMIGAMETIC HAPLOID METHOD[1]

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ABSTRACT

The semigametic phenomenon, which allows the production of haploid plants of any specific cotton breeding strain, was used to produce a haploid plant from a male gamete of an interspecific hybrid between *Gossypium barbadense* L. and *Gossypium hirsutum* L. The treatment of this haploid plant with colchicine, doubled the chromosome number, giving an immediate early generation pureline hybrid. This study demonstrates the usefulness of the semigametic phenomenon in producing a genetically stable breeding line in two generations from interspecific crosses.

Additional index words: Breeding stocks, Genetic stocks, Pure line.

TURCOTTE and Feaster (2) reported that a doubled haploid 57-4 produced a high frequency of polyhaploids (n=2×=26) from single-embryo seeds in Pima cotton, *Gossypium barbadense* L. In 1967, they reported that crosses of various strains of cotton with 57-4 gave progenies that were chimeral for maternal and parental tissue (3). This phenomenon was explained as semigamy, an abnormal type of fertilization in which genetic fusion is lacking. The male and female gametes subsequently divide independently, giving rise to haploid-male and -female chimeral tissue.

In 1968, Turcotte and Feaster (4) showed that haploids of specific marker stocks could be produced by semigamy, and that after chromosome doubling with colchicine, these doubled haploids were homozygous for all marker genes. The purpose of the present study was to produce a haploid plant from a gamete of an F_1 interspecific hybrid of *G. barbadense* × *G. hirsutum* L. and to double the chromosome number to produce a fertile homozygous interspecific F_2 hybrid. Certain morphological and agronomic traits were to be measured and compared to the parents to determine the fixation of specific traits of each parent.

MATERIALS AND METHODS

'Acala 9519,' *G. hirsutum*, tolerant to verticillium wilt, was crossed with 'Sea Brook Sea Island $12B_2$,' (SBSI $12B_2$), *G barbadense*, which is resistant to verticillium wilt, to produce an interspecific F_1 hybrid. This F_1 hybrid was then crossed as male to a virescent, semigametic strain, 'Vsg.' obtained from Dr. E. L. Turcotte, Phoenix, Ariz. The progenies (160 plants) of this cross were grown in peat pellets, and two seedlings chimeral for virescent (female) and normal green foliage (male) were transplanted into 18.9-liter containers. One haploid male tissue sector occurred, and the chromosomes were doubled with a 0.05% colchicine solution. A fruit from a selfed flower was harvested. Seed from this fruit resulted in 15 plants, which were grown in the greenhouse for seed increase. An additional seed increase was grown in Iguala, Mexico. In 1973, selfed seed from Iguala were planted with the Acala 9519 and SBSI-$12B_2$ parents in the field. Comparisons of 50 plants each of the doubled haploid and the parents were made for fiber, plant height, maturity, wilt tolerance, and certain morphological characteristics.

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Table 1. Comparison of fiber, plant height measurements, and maturity of Acala 9519, Sea Brook Sea Island $12B_2$, and the homozygous interspecific F_2 cotton plants.

	Parents		
	Acala 9519	Sea Brook Sea Island $12B_2$	Homozygous F_2
Plant height	84.9 ± 3.2 cm	79.7 ± 4.0 cm	51.0 ± 2.0 cm
Fiber			
Length 2.5% span	1.20 in.	1.33 in.	1.08 in.
Strength (T_1)	26.4 g tex.	31.5 g tex.	26.3 g tex.
Elongation (E_1)	8.0	7.3	7.9
Uniformity ratio	54.2	37.5	50.4
Maturity			
Open bolls 16 Oct.	39.3%	1.2%	15.4%

Table 2. Comparison of morphological characters of the Acala 9519 and the Sea Brook Sea Island $12B_2$ with the homozygous F_2 cotton plants.

	Parents		
	Acala 9519	Sea Brook Sea Island $12B_2$	Homozygous F_2
Pollen color	cream	yellow	cream
Petal color	cream	yellow	cream
Petal spot	none	present	none
Leaf shape	*G. hirsutum*	*G. barbadense*	*G. hirsutum*
Locules (no.)	4-5	3	4-5
Boll size	medium	small	small
Nectaries	typical *G. hirsutum*	typical *G. barbadense*	rudimentary†
Gland distribution	medium	dense	dense

† Nectaries were smaller and secreted less nectar than those of either parent.

RESULTS AND DISCUSSION

The homozygous F_2 plants grown in the field were of a more determinate type and, consequently, shorter than either parent. These plants from a doubled haploid varied less in plant height than the parents, which had been inbred for about eight generations.

The fiber of the doubled haploid was shorter than that of either parent, whereas the strength, elongation, and uniformity were comparable to those of the Acala parent (Table 1). The hybrid was intermediate to the two parents in earliness of maturity, as measured by the percentage of open bolls by 16 October. The hybrid had the characteristics of the Acala parent for pollen and petal color, no petal spot, leaf shape, and number of locules (Table 2). It had the small boll and gland density of the SBSI-$12B_2$ parent.

The Acala parent has good tolerance to verticillium wilt. Even though it develops foliar symptoms, it still maintains a good fruit load and sufficient leaves to mature the crop. The wilt resistance of the SBSI-$12B_2$ appears to be associated with lateness, unproductivity, and a vigor that allows it to outgrow the infection. The hybrid expressed symptoms early in the season, and some defoliation occurred. The plants developed new leaves and did not show any further symptoms during the rest of the season. This type of resistance appeared to be similar to that of the *G. barbadense.*

Both species secrete nectar, with *G. barbadense* being the most active. The hybrid, however, had a nectary distribution similar to that of *G. hirsutum,* except that the nectaries were rudimentary in development and secreted almost no nectar. The formation of nectaries is determined by two dominant genes. Ne_1 and Ne_2, which were found by Holder et al. (1) to be linked to Gl_2 and Gl_3, respectively. Gl_2 and Gl_3 are major gland-determining genes. Gl_2-Ne_1 are in the A genome, andGl_3-Ne_2 are in the D genome of allotetraploid cotton. As was stated earlier, the gland distribution was as dense in the hybrid as in the SBSI-12B_2 parent. Both parents and the hybrid were of the Gl_2Gl_2, Gl_3Gl_3; Ne_1Ne_1, Ne_2Ne_2 genotype. The differences in expression between glands and nectaries were probably caused by other modifying genes.

The homozygous F_2 hybrid shed blooms at a higher rate than either parent. Meiosis appeared to be normal with normal pollen production. After four generations, there was no apparent breakdown of plant type or fertility. The hybrid in itself, however, lacked many desirable agronomic traits. Even after producing large numbers of homozygous F_2 interspecific hybrids, it would be expected that plants with only desirable traits of both species would be rare. However, the haploid method could produce doubled haploids with desirable traits that could be used as breeding stocks. The F_1 hybrid selected to produce the gametes for the doubled haploid contains a maternal set of chromosomes from one species and a paternal set from the other species. There is no doubt that some crossing over between species chromosomes occurs in F_1 at meiosis. It would also be expected that large blocks of genes from both species are represented in the gametes used to synthesize the haploid tissue. Natural selection eliminates all nonviable combinations in the haploid phase. The doubled haploid generally would carry varying amounts of genetic material from both species.

It would seem likely that the homozygous, interspecific doubled haploid with less genetic diversity than regular crosses between the two species would be more manageable for breeding and selection.

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COMPARISON OF POLLINATION METHODS IN MAIZE[1]

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ABSTRACT

Paired-row plantings of maize (*Zea mays* L.) with different endosperm colors were made to compare pollinations made from tassels bagged overnight with pollinations made by shaking a tassel into a bag just prior to pollination. Outcrosses were determined by xenia effects on kernel color. The superiority of bagged over open (unbagged) tassels was clearly evident among hybrids but the two methods were not so distinctly different for inbreds. Tearing tops from shoot bags at pollination significantly reduced contamination. If plants are short or are inbred sufficiently that outcrosses can be accurately identified, pollinations can be made more efficiently without tassel bagging and an acceptable level of outcrosses can be maintained. However if plants are tall, or if freedom from outcrosses is of major importance, bagging tassels overnight and tearing tops from shoot bags at pollination appears to be the most reliable technique.

Additional index words: **Pollen control, Pollination technique, Outcrosses, Seed set, Corn, *Zea mays* L.**

THE usual method of making hand pollinations in a maize (*Zea mays* L.) nursery involves bagging tassels and leaving the bags on overnight. This method is time-consuming; tassels sometimes break during or following bagging; and bags often collect water during rainstorms. Under favorable conditions, these difficulties can be reduced or eliminated by simply shaking a shedding tassel into a bag to pollinate. Pollinations have been made successfully throughout the day by this method for several years in Virginia. In a preliminary study in Virginia (unpublished data), a saving of 30 sec/pollination was realized using the open-tassel rather than the bagged-tassel method.

Occasional outcrosses in hand-pollinated seed are virtually impossible to prevent. If they can be easily detected they cause little concern. My interest in the development of vigorous, productive inbred populations revived my concern for pollen control. Tall plants are difficult to pollinate without jarring surrounding plants and causing them to shed and outcrosses in vigorous progenies are often difficult to detect. This experiment was devised to compare the frequencies of outcrosses that might be expected when maize pollinations are made with bagged and open (unbagged) tassels.

MATERIALS AND METHODS

Two vigorous inbred lines of similar maturity but different endosperm color, K9408 (white) and Va35 (yellow), were planted in 1971 in alternate rows for three replications of five pollen-control treatments. Rows were 3.7 m long and 0.9 m apart with 15 plants/row. An attempt was made to obtain data on 10 or more ears/row. Two hybrids of similar maturity, 'Pioneer 509'

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