Genetics of Flowering Response in Cotton. VI. Flowering Behavior of *Gossypium hirsutum* L. and *G. barbadense* L. Hybrids[1]

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ABSTRACT

The center-of-origin cottons (*Gossypium* spp.) are isolated from cultivated American cottons by their short-day flowering response, and an understandng of the inheritance of flowering response is important in attempting to transfer genes from these primitive cottons. The genetic systems controlling flowering response in *Gossypium hirsutum* and *G. barbadense* cottons were studied in diallel crosses involving short-day anl day-neutral lines of each species. The *G. hirsutum* lines were Texas Marker-1 and M-11 (day-neutral) and Texas 220 and Texas 371 (short-day). The *G. barbadense* lines were 'Pima S-1' and 3-79 (day-neutral) and 'Lengupa' (short-day). These lines were crossed in all possible combinations, and the F_1, F_2, backcrosses to both parents, and a testcross to Texas Marker-1 were grown for each combination. Flowering response was recorded as percentage of flowering plants and the mean date of flowering for each line in the field at College Station, Texas. A failure to meet the preanalysis tests of assumptions prevented the analysis of these data by the Hayman-Jinks diallel analysis. The data were analyzed by comparison of mean performance. We concluded that *G. hirsutum* and *G. barbadense*, represented in this study, have nonhomologous systems controlling flowering response. Within *G. barbadense*, flowering response is recessive to nonflowering and under single-gene control. In *G. hirsutum*, however, flowering response is under multigenic control, which is partially dominant or additive to nonflowering. In interspecific cross combinations, the segregation for flowering response follows a multigenic pattern.

Additional index words: Genetic nonhomology, Interspecific hybrids, Germplasm, Transference of genes.

BOTANISTS who have studied the history and evolution of cotton (*Gossypium* spp.) tell us that the center of origin for *Gossypium hirsutum* L. is in Mexico and Central America. Certain of the feral *G. hirsutum* stocks collected from that area shortly after the turn of the century contributed to the establishment of commercial varieties in the United States. Extensive collections made by U.S. workers some 20 to 25 years ago represent a major genetic resource that has been exploited very little in the improvement of cultivated American Upland cotton. In recent years, however, there has been a revival of interest in the potential value of center-of-origin cottons for agronomic improvement and pest management.

By agreement reached through a regional research project (S-77), cotton researchers at College Station, Texas have responsibility for maintenance of the *G. hirsutum* primitive germplasm. The present collection comprises over 600 entries that have been catalogued and placed in storage at the National Seed

Storage Laboratory, Fort Collins, Colorado. These center-of-origin stocks, native to Mexico and Central America, were classified into seven geographic races: *latifolium, marie-galante, morrilli, palmeri, puncta- tum, richmondi,* and *yucantanense* (Hutchinson, 1951). The cultivated American Uplands belong to the *lati- folium* race.

One problem is that most of these tropical stocks do not flower under conditions of the relatively long summer day in the U.S. Cotton Belt. In their native habitat these cottons flower and set fruit during the winter months, and they are vegetative during the summer. This coincides with the winter dry season and the summer wet season.

A first step torward using this germplasm in the U.S. Cotton Belt has been an attempt to understand the factors that control the flowering response. A. Lang (unpublished) established that the primitive cottons were sensitive to daylength, and that they could be induced to flower by controlled short day- lengths or by grafting to flowering plants. The cul- tivated American Upland plants are insensitive to daylength and are said to be day-neutral.

In working with these primitive cottons, it becomes apparent that photoperiod is not the only factor con- trolling flowering response; temperature, maturity, and water balance also play a part. Hutchinson (1959) concluded that photoperiod, maturity, and water bal- ance were important in the induction of flowering in the races. He recognized photoperiod as the primary control.

Since the primitive *G. hirsutum* cottons, in their natural habitat, are perennials and have an indeter- minate growth habit, he proposed that the plants had to produce a critical number of nodes before flower- ing was initiated. In cultivated plants his response relates to differences in earliness when they are man- aged as annuals. We have observed that certain plants growing in a tropical environment do not flower dur- ing the first growing season, but flower profusely dur- ing their second season.

Hutchinson (1959) concluded that water balance was important since some strains would initiate floral buds during the rainy season, but the buds would be shed until the beginning of the dry season.

Waddle, Lewis, and Richmond (1961) noted that flowering response varied at different locations in the Cotton Belt, and they attributed this response to temperature differentials. Mauney and Phillips (1963) showed experimentally that reduced night tempera- ture hastens the time of floral initiation.

To investigate the problems and potential success of introducing other race germplasm into Upland cotton, inheritance studies of the flowering response in field-grown conditions were conducted at College Station, Texas. These studies involved the hybrid progeny between wild accessions of the races and cultivated Upland cottons. Three *latifolium* lines were included in the studies by Waddle, Lewis, and Richmond (1961) and Kohel and Richmond (1962). Flowering response was inherited as a complex trait, and flowering was partially dominant.

Another race studied was *marie-galante* (Lewis and Richmond, 1957). It was selected to represent more pronounced perennial growth habits. In the hybrid

progeny, flowering was influenced by photoperiod response and maturity factors. The inheritance of flowering was complex and primarily conditioned by additive gene action, but the backcrosses showed dominance toward the recurrent parent.

Lewis and Richmond (1960) investigated the flowering response in *G. barbadense* L. In the progeny of a cross between short-day 'Lenggupa' and day- neutral 'Pima S-1,' they found that a single gene controlled flowering, and that flowering was recessive to nonflowering.

The interrelations between *G. hirsutum* and *G. barbadense* were investigated by Kohel, Lewis, and Richmond (1965). In crosses between short-day *marie- galante* (*G. hirsutum*) and day-neutral Pima S-1 (*G. barbadense*), and between short-day Lenggupa (*G. barbadense*) and day-neutral Upland (*G. hirsutum*), flowering response, in both sets of hybrid progeny, was inherited as a complex trait as seen in intraspecific *G. hirsutum* crosses.

The present study was conducted to define more clearly the relations between the genetic systems controlling flowering response in *G. hirsutum* and *G. barbadense*. A complete diallel cross used short- day and day-neutral lines of both species. The diallel cross was supplemented with F_2, backcross, and test- cross progeny.

MATERIALS AND METHODS

The following parental lines were used in these experiments:

Gossypium hirsutum

    Texas Marker-1 (TM-1)—a day-neutral and long-term inbred line that was used in earlier studies (Kohel, Richmond, and Lewis, 1970) with *latifolium* and *marie-galante*.

    M-11—A day-neutral line derived from a doubled haploid of 'Empire.'

    Texas 220—A short-day *latifolium*.

    Texas 371—A short-day *marie-galante* used in an earlier study (Lewis and Richmond, 1957).

Gossypium barbadense

    Pima S-1—A day-neutral cultivar used previously in the study with Lenggupa.

    3-79—A day-neutral line derived from a doubled haploid of a line involved in the pedigree of Pima S-1. It was included because it was considered to have less possible *G. hirsutum* introgression than Pima S-1.

    Lenggupa—A short-day cultivar from Colombia that was used in two previous studies involving *G. barbadense* (Lewis and Richmond, 1960; Kohel, Lewis, and Richmond, 1965).

The lines were crossed in all possible combinations in the greenhouse during the winter months. For each hybrid combina- tion, we attempted to obtain the F_1, F_2, BC^1, BC^2, and a test- cross to TM-1. We were not able to obtain enough seed to test the backcross generations between *latifolium* and *marie-galante*. The entire test was grown in a single year, with a few excep- tions. Some entries that were not available for the first test were obtained after additional greenhouse crossings; these were grown in a supplemental test.

Seeds were germinated the first week of April in peat pellets in the greenhouse and transplanted to the field at 3 weeks of age. Nonsegregating entries were grown in single 20-plant rows (45 cm between plants and 102 cm between rows), and segre- gating material was represented by 2 rows. The test was repli- cated three times. Larger populations were grown for some entries in the supplemental test.

The date on which each plant flowreed was recorded along with the day of the year. The first flower appeared about June 9 (160) and the plants were scored from that date until they had ceased to initiate flowering, August 18 (230). This period was longer than the effective flowering period previously recorded. Usually the flowering period is terminated by lack of moisture

and high temperatures. The test year and the year of the supplemental test were similar, and both had more rainfall and cooler temperatures than usual. These factors no doubt prolonged flowering.

Data on flowering were expressed in three ways: 1) mean date of first flower of the flowering plants, 2) percentage of flowering plants, and 3) a flowering score. The flowering score was the mean date of first flower and the nonflowering plants were entered into the computations by assigning them the value of 260. This value was chosen to approximate the mean date of first flower for all those plants that had not flowered by the termination of the field test.

RESULTS AND DISCUSSION

The original plan called for an analysis of parental, F_1, and F_2 data for percentage flowering and flower score by the Hayman-Jinks diallel analysis. However, the data failed to meet the assumptions. Tests of the slope of the regression line of W_r on V_r, analysis of variance of $(W_r - V_r)$, test of epistasis, and analysis of variance of the deviation of the components were all statistically significant for the raw data and their transformations. The tests indicate that at least the assumptions of no epistasis, no multiple alleles, and no correlated gene distributions were not met. The analysis was confined to a comparison of mean responses.

G. hirsutum and G. barbadense are allotetraploids with presumed common ancestry. They are now discrete species, and each has undergone a high degree of diploidization in gene function, so that they are functionally diploids. The preliminary analysis results and the fact that these species have evolved under distinct species specific diploidization suggest that we may be dealing with nonhomologous genic systems controlling flowering response.

Flowering data from TM-1 and M-11 were pooled because there was so little difference in flowering response between them. The day-neutral barbadense stocks, Pima S-1 and 3-79, behaved in a similar manner, and flowering data from them also were pooled. This pooling reduced number of combinations from 21 to 10. Both the Upland and Pima parental stocks flowered at about the same time, days 176 and 180, respectively. The short-day G. hirsutum race marie-galante and the short-day G. barbadense, Lengupa, were chosen to represent lines that do not flower in this temperate-zone environment. Neither of these lines flowered during the course of the test. Though classified in general photoperiodic response as a short-day cotton, the latifolium line used in these experiments was chosen, on the basis of earlier data, to represent an intermediate flowering response—one in which less than half the plants in a progeny would flower. Furthermore, flowering on those plants that flowered was sparse and considerably later than that of normal day-neutral stocks. However, in this test 69% of the plants in this latifolium line flowered and, on the average, they flowered 205 days after planting. The flowering data just reported are well within the range of flowering response shown by members of race latifolium, but in view of its previous record we did not expect the latifolium stock that we used to give as high a percentage of flowering plants as it did, nor did we expect those plants that flowered to do so as early as they did. Any number of reasons for this behavior may de advanced but all

are speculative. Possibly the data recorded earlier did not represent the usual flowering behavior of the stock. On the other hand, the environment during the present test could have been exceptional or sufficiently different from that in which the earlier data were taken to have produced the observed effects.

A comparison of the intraspecific day-neutral × short-day cross combinations (Upland × marie-galante and Pima × Lengupa) illustrates the differences in flower response between the two species (Table 1). In the G. hirsutum cross combinations, multifactored inheritance was evident, with partial dominance or additive gene action controlling flowering. In the G. barbadense cross combinations, single-factor segregation was apparent, nonflowering was dominant, and the mean flowering dates of the flowering plants were similar and only slightly later than those of the flowering Pima stock. That there was little variability among the flowering plants in respect to time of flowering is worthy of special note.

The performance of the intraspecific crosses is contrasted with that of the interspecific day-neutral × short-day cross combinations (Upland × Lengupa and Pima × marie-galante). In the combinations involving Upland × Lengupa, there was no apparent expression of the dominant nonflowering gene; the values for the various generations paralleled those of the Upland × marie-galante generations, but at a lower level of flowering (plants flowered at a later date and fewer plants flowered). The lower level of flowering suggested that flowering response is under additive or partial-dominant multifactorial control. This response is in contrast to the single type of control shown by the Pima × Lengupa cross.

The cross combinations involving Pima × marie-galante gave a lower level of flowering response than the Upland × Lengupa combinations, but did not reach the low level of the Pima × Lengupa combinations. The distribution of percentage flowering plants between generations of the Pima × marie-galante combinations were reminiscent of the Pima × Lengupa combinations, but mean flowering date did not follow the same pattern. In the Pima × marie-galante combinations, flowering response was recessive.

The marie-galante × Lengupa combinations revealed that marie-galante and Lengupa do not have homologous nonflowering genes, because 1% of the F_2 (4 plants) and 1% of the BC^1 (1 plant) flowered. This low level of flowering suggests that the two nonhomologous systems complement each other.

Inspection of the array means (Table 1) reveals that there is a progressive decrease in dominance of flowering from the Upland array to the latifolium, marie-galante, Lengupa arrays, and distinct recessiveness in the Pima array when the F_1, F_2, and BC generation means are compared.

From these results, we can be reasonably sure that in the G. barbadense crosses, flowering response was recessive to nonflowering, and there were only a few genes that control time of flowering. In the G. hirsutum crosses, on the other hand, flowering response was controlled by many genes, and these genes had a cumulative effect that determined the time of flowering. In the interspecific combinations

Table 1. Mean flowering date, percentage flowering plants, flowering score, and number of plants in the parental F_1, F_2, BC^1, BC^2, and testcross populations of selected *Gossypium hirsutum* and *G. barbadense* short-day and day-neutral lines.*

Parent and population	Upland $\overline{X}_F$	$\%_F$	$\overline{X}_C$	n	latifolium $\overline{X}_F$	$\%_F$	$\overline{X}_C$	n	marie-galante $\overline{X}_F$	$\%_F$	$\overline{X}_C$	n	Lengupa $\overline{X}_F$	$\%_F$	$\overline{X}_C$	n	Pima $\overline{X}_F$	$\%_F$	$\overline{X}_C$	n	Array mean $\%_F$	$\overline{X}_C$
Upland	176	100	176	207																		
F_1					198	98	199	109	209	68	225	109	213	39	242	115	179	100	179	225	76	211
F_2					198	96	201	213	205	54	230	110	195	20	247	200	179	100	179	396	68	214
BC^1					179	100	179	300	183	100	183	403	187	85	198	307	176	100	176	651	96	184
BC^2					206	70	222	256	226	1	259	136	219	1	260	102	180	100	180	414	43	230
TC					179	100	179	300	183	100	183	403	187	85	198	307	176	100	176	651	96	184
Mean					192	93	196		201	65	216		200	46	229		178	100	178		76	205
Latifolium					205	69	218	118														
F_1									225	12	256	110	224	4	255	117	208	15	252	136	32	240
F_2									223	11	256	172	212	8	256	458	198	37	237	106	38	238
BC^1									–	–	0		209	29	245	124	213	27	247	199	42	238
BC^2									–	–	0		200	2	259	177	185	63	213	206	41	228
TC									207	92	211	112	202	74	217	141	190	84	201	184	88	202
Mean									218	31	244		209	23	246		199	45	230		48	229
marie-galante									–	0	260	115										
F_1													–	0	260	119	200	3	258	177	21	250
F_2													200	1	260	466	196	31	240	478	24	246
BC^1													199	1	259	118	225	2	250	117	1	259
BC^2													–	0	260	109	186	59	213	181	50	223
TC													207	55	231	114	189	89	197	92	84	206
Mean													202	11	254		199	37	233		36	237
Lengupa													–	0	260	102						
F_1																	–	0	260	153	11	254
F_2																	184	33	235	301	16	250
BC^1																	–	0	260	66	1	260
BC^2																	186	58	217	126	43	230
TC																	188	84	200	156	74	212
Mean																	186	35	234		29	241
Pima																	180	100	180	208		
F_1																					30	237
F_2																					50	223
BC^1																					70	206
BC^2																					32	236
TC																					89	194
Mean																					54	219

* BC^1 and BC^2 refer to the first backcross to the parents listed on the left side and top of the table, respectively. TC refers to the testcross of the F_1 to Texas Marker-1. $\overline{X}_F$ = mean flowering date, $\%_F$ = percentage flowering plants, $\overline{X}_C$ = flowering score, and n = number of plants.

with Lenmpa, the Lengupa nonflowering gene had no apparent effect; we assumed there was no similar flowering control system present on which it could act. However, in the segregating generations, F_2 and BC, there was some expression because of reassociation of the *G. barbadense* genes. In the interspecific combinations involving Pima and short-day *G. hirsutum*, the *G. hirsutum* genes would essentially be in a hemizygous condition and have a reduced effect on flowering. This effect is illustrated by the greater flowering response of the *latifolium* parental line than of the *latifolium* × Pima F_1.

These interpretations of genetic control of flowering response to the transfer of genes from the race stocks imply that transfers within *G. hirsutum* can be accomplished in a routine manner. In actual practice, such transference programs have been employed with success. The only difficulty is that the stronger the nonflowering response in the race line, the fewer flowering recombinants recovered, and these would be later maturing.

Since the collection of *hirsutum* races has been evaluated to a greater extent than has the *barbadense* collection, it is more likely that short-day *hirsutum* germplasm would be transferred into *barbadense* than the reverse. This is true for the immediate future, at least. The situation in which short-day *G. hirsutum* germplasm is being transferred into day-neutral *G. barbadense* presents a more difficult problem than the transfer of short-day *G. hirsutum* to day-neutral

G. hirsutum because there would be fewer flowering recombinants and they may be late maturing.

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