

# Breakup of Initial Linkage Blocks Through Intermating in a Cotton Breeding Population<sup>1</sup>

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## ABSTRACT

Restricted character recombination was observed in the  $F_2$  generation of a cross between two contrasting inbred lines of cotton. If initial linkage blocks were an important cause of the restricted recombination, intermating for several generations should break up such linkages and increase genetic recombination. Beginning with the  $F_2$  generation, the population was maintained for six successive generations in an isolation block where the natural mating system was mixed intermating and selfing (approximately 50% self-pollination). The population was resampled to study the effects of intermating on genetic recombination for seven quantitative traits. Genotypic variances decreased for six traits for which coupling-phase linkages would be expected to predominate and increased for one trait for which repulsion linkages might have been more important. Genotypic correlations between traits tended to shift toward values observed in populations assumed to be more nearly at linkage equilibrium. These observed changes conform with expectations that intermating dissipates initial linkage disequilibrium. Changes in gene frequency which occurred during intermating for some of the traits, however, confounded the interpretation of the observed results.

**Additional Index Words:** genetic recombination, genetic variance, genetic correlation, linkage equilibrium.

THE segregation observed in the  $F_3$  generation of a cross between an unadapted cotton strain of interspecific origin and an adapted Upland type was observed by Al-Jibouri et al. (1). The breeding objective was to combine the desirable fiber strength of the first parent with the high yield and good adaptation of

the second. Replicated tests of the  $F_3$  progenies, however, revealed that character recombinations in the direction of nonparental types were very restricted. Combinations similar to the original parental types were recovered quite frequently. As discussed by Anderson (2) and Dempster (6), these character associations may be due to linkage, pleiotropism, or a combination of both.

The immediate effects of pleiotropism and close linkage would be generally similar in the first segregating generation following a cross. Thus, in the study cited, it was not possible to distinguish between these possible causes of restricted recombination. Their consequences in regard to the potential breeding value of the material, however, would be different and of critical importance.

The investigation reported here is a continuation of the above study. It was hypothesized that if initial linkage blocks were an important cause of the observed restricted recombination, intermating for several generations might tend to break up such linkages and increase genetic recombination in the population. Manual intercrossing of a large number of cotton plants is tedious. Since natural outcrossing of cotton in the Raleigh, N. C., area is of the order of 50%, considerable intercrossing between plants within the population would occur if the material was simply maintained for several generations in an isolated block. Such a program was initiated with the  $F_2$  generation of this cross. After six generations, the population was resampled to study the effects of this mixed random mating and selfing on genetic recombination.

## MATERIALS AND METHODS

The populations studied were generated from the cross Empire 10  $\times$  TH 131-5. Empire 10 is a component strain of the 'Empire' variety (*Gossypium hirsutum* L.) and is adapted to the southeastern cotton-producing areas. TH 131-5 is one of the strains descending from the tri-species hybrid (*Gossypium thurberi* Tod.  $\times$  *Gossypium arboreum* L.  $\times$  *G. hirsutum* L.) originally reported by Beasley (3). Six generations of recurrent

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crossing of the tri-species hybrid to locally adapted Upland varieties, coupled with strong selection pressure for high lint strength, resulted in fully fertile strains. Except for their desirable lint strength and acceptable fiber length, however, these strains are low in yield and generally undesirable from an agronomic standpoint. Both the parental strains used in this study were inbred by a pedigree (individual plant) system for at least six generations prior to making the cross and are assumed to be homozygous.

The  $F_2$  and the five succeeding generations were grown in successive years in a field near Raleigh, N. C., which was isolated by a distance of at least a half mile from other cotton. Seed was drilled thinly within the row, with no artificial thinning following emergence. Due to failures in seed germination and in seedling emergence and survival, it is estimated that the plants established in the field represented about 1/3 to 1/2 of the seed planted. Such stands are quite normal in the area where the experiment was conducted. The  $F_2$  generation consisted of approximately 500 plants. Twenty-five seeds were harvested from each  $F_2$  plant and bulked to plant the following generation. Populations of 4,000 to 6,000 plants were established for each of the remaining generations. After the first year, six to seven seeds were harvested from each plant, bulked, and approximately 1/2 of the seed was used for the next year's planting. At the end of six generations, a sample of the seed harvested was planted in the breeding nursery and self-pollinated seed was obtained from 96 random plants. The same year 96 random plants from an  $F_2$  population of the original cross were likewise self-pollinated. Thus, seed was available for replicated progeny tests comparing the population which had undergone six generations of mixed selfing and random mating (designated as the intermated population) with the original  $F_2$  population.

In order to decrease the effects of soil heterogeneity, the above progenies were randomly divided into six sets. Each set consisted of 16  $F_3$  progenies and 16 progenies from the intermated population. In addition, two entries each of the two parental lines, Empire 10 and TH 131-5, were included in each set. This material was grown at the Upper Coastal Plains Experiment Station near Rocky Mount, N. C. in two consecutive years. A randomized complete block design was used for each set with three replications the first year and two replications the second. Plots were single 457-cm (15-foot) rows, 91.4 cm (36 inches) apart, thinned to a stand of one plant per 10 cm (3 plants per foot).

Data were recorded on seed cotton yield, lint-yield, lint percent, and weight per boll. A random sample of 25 bolls was harvested from each plot to measure lint percentage and weight per boll. Lint yield values were calculated by multiplying the total weight of seed cotton per plot by the lint percentage value for that plot. Lint samples were taken from all plots and analyzed by the U. S. Department of Agriculture Fiber Laboratory at Knoxville, Tenn., to obtain data on fiber properties. Fiber property measurements were as follows:

Fiber length, U.H.M. (Upper Half Mean) — the length in inches of the half of the fibers by weight that contains the longer fibers.

Fiber strength,  $T_1$  units — the fiber strength of a bundle of fibers measured on the Stelometer with two jaws holding the fiber bundle separated by an  $1/8$ -inch (.32-cm) space. Strength is expressed as grams/grex.

Fiber elongation,  $E_1$  units — the percentage elongation at break of the center  $1/8$  inch (.32 cm) of the fiber bundle measured for  $T_1$  strength on the Stelometer.

Fiber fineness, Mic. (Micronaire) — the fineness of the sample measured on the Micronaire and expressed in standard (curvilinear scale) Micronaire units.

Effects of the several generations of intercrossing on genetic recombination were studied by comparing estimates of genetic variances and covariances from the two populations assuming no linkages. Discrepancies between the two estimates would then reflect the effects of intercrossing on breakup of initial linkage blocks, provided the intermated population had been developed without selection. Throughout this report, epistasis is assumed to be absent, although in some cases the genetic expectations include the additive  $\times$  additive epistatic component.

Estimates of the components of variance and covariance among selfed progenies were obtained from separate analyses of the two populations. The pertinent portion of the analysis of variance or covariance and the mean square or product expectations are presented in Table 1. Due to the unequal number of replicates in the 2 years, the coefficients ( $k$  values) in the expectations were calculated as outlined by Cochran and Cox (4). Of

Table 1. Pertinent portion of analysis of covariances between traits  $i$  and  $j$  of data from 2 years (analysis of variance if  $i = j$ ).

Source	df	Mean product	Expectation of mean product*
Progenies in sets	$t_1 = (p-1)s$	$M_{1ij}$	$\sigma_{ij} + k_2 \sigma_{pyij} + k_3 \sigma_{pij}$
Progenies in sets $\times$ years	$F_2 = (p-1)s(y-1)$	$M_{2ij}$	$\sigma_{ij} + k_1 \sigma_{pyij}$
Pooled error	$t_2 = (r_1-1+r_2-1)(p-1)s$	$M_{3ij}$	$\sigma_{ij}$

\*  $\sigma_{ij}$  = pooled plot error covariance (variance),  $\sigma_{pyij}$  = interaction covariance (variance) of progenies  $\times$  years,  $\sigma_{pij}$  = genotypic covariance (variance) among progenies,  $k$  values are expected coefficients due to unequal number of replications where  $k_1 = 2/4$ ,  $k_2 = 2/6$ , and  $k_3 = 5$  (see text),  $p$  = number of progenies,  $s$  = number of sets,  $y$  = number of years,  $r_1$  and  $r_2$  = number of replications.

primary interest for this study are the progeny or genotypic components of variance ( $\sigma_{pij}$ ) or covariance ( $\sigma_{pij}$ ) which were estimated as:

$$\hat{\sigma}_{pij} = \frac{1}{k_3} \left[ M_{1ij} - \frac{k_2}{k_1} M_{2ij} + \left( \frac{k_2}{k_1} - 1 \right) M_{3ij} \right]$$

These components were used to estimate the genotypic correlations,

$$\text{Genotypic } r_{ij} = \frac{\sigma_{pij}}{\sqrt{\hat{\sigma}_{pi} \hat{\sigma}_{pj}}}$$

which is the additive genetic correlation only if all genetic variance is additive.

Variances of estimated genotypic components of variance and covariance were estimated as:

$$V(\hat{\sigma}_{pij}) = \frac{1}{k_3^2} \left[ \frac{M_{1ij} M_{1ij} + M_{2ij}^2}{f_1 + 2} + \left( \frac{k_2}{k_1} \right)^2 \left( \frac{M_{2ij} M_{2ij} + M_{3ij}^2}{f_2 + 2} \right) + \left( \frac{k_2}{k_1} - 1 \right)^2 \left( \frac{M_{3ij} M_{3ij} + M_{3ij}^2}{f_3 + 2} \right) \right]$$

Variances of genotypic correlation coefficients were estimated as outlined by Mode and Robinson (14).

If the  $F_2$  population had been randomly mated for six generations rather than allowing natural mating (50% selfing), the parameters estimated from the two populations would have been directly comparable, so that any differences would have been ascribable, assuming no selection, to the effects of recombination destroying linkage blocks. It can be shown that a system having a constant probability of selfing,  $\alpha$ , and  $(1-\alpha)$  probability of random mating, has an inbreeding coefficient after  $n$  generations of  $F = [\alpha/(2-\alpha)] [1 - (\alpha/2)^n]$  which rapidly approaches an equilibrium with inbreeding of  $F = [\alpha/(2-\alpha)]$ . (8). Thus, comparison of the estimates of the genetic parameters requires that they be translated from one generation of inbreeding to another. This translation can be made if gene frequency is 1/2, presumably satisfied by the origin of the population, and if the genetic model is limited to exclude any types of epistasis having dominance in the nomenclature (5). In relating the genetic parameters in the two populations, it is assumed that there is no linkage, that the intermated population was at equilibrium with respect to inbreeding ( $F = 1/3$ , when  $\alpha = 1/2$ ), and that the gene frequencies did not change in producing the intermated population. Under these conditions, the genetic expectation of the genotypic component of variance for the original population is

$$\sigma_{pij} = \sigma_A^2 + 1/4 \sigma_D^2 + \sigma_{AA}^2$$

and for the intermated population,

$$\sigma_{pij} = 4/3 \sigma_A^2 + 2/9 \sigma_D^2 + 16/9 \sigma_{AA}^2$$

where the model is restricted to additive, dominance, and additive  $\times$  additive two-factor epistasis and using the  $F_2$  population as the base of reference.

Similarly, the means of the selfed progenies in the two populations would be expected to differ due to inbreeding alone if dominance is of any importance. Assuming a nonepistatic model, the genetic mean of the progenies from the intermated population ( $\bar{x}_i$ ) can be related to the genetic mean of the progenies from the original population ( $\bar{x}_o$ ) as

$$\bar{x}_i = \frac{2}{3} \bar{x}_o + \frac{1}{3} \left( \frac{\bar{P}_1 + \bar{P}_2}{2} \right),$$

where  $\bar{P}_1$  and  $\bar{P}_2$  are the means of the two original parental

Table 2. Average performance of parental lines.

Trait	Empire 10	TH 131-5	High parent as percent of low parent
Lint yield, kg/ha	827 ± 43	558 ± 43	148
Lint, %	36.9 ± 0.3	33.7 ± 0.3	109
Weight per boll, g	7.22 ± .16	5.76 ± .16	125
Fiber length, U.H.M.	1.105 ± .008	1.138 ± .008	103
Fiber strength, T <sub>1</sub>	1.77 ± .04	2.60 ± .04	147
Fiber elongation, E <sub>1</sub>	6.8 ± 0.2	5.8 ± 0.2	117
Fiber fineness, Mic.	3.71 ± .09	4.18 ± .09	113

lines. This relationship would be unaffected by linkages. Thus, tests for deviations from this model are tests of the composite hypothesis of no selection in the formation of the intermated population, no epistasis, and constant percent selfing. Unfortunately, an independent test for selection alone is not available from these data. Deviations from the above model were tested using a weighted analysis of generation means, i.e.,  $\bar{P}_1$ ,  $\bar{P}_2$ ,  $\bar{X}_0$ , and  $\bar{X}_1$ , where the weighting was by the inverse of their respective variances.

## EXPERIMENTAL RESULTS

### Means

Performance data for the two parental lines are presented in Table 2. The two parents contrast substantially for all traits except fiber length.

Data comparing the means of the 96 F<sub>3</sub> progenies of the original population with those of the 96 progenies of the intermated population are presented in Table 3. These comparisons are of interest to determine whether selection occurred during the successive intermating generations. Direct comparisons of the observed population means, however, are confounded with the differing coefficients of inbreeding. Accordingly, "expected" means were calculated for the intermated population assuming no selection and adjusting for the change in the coefficient of inbreeding (see Methods Section). It should be noted that, if average gene effects are additive, no changes in means would be expected during the successive intermating generations. Adjusted means for the intermated population are presented in the fourth column of Table 3 and varied only slightly from the observed means of the original population.

The tests for the composite hypothesis of no selection, no epistasis, and 50% selfing were significant or highly significant for all characters except weight per boll and fiber fineness. Errors in the assumption regarding degree of inbreeding would be expected to affect the observed changes in means similarly for all traits. For any degree of inbreeding without selection, the mean value of a trait in the original population ( $F = 0$ ) would be expected to shift toward the mid-parental value. In the cases of yield and length, however, the shifts were actually in an opposite direction. For fiber strength, the change was in the direction expected for inbreeding, but the shift was much larger than could be accounted for by complete inbreeding, i.e., further than the midparental value. Thus, observed deviations from "expected" values do not appear explainable by simple error in the assumption regarding degree of inbreeding. No information is available concerning the possible effects of epistasis. It would appear, however, that the primary cause of the observed deviations must be selection, principally for Empire 10 characteristics. Except for yield and fiber strength, the percentage changes were relatively small.

Table 3. Comparison of original and intermated population means.

Trait	Original	Mid-parental value	Intermated	Expected means intermated population†	Deviation from expected %
Lint yield, kg/ha	720±14	692	792±14	711±16	+11.4**
Lint, %	34.7±0.1	35.3	35.4±0.1	34.9±0.1	+1.4**
Weight per boll, g	6.31±.06	6.49	6.32±.06	6.37±.06	~ 0.8
Fiber length, U.H.M.	1.130±.003	1.121	1.135±.003	1.127±.003	+0.7*
Fiber strength, T <sub>1</sub>	2.25±.01	2.18	2.11±.01	2.23±.02	-5.4**
Fiber elongation, E <sub>1</sub>	6.1±.1	6.3	6.4±.1	6.2±0.1	+3.2*
Fiber fineness, Mic.	3.92±.03	3.94	3.92±.03	3.93±.04	~ 0.3

\*, \*\* Significant deviation from an additive plus dominance generation analysis at 5% and 1% levels. † Predicted from the original population mean and the mid-parental value, assuming 50% self pollination, no selection and adjusting for change in coefficient of inbreeding (see text).

Table 4. Comparison of genotypic variances ( $\sigma_{pi}$ ) of original and intermated populations. Standard errors in parentheses.

Trait	Observed original	$\sigma_{pi}$		Ratios	
		Observed intermated	Expected intermated*	Obs. interm./Expected	Differ./S.E. of differ.
Lint yield, kg/ha	3332(1281)	3486(1375)	4442(1707)	0.78	< 1
Lint, %	2.136(.358)	2.370(.388)	2.848(.477)	0.83	< 1
Weight per boll, g	.0923(.0242)	.0970(.0275)	.1231(.0323)	0.79	< 1
Fiber length, U.H.M.†	.0487(.0102)	.1112(.0186)	.0622(.0136)	1.79	> 2
Fiber strength, T <sub>1</sub>	.0312(.0054)	.0199(.0035)	.0416(.0072)	0.48	> 2
Fiber elongation, E <sub>1</sub>	.1619(.0468)	.1248(.0410)	.2159(.0624)	0.58	> 1
Fiber fineness, Mic.	.0663(.0129)	.0707(.0147)	.0884(.0172)	0.80	< 1

\* Assuming all additive variance, no linkage, 50% self pollination, and no selection in the development of the intermated population (see text). † Actual values divided by 100.

### Variances

Comparisons of the genotypic components of variance for the original and intermated populations are presented in Table 4. As with the means, direct comparisons of the original components are confounded with the differences in degree of inbreeding in the two populations. Consequently, it was necessary to examine the expected effects of inbreeding on the genotypic components of variance. Pertinent procedures for an additive and dominance model have been outlined in the Methods Section. Ramey and Miller (15) estimated the relative magnitude of additive genetic variance and dominance variance in the F<sub>2</sub> generation of the same cross that is being considered here. Significant estimates of dominance genetic variance were obtained for only two traits, lint percentage and fiber fineness. For both of these traits, however, the actual magnitude of the additive genetic variance was many times larger than that for dominance variance. Even if dominance were important in the F<sub>2</sub> population, bias due to dominance in the difference between the F<sub>2</sub> and expected variances for the intermated population would be relatively small (only 1/9  $\sigma_D^2$ ). Thus, the  $\sigma_D^2$  component in the model may be ignored for practical purposes and adjustments for differences in inbreeding may be restricted to  $\sigma_A^2$ .

Expected genotypic variances for the intermated population, assuming 50% selfing and no selection, are listed in Table 4. Comparisons were then made between the observed and expected values in the form of ratios. Ratios for six of the seven traits (Table 4) are less than unity, indicating that after adjustment for inbreeding there was a decrease in genotypic variance for these traits during the intermating generations. For fiber length, on the other hand, there was a sizeable increase in variance.

### Genotypic Correlations

Estimates of the genotypic correlation coefficients between all possible pairs of the seven traits studied

Table 5. Genotypic correlations ("r" values) between pairs of traits in the original and intermated populations. Standard errors in parentheses.

Traits correlated	Genotypic "r"		Diff./S.E. of diff.	Avg "r" value*
	Original	Intermated		
Lint yield vs lint %	.90 (.12)	.67 (.12)	> 1	.80
vs weight/boll	.14 (.23)	-.20 (.24)	> 1	-.42
vs fiber strength	-.69 (.14)	-.35 (.16)	> 1	-.20
vs fiber elongation	.71 (.19)	.37 (.25)	> 1	--
vs fiber fineness	.42 (.17)	.46 (.19)	< 1	--
Lint % vs weight/boll	.19 (.15)	-.04 (.15)	> 1	-.36
vs fiber strength	-.70 (.07)	-.62 (.08)	< 1	-.12
vs fiber elongation	.74 (.11)	.23 (.15)	> 2	--
vs fiber fineness	.26 (.12)	-.08 (.13)	> 1	--
Wt/boll vs fiber strength	-.53 (.12)	-.36 (.14)	< 1	.95
vs fiber elongation	.24 (.20)	.13 (.23)	< 1	--
vs fiber fineness	-.56 (.17)	-.49 (.20)	< 1	--
Fiber strength vs				
fiber elongation	-.51 (.13)	-.22 (.16)	> 1	--
fiber fineness	.14 (.13)	.33 (.13)	> 1	--
Fiber elongation vs				
fiber fineness	.45 (.16)	.20 (.19)	> 1	--
Fiber length vs				
lint yield	.02 (.20)	-.37 (.17)	> 1	-.41
lint %	-.13 (.11)	-.50 (.08)	> 2	-.52
weight/boll	-.14 (.17)	.22 (.14)	> 1	.41
fiber strength	.36 (.13)	.45 (.10)	< 1	.12
fiber elongation	.15 (.18)	-.07 (.15)	< 1	--
fiber fineness	.12 (.15)	.35 (.12)	> 1	--

\* Observed in 3 independent populations. Reported by Miller et al. (13).

in the two populations are presented in Table 5. Since correlation coefficients are ratios of covariances and variances which would be affected similarly by the difference in inbreeding between the two populations, no adjustments for inbreeding are necessary for comparison of the observed values.

Genotypic correlation coefficients are presented in two groupings. The first group includes all possible pairs among six traits, excluding fiber length. The "r" values for the original population were relatively high and all but 3 of the 15 correlations decreased in magnitude (ignoring sign) after the 6 generations of intermating. Of considerable interest in this population, for example, was the change in the genotypic correlation between yield and fiber strength from  $-.69$  in the original population to  $-.35$  after intermating. The second group of correlations was that involving fiber length with the other six traits. These "r" values were generally small in the original population but increased in magnitude (ignoring sign) in the intermated population. Thus, for example, the correlation between yield and fiber length changed from  $.02$  to  $-.37$ .

## DISCUSSION

### Assumptions

Interpretation of the observed changes of means and variances in the population following the six generations of intermating depends on the validity of the assumptions of the pertinent comparisons. The assumption of 50% self-pollination in the isolation block appears reasonable. Simpson (16) reported approximately 50% natural crossing in cotton in parts of the southeastern region of the Cotton Belt, with an average of 49% for the Raleigh, N. C. location. Even if the actual proportion of selfing was somewhat higher or lower than 50% (e.g. 35 to 65%), such differences would have only a minor effect on the inbreeding adjustments for the present study. For example, the average selfing would have to be of the order of 10% for selfing alone to explain the change in variance for yield. Furthermore, the deviations in the three cases where the deviations from "expected"

Table 6. Theoretical genetic variances for the original and intermated populations relative to the genetic variance under linkage equilibrium.

Chromosome map length, s	Number of loci, n	q = 0.5		q = 0.9	
		Original	Intermated	Original	Intermated
0.5	2	.64	.93	1.10	1.01
	11	.32	.62	5.10	3.30
	large	.26	.51	--	--
1.0	2	.86	1.00	1.04	1.00
	11	.50	.80	4.02	2.20
	large	.44	.72	--	--

Table 7. Predicted effects of selection for a 35% increase in fiber strength over adapted parent in original and intermated populations.

Population	Lint yield, kg/ha	Lint %	Wt. per boll, g	Fiber		
				length UHM	strength T <sub>1</sub>	elongation E <sub>1</sub>
Original	639	33.9	6.18	1.14	2.39	5.9
Intermated	750	33.5	6.10	1.17	2.39	6.2
Int.-Orig./Orig., %	+ 8.9	-1.2	-1.3	+2.6	--	+5.1

intermated variances were larger than the standard error (Table 4) were of such a nature as to be unexplainable by inbreeding alone. In two cases, there was a reduction in variance with intermating which is inconsistent with the effects of inbreeding and, in the other case, the variance of the intermated population was more than twice the variance of the original population, the maximum increase in variance if  $F = 1$  for an additive model. Also, the inconsistency of the change of variance from the original to the intermated population is evidence in itself that factors other than inbreeding are operative. That is, the inbreeding effect on variances would be similar in nature for the different characters in the absence of linkage, selection, and epistasis.

Comparison of variance components of the original and intermated populations assumes that the genotypic variance among progenies was due entirely to additive effects. Ramey's data (15) were cited in support of this assumption. Miller and Marani (12), in a diallel cross analysis of eight American Upland cotton strains, also concluded that the major portion of the genetic variance was additive. White (17), in diallel analyses of various quantitatively inherited characters, reported that yield and boll size were the only characters for which dominance was significant and that epistasis was not operating to control any of the characters measured. Although extensive and critical data on this point are not available for cotton, it appears that the assumption of additivity may be the most reasonable one available from present information.

### Means

The observed increase in the yield of the intermated population as compared with the original suggests natural selection occurred during the intermating generations. Although an equal number of seed were harvested from each plant in the intermating block, there was opportunity for selection at germination and seedling establishment as well as flowering time. It is estimated that the plants established in the field represented about 1/3 to 1/2 of the seed planted. No information is available as to how much, if any, of this seedling establishment differential might be related to genotypic differences in mature plant yield. It would seem logical to suppose, however, that such

a relationship might account for at least a portion of the increased yields of the intermated populations. Likewise, it might be supposed that the more vigorous and higher yielding plants produced more pollen at flowering time and thus contributed more than their proportional share of gametes to the succeeding generation.

Changes for characters other than yield could be accounted for by correlated responses. In all cases, the changes were of the magnitude and direction expected for changes in unselected traits due to the observed genotypic correlations between yield and these traits (Table 5).

### Variances

Genotypic variances decreased for six of the seven traits during the intermating generations. For fiber length, on the other hand, there was a sizeable increase in variance. If the assumptions of 50% random mating and predominance of additive genetic variance are realistic, these observed changes in variances could reflect either changes in linkage disequilibrium or changes in gene frequency which occurred during the successive intermating generations.

Linkage disequilibrium will be considered first. If a number of loci with interrelated effects on a trait were linked in the original parents and the coupling and repulsion-phase linkages were not in balance, genetic variances would be expected to move with intermating towards the linkage equilibrium values. Furthermore, studies involving an adapted genotype and a primitive or exotic genotype might be expected to involve gene frequency values near one or zero and extreme linkage effects would be expected. Certainly this might be the case in the present population which resulted from crossing the adapted Empire strain with a derivative from *G. arboreum*, an exotic species, and *G. thurberi*, a wild species. Magnitudes of the variances would be expected to decrease if coupling-phase linkages predominated but to increase if repulsion linkages were of major importance. In view of the wide spread between the parental strains for all traits other than fiber length, coupling-phase linkages might be expected to predominate for these characters in the population under study. For fiber length, on the other hand, the similarity in the phenotypes of the parents with significant segregation in the  $F_2$  generation suggests repulsion-phase linkages. Thus, any breakup of initial linkage blocks that might have been present would be expected to decrease variances of the former traits and to increase variance for fiber length. The direction of the observed changes is in agreement with such expectations.

It is of interest to examine briefly the theoretical effects of six generations of intermating, 50% selfing, and 50% random mating. The developments of Hanson (9, 10) and Hanson and Hayman (11) were used to develop a description of the effects of intermating for systems with an arbitrary degree of mixed selfing and random mating and are being published separately (Rawlings, manuscript in preparation). For a system involving a cross between two homozygous parents, the average effective map length,  $s'$  (i.e., the map length required to give the same amount of recombination in one meiotic division), after six generations of 50% selfing and 50% random mating, is

83% as long as that for complete random mating. The average effective map length with 6 generations of complete selfing is 56% as long as that for complete random mating. Thus, 50% selfing has the net effect of reducing effective recombination by 17%, as compared to complete random mating. Any degree of selfing increases the average segment length remaining. However, with 50% selfing, the increase in remaining segment length after six generations is approximately 1/3 as much as with complete selfing for true chromosome map lengths between 0.5 and 2.0.

Quantification of these differences in terms of their effects on genetic variances requires a more complete specification of the genetic system. For purposes of comparison, it is assumed that there are "n" equally spaced loci on a genetic map length "s," with all loci contributing equally to the trait. Further, q% of the favorable alleles come from the best parent. That is, if  $q = 0.5$ , the parents are equal in performance and there will be maximum repulsion linkages. If  $q = 1.0$ , there is maximum difference between the parents and maximum coupling linkages. Table 6 gives the theoretical genetic variance present relative to that present under linkage equilibrium for  $s = 0.5, 1.0$ ;  $n = 2, 11$ ; and  $q = 0.5, 0.9$ . These values are computed using the relationship developed by Hanson and Hayman (11), the relative genetic variance =  $n(2q-1)^2(1-2p) + 2p$ , where p, the average recombination for the mating system, was determined for 50% selfing and 50% random mating.

These figures illustrate the degree of difference in variances that might be expected between the original and intermated populations and how much additional change that might be expected with continued intermating to linkage equilibrium. It is clear that the amount of increase observed for fiber length, 79% is compatible with the increases expected in a repulsion system,  $q = 0.5$ , and the decreases of 20 to 50% for the other traits are equally acceptable for a coupling system. If the number of loci is sizable, the intermated population after six generations still retains a sizable amount of linkage disequilibrium.

In respect to changes in gene frequency, comparisons of the means indicated that at least some change occurred for certain traits, particularly yield and fiber strength. Since the original population was a cross of two homozygous parents, initial gene frequency can be assumed to have been 0.5. Assuming that additive genetic variance predominated, any change in gene frequency would be expected to decrease genotypic variance (7). Thus, change in gene frequency as well as possible breakup of initial linkage disequilibrium could account for the observed results for those six traits for which the variances decreased. In the case of the fiber length, however, change in gene frequency could not contribute to the observed increased genetic variance.

### Genotypic Correlations

If loci affecting two different traits were linked in the original parents and the coupling and repulsion phases of such linkages were not at equilibrium, the genotypic correlation arising from such linkages would be expected to move toward the equilibrium value with intermating. As mentioned previously, extreme linkage effects might be expected in the wide cross

under study. The average correlation values for experiments which involve random pairs of homozygous parents should estimate the coefficients for equilibrium condition. Miller, et al. (13) reported genotypic correlations between several pairs of traits for three independent cotton breeding populations representing adapted Cotton Belt material. The average "r" values from the three populations are listed in column 3 of Table 5. For those pairs of traits for which comparative data are available, there is a general tendency for the "r" values of the original population of the study to shift toward these average values with intermating. If it can be assumed that the average "r" values from the three populations are representative of equilibrium values (or at least closer to equilibrium values than those of the original population of this study), this suggests that the shifts observed after intermating reflect a breakup of initial disequilibrium. It is recognized that the correlation values from only three populations is a very restricted sample of equilibrium values.

Observed changes in the genotypic correlation coefficients could likewise be due to changes in gene frequency during the intermating generations. In those cases where substantial changes in means were observed, it is not possible to separate the relative importance of these two factors in shifting the correlation coefficients.

#### Implications on Selection Programs

The extent to which linkage blocks are broken and to which genetic recombination occurs is of critical interest to the plant breeder, since the success of his breeding program depends upon obtaining desirable character recombinations. Following the crossing of two inbred lines, intermating of the original  $F_2$  population for several generations prior to selfing and selection should insure a degree of breakup in the linkage groups. Predicted effects of one generation of selection in both the original and intermated populations were examined in the present study. In order to make a direct comparison, it was assumed that the selection in both populations would be for a level of fiber tensile strength which would be 35% higher than that of the standard commercial parent. Applying the usual formulae (13), it was possible to calculate the expected change in the remaining unselected traits, utilizing the variance and covariance estimates from the analyses of each population. Additive gene action is assumed for such predictions. Results of these calculations are presented in Table 7. With sufficient selection intensity in each population to obtain a fiber strength of 2.39, lint yield of the group of selections from the intermated population is predicted to be 8.9% higher than those from the original  $F_2$  population. Similarly, increases of 5.1% in fiber elongation and 2.6% in fiber length are predicted. Such changes

favoring the intermated population would be desirable. Lint percentage and weight per boll would be expected to be slightly less in the selections from the intermated population than from the original. Neither of these traits, however, are of direct economic importance. It appears that the intermated populations should provide a better source of material for selection than the original  $F_2$  population, perhaps due to a partial breakup of linkage blocks in the original material. Hanson (9), on the basis of theoretical calculations, suggests that at least one or more and preferably four intermating cycles should precede selfing generations to insure a degree of breakup of the linkage groups and to increase the genetic recombination within the linkage groups. Present data tend to support this conclusion. Such intermating cycles may be of particular importance in wide crosses involving an unadapted parent.

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