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Heterosis and Combining Ability in Varietal Top Crosses of Upland Cotton, *Gossypium hirsutum* L.[1]

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DEVELOPMENT of methods for the commercial production of hybrid cotton seed is receiving considerable emphasis (2, 3, 4, 5, 9). Almost any method likely to be developed in the foreseeable future, however, would undoubtedly increase seed costs. Hence, in order to evaluate the economic feasibility of producing hybrid seed by any of these methods, specific information is needed concerning the performance of hybrid cottons relative to currently produced commercial varieties. If hybrid seed production appears to be feasible, it would also be desirable to study selection criteria for parental strains.

Variable results have been reported concerning the amount of heterosis exhibited in crosses among varieties and strains of Upland cotton. Miller and Marani (6) and White and Richmond (10) have reviewed the earlier literature concerning this subject. Generally, certain crosses exhibit substantial heterosis for yield and certain components of yield as measured by comparison of the F_1 with the average performance of the parental lines. When such F_1's are compared with the best of all parents in the study or with the standard commercial checks, however, there are very few instances of significantly superior performance of the hybrids. It should be pointed out that many of these studies have involved noncommercial types of material since the hypotheses being tested often impose such limitations as homozygosity of the parents, presence of marker genes, etc. Furthermore, due to the difficulty of producing adequate amounts of hand pollinated seed, even those studies involving acceptable agronomic types generally have been rather limited in terms of number of crosses studied, number of replications, and repetition over environments.

The present investigation was initiated in 1958 at the time the chemical compound 2,3-dichloroisobutyrate showed promise as a male gametocide (1). With the immediate possibility of producing hybrid seed on a commercial scale, information on the combining ability of existing varieties and strains was desired. Based upon information reported on hybrid corn breeding, it appeared that the most efficient approach in the preliminary screening of materials for desirable combining ability would be top-cross tests. Accordingly, such a program was inaugurated. Results to be presented concern *(1)* agronomic performance of certain top-cross combinations relative to standard varieties, *(2)* criteria for the selection of parents which might be expected to produce superior hybrids, and *(3)* observations on heterotic effects under different environments.

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METHODS AND MATERIALS

The variety 'Coker 100A' was chosen as the tester parent. A group of 22 varieties and strains designated as "parental varieties" were crossed to this tester. Thirteen of these parents were commercial releases from various public and private breeding agencies in the Southeastern and Delta regions of the cotton belt. The remaining parents were noncommercial strains of diverse origins which had been maintained by bulked self-pollinations at the North Carolina Experiment Station for several years. All crossed seed were produced by hand pollination.

In 1959, the first year of yield tests, 9 parental varieties, their topcross hybrids, and entries of the tester parent were evaluated at 3 locations, Lewiston, Rocky Mount, and Salisbury, North Carolina. In 1960 and 1961 these same 9 sets of material as well as an additional 13 sets were grown at Lewiston and Rocky Mount. Three replications were grown for all tests. A split-plot design was used with each parental variety, the tester variety, and the F_1 generation of the respective topcross hybrids making up the subplots. Subplots were 1 row, 15 feet in length, thinned to a final stand of 3 plants per foot of row.

Data were recorded on lint yield, lint percent, and weight per boll from all plots and all tests. 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. In 1960, fiber samples were taken from 2 replications of each test and analyzed by the U. S. Department of Agriculture Fiber Laboratory at Knoxville, Tenn. to obtain information on fiber quality. Fiber property measurements were as follows:

- Fiber length, U.H.M. (Upper Half Mean)—Length, in inches, of the half of the fibers, by weight, that contains the longer fibers.
- Fiber length, M (Mean)—Average length, in inches, of all fibers longer than 1/4 inch.
- Fiber strength, T_1 units—Strengtth of a bundle of fibers measured on the Stelometer with 2 jaws holding the fiber bundle separated by an 1/8-inch space. Strength is expressed in terms of grams per grex.
- Fiber elongation, E_1 units—Percentage elongation at break of the center 1/8 inch of the fiber bundle measured for T_1 strength on the Stelometer.
- Fiber fineness, Micronaire—Fineness of the sample measured by the Micronaire and expressed in standard (curvilinear scale) Micronaire units.

Data were summarized and analyzed on the basis of two groupings of the material. One grouping included the 9 sets of parents and hybrids common to all 7 tests over the 3-year period. The second grouping included the 22 sets tested at 2 locations each in 1960 and 1961. Analyses of variance were made only for lint yield. In the combined analyses each test was treated as an "environment" with no attempt to separate year and location effects as such.

RESULTS

Average Heterosis for Different Traits

Comparisons of average variety and top-cross performance for various yield and fiber quality traits for the two groupings of the material are presented in Tables 1 and 2. The top-cross hybrids on the average had higher lint yields and larger bolls than the parental varieties. In respect to lint percentage and the various fiber properties, however, average top-cross performance was very similar to that of the mean of the mid-parent values. Data from the individual top crosses and their corresponding parents are in close agreement with the overall average results, i.e., topcross values deviated substantially from their corresponding midparent value only for lint yield and boll size. The

Table 1. Mean performance and comparisons of parental varieties, the tester parent, and top-cross hybrids of 22 sets of cotton material evaluated at 4 environments.

	Lint, lb./A.	Wt. /boll, g.	Lint %	Fiber†				
				Length		Strength, T_1	Elongation, E_1	Fineness, Micronaire
				U.H.M.	M.			
Mean of								
Parental varieties	717	6.38	38.9	1.04	0.87	1.76	7.8	4.73
Tester parent*	814	6.21	39.6	1.05	0.88	1 69	8.1	4.77
Top-cross hybrids	904	6.59	39 4	1 05	0.87	1 69	8.0	4.82
Mid-parent values	766	6.30	39.2	1.04	0.88	1.72	8.0	4.75
Top crosses as								
% of midparents	118.0	104.6	100.5	101.0	98.9	98.2	100.0	101.5
% of tester	111.0	106.1	99.5	100.0	98.9	100.0	98.8	101.0

* Tester parent included in each whole plot (see text). † Fiber data obtained from 2 replicates at each of 2 environments only.

Table 2. Mean performance and comparisons of parental varieties, the tester parent, and top-cross hybrids of 9 sets of cotton material evaluated at 7 environments.

	Lint, lb./A.	Wt. /boll,	Lint %	Fiber†				
				Length		Strength, T_1	Elongation, E_1	Fineness, Micronaire
				U.H.M.	M.			
Mean of								
Parental varieties	616	6.19	37.8	1.03	0.86	1.77	7.8	4.67
Tester parent *	669	6.04	37.7	1.06	0.88	1.70	8.1	4.77
Top crosses	769	6.46	37.8	1.05	0.87	1.70	8.0	4.78
Mid-parent values	643	6.12	37.8	1.05	0.87	1.74	8.0	4.72
Top crosses as								
% of midparents	119.6	105.6	100.0	100.0	100.0	97.7	100.0	101.3
% of tester	114.9	107.0	100.3	99.0	98.9	100.0	98.8	100.2

* Téster parent included in each whole plot (see text). † Fiber data obtained from 2 replicates at each of 2 environments only.

remainder of this report will concern only lint yield since little economic significance can be attached to the differences in boll size observed in these tests.

Heterosis for Yield in Individual Top Crosses

Yield data for the individual varieties, the average of the tester parent and each top-cross hybrid are presented in Tables 3 and 4. Appropriate least-significant-differences are also presented. In both groupings of material all top-cross hybrids appeared to be significantly higher yielding than their corresponding parental varieties. A more meaningful comparison, however, is that of the top crosses with the tester parent Coker 100A, since this variety was generally the higher yielding parent of each top-cross hybrid. Yields of the top crosses ranged from 100 to 128% of the yield of Coker 100A. Appropriate LSD values indicate that such yield differences were statistically significant in over 1/2 of the top-cross versus tester-parent comparisons for the material tested at 4 environments and in over 2/3 of the corresponding comparisons for the material grown at 7 environments. Thus certain individual top-cross hybrids were clearly superior in yield to the best varieties grown in the test. Other top-cross hybrids were only average or slightly above average relative to the better varieties.

Relationship Between Varietal Performance and Top-Cross Performance

The data in Tables 3 and 4 suggest a close relationship between varietal performance and the performance of those varieties when top-crossed to a common tester parent. Simple correlation coefficients measuring this relationship were calculated. The design of this experiment, such that the variety and its top-cross hybrid always occurred together in the same whole plot in the field, introduced an upward bias in the covariances and variances. It was possible, however, to estimate the magnitude of this bias factor from the mean square expectations of an analysis of variance of the whole plots where only parent-variety and top-cross data are used (Coker 100A tester parent excluded). The estimates of these correlation coefficients using the unbiased variances and covariances[3] are presented in Table 5. The "r" values of +.66 and +.58 for the 2 groupings indicate a close relationship between the varietal performance *per se* and the performance of those varieties in top crosses with a common variety.

Table 3. Mean yields of each of 22 varieties of cotton, their top-cross hybrids, and the tester parent grown at 4 environments.

Variety or strain	Lint yield, lb./A.†		Top-cross yield as % of tester	Variety or strain	Lint yield, lb./A.		Top-cross yield as % of tester
	Variety	Hybrid			Variety	Hybrid	
Coker 100A (tester)*	814	--	--	Rex	675	899	110
M8	786	1019	125	Z 106	639	898	110
Coker 124	820	1017	125	Fox	676	892	110
Plains	807	989	122	Acala 5675	614	880	108
Auburn 56	824	942	116	Moore Special	612	867	106
G-4	789	933	115	TH 149	686	858	105
Pope	773	931	114	TH 73	662	849	104
Stoneville 7	748	930	114	T 317	749	843	104
A-1	716	930	114	V 107	715	842	103
Moore 33	682	913	112	Empire	706	835	103
Stardel	752	902	111	Dixie King	674	814	100
V-3	675	900	111				

* Average of all entries of the tester parent which was included in each whole plot.

† LSD's for comparison of:		.05	.01
	Any entry vs. tester	79	104
	Variety vs. its top cross	92	121
	Entries in different whole plots	115	150

Table 4. Mean yields of each of 9 varieties of cotton, their top-cross hybrids, and the tester parent grown at 7 environments.

Variety or strain	Lint yield, lb./A.†		Top-cross yld. as % of tester	Variety	Lint yield, lb./A.		Top-cross yld. as % of tester
	Variety	Hybrid			Variety	Hybrid	
Coker 100 A (tester)*	669	--	--	G-4	646	766	114
Pope	684	855	128	Empire	633	732	109
M8	652	850	127	Dixie King	614	730	109
Moore 33	552	793	119	Acala 5675	515	712	106
Auburn 56	697	782	117	Fox	554	697	104

* Average of all entries of the tester parent which was included in each whole plot.

† LSD for comparison of:		.05	.01
	Any entry vs. tester	53	69
	Variety vs. its top cross	65	86
	Entries in different whole plots	79	104

Table 5. Simple correlation coefficients measuring the relationship between parental yields and yields of their hybrids.

Data source	"r" value
Parent variety vs. top-cross hybrid	
Top crosses of 22 varieties in 4 environments.	+0.66**
Top crosses of 9 varieties at 7 environments.	+0.58
Diallel cross study† -- 28 mid-parent values vs. 28 F_1 hybrids	+0.78**

** Significant difference from zero at 1% level. † See text.

Additional information on the relationship between parental and hybrid performance is available from a study of an unrelated group of material which included eight inbred lines and all possible F_1 hybrids among them. The genetic materials used and the pertinent procedures have been presented elsewhere (6). For the present report a correlation coefficient was calculated to express the relationship between midparent yields and their corresponding F_1 hybrid yields. The "r" value of +.78 supports the present findings of a close relationship between parental and hybrid performance in respect to yield.

Using a different approach to further examine the relationship in question, analyses of variance of the present top-cross tests were made to examine the interaction of of heterosis × crosses. For this purpose heterosis was measured as the difference between each top-cross hybrid and the mean of its two parents. Although in all cases the top-cross hybrid was significantly higher yielding than

[3] The assistance of Dr. J. O. Rawlings, Department of Experimental Statistics, North Carolina State, in the development of the pertinent procedures is gratefully acknowledged.

the average of its two parents, the interaction among crosses was nonsignificant in both groupings of the material; that is, F_1 minus mid-parent values were not significantly different for the different crosses. Since there were significant differences among the different top-cross hybrids, the results of these analyses suggest that the major portion of the differences between hybrids may be associated with the differences among the original mid-parental values rather than differences in the amounts of heterosis expressed in the different crosses. These analyses lend further support to the contention of a close relationship between the performance of the parental varieties *per se* and the performance of the hybrids between them.

Heterosis Under Different Environments

Casual inspection of earlier data collected in North Carolina had suggested that greater heterotic responses may occur under unfavorable environments than when the material is grown under conditions approaching a more optimum environment for high yield. Data are presented in Table 6 on the amounts of heterosis averaged across all entries at the different environments at which the parental material was grown. Heterosis is expressed as the difference between average top-cross hybrid yields and the average of the mid-parent values on a pounds of lint per acre basis. This also is expressed as a percentage of the mid-parent value. Environments are listed in the table in ascending order of yield with a range from less than 1 bale per acre to more than 2 bales per acre. Examination of heterosis expressed on a pounds per acre basis does not reveal any consistent pattern of response. An analysis of variance of heterotic effects (F_1 minus mid-parent values) over the different environments indicated that although the average heterotic effects were highly significant at each environment, there were no significant differences in the magnitude of heterosis recorded for the different yield-level environments.

It should be noted, however, that when heterosis is expressed on a percentage basis, there is a marked tendency for the percentage increase in yield over the mid parent to decrease as average yield level increases from one environment to another. This, of course, is expected if the actual amount of heterosis on a pounds per acre basis remains essentially constant over a range of yield levels.

DISCUSSION

Present data indicate that certain hybrids are clearly superior in lint yield to the best varieties. A conservative estimate of the magnitude of this superiority might be of the order of 15 to 20%. Since seed cost and somewhat higher harvesting costs would be the only additional investment required for a potentially increased return of this magnitude, such information can be used to evaluate the economic feasibility of various methods of hybrid seed production. A great deal more information and developmental research is needed, however, concerning the methodology of producing F_1 hybrid seed in cotton (4, 7, 8).

If efficient procedures for the production of hybrid seed are developed in the future, information will be needed concerning procedures for evaluating and choosing the parents of hybrids. Present data indicate that the magnitude of heterosis is largest for lint yield, medium for boll size, and of very minor consequence for lint percentage and the various fiber properties. Obviously, potential parents must perform at acceptable levels for these latter traits if the hybrid is to be commercially acceptable. The major advantage expected from hybrids would be increased yield.

Table 6. Average heterosis for lint yield for all crosses at each of the different environments.

Environment*	Average of 9 crosses			Average of 22 crosses		
	Mid-parent yield, lb./A.	Top cross minus mid-parent lb./A.	Heterosis, % increase over mid-parent	Mid-parent yield, lb /A.	Top cross minus mid-parent, lb./A.	Heterosis, % increase over mid-parent
1	455	82	18.0	-	-	-
2	468	122	26.1	-	-	-
3	486	140	28.8	-	-	-
4	533	148	27.8	533	131	24.6
5	735	116	15.8	708	127	17.9
6	742	130	17.5	749	126	16.8
7	1082	141	13.0	1070	171	16.0

* Environments listed in ascending order of mid-parent yield.

Data from the present study suggest that one of the primary criteria for choosing parents which might result in superior yielding hybrids would be the performance of the parental varieties or strains *per se*. Considering the grouping of the 22 varieties grown at 4 locations, the 5 highest yielding top-cross hybrids were those involving the 5 highest yielding parental varieties. It is recognized that the use of only one tester stock in the present study imposes a restriction on the interpretation of these observations. It would appear on the basis of the present data, however, that little is to be gained from testing hybrids in which one or more of the parents are clearly inferior in yield to the standard check varieties with which they would be compared. Preliminary screening of materials for potential hybrids might be entirely on the basis of the performance of the strains or varieties themselves, taking all economic characteristics into consideration. This procedure would permit the elimination of a large amount of undesirable parental material and thus avoid the necessity of testing numerous hybrid combinations. It should be pointed out, of course, that the correlation between parent-variety performance and top-cross performance is obviously less than unity. Although no significant differences were detected in the array of heterotic effects (F_1 minus mid-parent values) for the different crosses, differences of the magnitude observed might prove to be statistically significant with the use of more precise experimental techniques. Thus the final stage of testing would still necessitate actual evaluation of F_1 combinations among the screened parents.

The finding that heterotic effects were not significantly different for different crosses or for the average of all entries, when grown in different environments, was somewhat surprising. Perhaps this only reflects a lack of precision in detecting relatively small differences of the order observed. At any rate such differences did not appear to be a major source of variation in the present tests. Yield differences between hybrids appeared to be primarily associated with differences in the respective mid-parent values. This observation as well as the high parent-topcross correlation suggest that additive gene action might be of considerable importance in the determination of yield. If this should be so, the most efficient breeding and seed production system should be one that maximizes additive gene effects as well as heterotic effects.

Since heterosis in pounds of lint per acre tended to be constant in all top crosses, it follows that the highest *percentage* increase in yield of F_1 over parents often occurred in crosses between lower yielding lines. This is in general agreement with previously cited studies and emphasizes the need for careful interpretation of heterosis data expressed on a percentage basis.

SUMMARY

Comparisons of top-cross hybrids with their parental varieties are presented for 9 crosses evaluated at 7 environments and for an additional 13 crosses grown at 4 environments. Data were obtained on lint yield, lint percentage, weight per boll and various fiber property measurements. The top-cross hybrids generally had higher lint yields and larger bolls than the parent varieties. In regard to lint percentage and the various fiber traits, however, hybrid performance was very similar to the mid-parent value. Certain individual top-cross hybrids were clearly superior in lint yield to the better varieties included in the test. A conservative estimate of the magnitude of this superiority would be of the order of 15 to 20%.

A close relationship was observed between the yield of the parental varieties *per se* and the yields of their respective top-cross hybrids. Simple correlation coefficients expressing this relationship were +.58 and +.66 for the two groupings of the material. An analysis of F_1 minus mid-parent values revealed that although in all cases each top-cross hybrid was significantly higher yielding than the average of its two parents, the amount of heterosis was not significantly different for different crosses. Thus the observed differences among hybrids appeared to be associated primarily with differences in the "base" performance of the parental varieties *per se,* rather than with differences in the amount of heterosis expressed in the different crosses.

Observations on heterotic effects (F_1 minus mid-parent values) at different environments indicated that the magnitude of heterosis on a pounds per acre basis remained rather constant over a range of yield levels.

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