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# Inheritance of Seedling Root Length and Relative Root Weight in Cotton<sup>1</sup>

A. M. Eissa, J. N. Jenkins, and C. E. Vaughan<sup>2</sup>

## ABSTRACT

A perennial problem in cotton, *Gossypium hirsutum* L., involves obtaining a vigorous, uniform stand when planted early. Strains that grow roots longer and higher in relative root weight (mg/mm length) than currently used cultivars when temperatures are cool may help alleviate this. Using the standard cotton, cool temperature (18°C) germination test we determined root lengths and relative root weights for 124 day-neutral composite F<sub>3</sub> strains. Variability among lines was significant for each seed and seedling parameter. Fixed model diallel analyses of five strains indicated that among the five strains one would have difficulty in selecting for long roots with high relative root weight. Generation mean analysis indicated from one of two crosses that large amounts of additive, dominance, and additive by additive epistatic effects were present for the two primary root traits. One should thus be more successful if selection is delayed to the F<sub>3</sub> to allow genetic recombination of additive and additive by additive epistatic genes to occur. Recurrent selection should be a useful method of breeding. The cross of strains 3822 × 3851, involving accessions T-214 and T-225, respectively, should be useful for developing lines with root length equal or longer than ST 213 and relative root weight greater than ST 213. We speculate that plants with long roots and a high relative root weight should possess increased levels of resistance to seedling disease through a mechanical type of resistance; i.e., the plants should have more root tissue to slough off as diseased tissue and yet maintain a viable root system and a healthy plant than would plants with short roots with a low relative root weight.

**Additional index words:** Seedling disease, Cotton genetics, Diallel analysis, Generation mean analysis.

**E**STABLISHMENT of a uniform, vigorous stand is a perennial problem in cotton, *Gossypium hirsutum* L., production. A combination of early planting and a good healthy stand generally enhances early maturity, insect control, and yield. However, cotton planted early in the season is often subjected to a period of unfavorable temperatures resulting in poor stands.

Germination at low temperatures has received the attention of a number of investigations. The minimum temperature for germination is 12°C with better activity at 15°C (Ludwig, 1932). *G. barbadense* L. germinates better at the low 12°C temperature than *G. hirsutum* (Marani and Dag, 1962). Seed which are acid delinted germinate better at low temperatures (Marani and Amirav, 1970). There seems to be a potential for developing cotton lines with improved germination at cool temperatures (Buxton and Sprenger, 1976).

Investigations of the magnitude and nature of gene action involved in the inheritance of seed and seedling traits indicated multiple gene action (Balls, 1919;

Ware, 1936; Al-Rawi and Kohel, 1969; and Abd-Alla, 1970). Seed weight had large additive and dominance components (Bourland, 1980). There is very little genetic information available on the inheritance of root length and weight per mm of length.

Information to predict consequences of various breeding procedures and to make more accurate decisions about effective breeding practices should come from experiments designed to determine the relative magnitude of additive, dominance, and epistatic variances.

The objectives of this research were to: a) evaluate F<sub>3</sub> day-neutral composites from a cross of 124 exotic cottons with an upland parent for variability for root length and relative root weight when germinated at 18°C, b) determine gene action involved in inheritance of cotton root length and relative root weight when germinated at 18°C, and c) determine the feasibility of genetically combining long roots with high relative root weight.

## MATERIALS AND METHODS

### Evaluation Studies

Seeds of six cultivars and day-neutral F<sub>3</sub> composites from crosses of 124 exotic accessions of short day *G. hirsutum* with 'Deltapine 61' were planted on the Plant Science Farm at Mississippi State, Miss. in a Leeper silty clay loam (fine, montmorillonitic, nonacid, Thermic Chromodertic Haplaquept). A randomized complete block design with two replicates was used with each entry planted in a single-row 19 m plot. At harvest two bolls were harvested from each plant for a total of 80 bolls per row. Bolls were harvested within 10 days after at least one boll on each plant had opened.

Cottonseed from each strain plus six cultivars were germinated on rolled paper towels using the standard cool germination test (Christiansen, 1964; McCarter and Roncadori, 1970, 1971) with four replicates of 100 acid-delinted seed from each entry. Each sample was placed randomly on two double-layered 38.5 × 25.6 cm moist germination paper towels. One additional towel was placed over the seed before rolling. The rolled towels were put in a plastic box and placed in a dark germinator at a constant temperature of 18°C. After 4 days, 35 germinating seeds having radicle length of 1.25 cm were chosen from each replicate. To measure root growth, the 35 germinating seeds were transplanted to another germination paper which was then rolled and put in a plastic box and placed back in the germinator for 12 more days. After 16 days the length of the primary root of each seedling was measured. Wet and dry weights of roots and the top part of the plant were determined. To determine total dry weight of roots and tops of 35 seedlings, they were dried separately at 105°C to a constant weight. Relative root weight was calculated as follows: dry weight roots (mg)/length of roots (mm). To determine root length each of the 35 roots was measured individually. We recognize that relative root weight could be a result of more dense cell material or a larger diameter root.

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Seed weight (weight of 100 acid-delinted seed), seed volume (cc displacement of methanol by 100 seed), and seed density (ratio of seed weight to seed volume) were determined using four replicates of 100 seeds each.

Differences among the lines were determined by the analysis of variance. All possible subset regression from BMDP Statistical Software, Inc.<sup>3</sup> was used to determine the relationship among traits. Coefficients of correlation among the characters were also determined.

### Inheritance Studies

Seven strains were selected from the evaluation study for use in the inheritance study. In the summer of 1979, 'Stoneville 213' (ST 213) and F<sub>4</sub> progeny from four lines, 3933, 3984, 3777, and 3900, were crossed in the possible 10 hybrid combinations without reciprocals. These strains represent day-neutral selections from accessions T-251, unknown, T-177, and T-297, respectively.

In the summer of 1979, ST 213 was crossed with F<sub>4</sub> progeny from 3768 and strain 3822 was crossed to 3851. These represent day-neutral selections from accessions T-154, T-214, and T-225, respectively. These strains and their F<sub>1</sub> hybrids were grown in the winter nursery at Iguala, Mexico where F<sub>1</sub>'s were selfed for F<sub>2</sub> and backcrossed to the parental strains. F<sub>1</sub> hybrids between parents were made again and the four parent strains were selfed. By this procedure seeds of all generations (P<sub>1</sub>, P<sub>2</sub>, F<sub>1</sub>, F<sub>2</sub>, BCP<sub>1</sub>, BCP<sub>2</sub>) were produced in the same environment at Iguala.

In the summer of 1980, all 10 diallel cross F<sub>1</sub>'s and parents as well as the six generations from each of the two sets of crosses utilized in the generation mean analysis were evaluated for root length and relative root weight using the cool germination test at 18°C reported herein. Six replications were used. Characters measured were seed weight, seed volume, seed density, wet weight of tops, wet weight of roots, dry weight of tops, dry weight of roots, and root length and relative root weight.

The fixed model diallel analysis developed by Griffing (1956a, 1956b) and the generation mean analysis procedure developed by Hayman (1958, 1960), as modified by Meredith and Laster (1975), were used to estimate relative amounts and types of gene action associated with all characters in these specified crosses. Thus, by using fixed models and different parents in the diallel and the generation mean analysis we are evaluating specific parents and the results from the two methods may differ because of different parents being involved. This manuscript reports inheritance results only for relative root weight and root length since these were our primary interests. Data on the other traits are available (Eissa, 1981).

## RESULTS AND DISCUSSION

### Evaluation Studies

Data for mean, range, LSD at 0.05 level, and coefficient of variation for seed and seedling traits are shown in Table 1. The mean squares for strains were significant for all traits. Thus, genetic differences exist among strains for all traits. The ranges indicate large differences among strains. These strains should be useful in selecting parents for genetic studies.

There were significant correlations between wet weight of tops and roots with seed weight and seed

**Table 1. Statistical measures for seed and seedling traits of 124 cotton strains.**

Characters	Mean	Range	LSD 0.05	CV (%)
Seed weight, gm	10.86	8.67 - 13.15	0.25	1.65
Seed volume, cc displacement	10.76	8.02 - 15.62	1.27	8.51
Seed density, gm/cc	1.01	0.89 - 1.18	0.07	5.06
Wet wt. tops/plant, mg	747.60	471.43 - 919.14	63.94	6.17
Wet wt. roots/plant, mg	96.06	60.57 - 123.43	12.43	9.32
Dry wt. tops/plant, mg	48.69	37.14 - 62.85	5.89	8.73
Dry wt. roots/plant, mg	6.97	4.28 - 11.43	2.09	21.60
Root length/plant, mm	89.77	56.40 - 124.42	10.51	8.44
Relative root weight, mg/mm	0.079	0.046 - 0.121	0.03	22.91
Percent dry wt. of plant	6.63	5.59 - 9.68	0.01	8.66
Percent dry wt. of tops	6.55	5.61 - 9.41	0.01	9.31
Percent dry wt. of roots	7.30	5.00 - 9.68	0.02	21.40

volume. Seed density was highly correlated with seed volume but not seed weight. Relative root weight was significantly correlated with dry weight of roots, percent dry weight of roots, and percent dry weight of plants. There was a significant negative correlation (-0.41) between relative root weight and root length. Root length and wet weight of roots were significantly correlated (Table 2).

All possible subset regression analysis was utilized to determine the best set of traits to predict root length. Since relative root weight is calculated from root length it was not included in the subset. The coefficient of determination ( $r^2$ ) was 0.41 and significant when the independent variable was wet weight of roots. It increased to 0.46 when the wet weight of tops was included in the regression equation. No further increases were obtained with any other combinations of traits up to subsets including all 11 variables.

When relative root weight was the dependent variable in the regression analysis (root length was not included) the percentage dry weight of roots gave a  $r^2$  of 0.75 and dry weight of roots gave a  $r^2$  of 0.61. However, dry weight of roots is used to calculate relative root weight. Including both in the regression equation only increased the  $r^2$  to 0.76. No other combination of subsets increased  $r^2$ . It seems that percentage dry weight of roots could be utilized as a measure similar to relative root weight but one would still need to measure root length in order to select plants with long roots which have a high relative root weight. Thus, there were no seed or seedling traits more easily obtained than root length and relative root weight which could be used to accurately predict these two.

In Table 3 are shown data for the seven parental strains we utilized to study the inheritance and breeding behavior of root length and relative root weight.

### Inheritance Studies

The general combining ability (GCA) and specific combining ability (SCA) mean squares were significant for root length and relative root weight (Table 4). Length ranged from 88.19 to 108.18 mm and relative root weight ranged from 0.066 to 0.088 among the 10 F<sub>1</sub>'s (Table 5). The GCA and SCA effects were calculated and only the GCA effect for root length for parent ST 213 was significant with that for parent 3900 being almost significant at the

<sup>3</sup> Mention of a trademark or proprietary product does not constitute a guarantee or warranty of the product by USDA or the Mississippi Agric. and For. Exp. Stn., and does not imply its approval to the exclusion of other products that may also be suitable.

**Table 2. Coefficients of correlation among seed and seedling traits of 124 cotton strains.**

Traits	Traits											
	Seed wt.	Seed vol.	Wet wt. tops	Wet wt. roots	Dry wt. tops	Dry wt. roots	Root length	Total dry wt.	Dry wt. tops	Dry wt. roots	Seed density	Relative root wt.
Seed weight	--											
Seed volume	0.71*	--										
Wet wt. tops	0.68*	0.44*	--									
Wet wt. roots	0.31*	0.24*	0.36*	--								
Dry wt. tops	0.61*	0.43*	0.47*	0.18	--							
Dry wt. roots	0.16	0.16	0.10	0.33*	0.13	--						
Root length	0.09	0.13	0.03	0.64*	0.08	0.22*	--					
% total dry wt.	-0.03	0.03	-0.48*	-0.22*	0.51*	0.25*	-0.01	--				
% dry wt. tops	-0.03	0.02	-0.48*	-0.18	0.54*	0.04	0.02	0.97*	--			
% dry wt. roots	-0.00	0.03	-0.12	-0.22*	0.03	0.84*	-0.14	0.40*	0.16	--		
Seed density	-0.09	-0.73*	-0.02	-0.10	-0.02	-0.05	-0.15	-0.00	-0.01	0.01	--	
Relative root wt.	0.09	0.07	0.07	-0.10	0.06	0.78*	-0.41*	0.24*	0.02	0.87*	0.04	--

\* Significantly different from zero at 0.05 level.

**Table 3. Root characteristics of strains and ST 213 used in genetic studies.**

Strain	Accession no.†	Race†	Preliminary screening		Diallel analysis		Generation mean analysis	
			Length	Relative root wt.	Length	Relative root wt.	Length	Relative root wt.
			mm	mg mm <sup>-1</sup>	mm	mg mm <sup>-1</sup>	mm	mg mm <sup>-1</sup>
3933	T-251	Punctatum	128	0.067	128	0.070		
3984	Unnumbered	Unclassified	124	0.057	124	0.060		
3777	T-177	Latifolium	92	0.140	92	0.140		
3900	T-297	Latifolium	93	0.130	93	0.090		
3822	T-214	Latifolium	56	0.115	--	--	77.50	0.110
3768	T-154	Latifolium	124	0.046			112.94	0.066
3851	T-225	Latifolium	70	0.112	--	--	100.74	0.084
ST 213	Commercial	Unclassified	90	0.060	90	0.060	93.72	0.084

† Designation of accession number and race type for the short day parent used in developing the day-neutral strains.

**Table 4. Analysis of variance and combining ability analysis for root traits of 10 F<sub>1</sub>'s from a diallel cross in cotton.**

Source of variation	df	Root length	Relative root wt.†
		mm	mg mm <sup>-1</sup>
Replication	5	225.78	0.23
Crosses	9	229.04**	0.29**
GCA	4	393.01**	0.35**
SCA	5	97.87*	0.24*
Error	45	33.05	0.08

\*,\*\* Significant at the 0.05 and 0.01 levels of probability, respectively.

† Mean square × 10<sup>3</sup>.

0.05 level (Table 6). GCA effects for parents ST 213 and 3900 indicated that these generally transmitted their root length to their progeny; however, both have short roots (Table 3). The F<sub>1</sub> of 3900 × 3933 did, however, have the longest roots (Table 5); differences in root length were significant among the 10 F<sub>1</sub>'s and the mean of the F<sub>1</sub>'s involving 3900 was longer than those involving other parents.

Using the five parents involved in this diallel analysis we would expect to experience difficulties in combining long roots with high relative root weight in pure breeding lines.

Four parents were involved in the generation mean analysis. Only ST 213 was common between the two genetic studies. In both studies we are considering fixed models; i.e., we are making interpretations of the data for these specific parents since the parental strains were not randomly chosen, but were selected for combinations of root length and relative root weight. In the generation mean analysis there were highly significant differences among the means of the

**Table 5. Mean root length and relative root weight of 10 F<sub>1</sub>'s from a diallel cross.†**

Parental lines or cultivar	Parental line or cultivar					Mean
	3777	3900	3984	3933	ST 213	
3777	100.09	91.38	100.29	96.17	96.17	96.98
	0.078	0.079	0.066	0.078	0.078	0.075
3900		100.43	108.18	96.22	96.22	101.22
		0.067	0.067	0.079	0.079	0.073
3984			99.15	88.19	88.19	94.79
			0.074	0.080	0.080	0.075
3933				88.54	88.54	99.04
				0.088	0.088	0.074
ST 213						92.28
						0.081

† Upper numbers are length (mm) and lower numbers are relative root weight (mg/mm). LSD (P = 0.05) for a single cross means = 6.67, and 0.010 for root length and relative root weight, respectively.

**Table 6. General and specific combining ability effects for root length and relative root weight.†**

Parental lines or cultivar	GCA effects of parental lines or cultivar					GCA effects of parental lines
	3777	3900	3984	3933	ST 213	
3777	-2.75	-2.88	0.37	5.26	0.16	0.16
	0.64	0.46	-0.65	-0.45	-0.07	-0.07
3900		0.52	2.59	-0.36	5.82	5.82
		-0.39	-0.23	-0.02	-0.37	-0.37
3984			2.15	0.21	-2.77	-2.77
			-0.17	-0.24	-0.07	-0.07
3933				-5.11	2.90	2.90
				-0.71	-0.24	-0.24
ST 213						-6.11**
						0.75

\*\* Significantly different from zero at the 0.01 level of probability.

† Upper lines of numbers are for root length and lower lines of numbers are for relative root weight. Relative root weight effects are × 10<sup>2</sup>.

**Table 7. Means and generation mean analyses of genetic components for root length and relative root weight.**

Root traits	Variables†											
	Generation means						Genetic variance components					
	$\bar{P}_1$	$\bar{P}_2$	$\bar{F}_1$	$\bar{F}_2$	$BC\bar{P}_1$	$BC\bar{P}_2$	LSD 0.05	A‡	D	AXA	Res.	Error
<b>Cross 3822 × 3851</b>												
Root length (mm)	77.50	100.74	114.13	93.65	113.10	112.00	8.13	1236.13**	3016.98**	1428.84**	283.72*	46.69
Relative root weight (mg/mm)	0.110	0.084	0.072	0.098	0.063	0.066	0.008	13.94**	33.15**	44.89**	6.36**	0.47
<b>Cross 3768 × ST 213</b>												
Root length (mm)	112.94	93.72	109.70	100.06	97.68	119.56	9.68	164.53	158.44	297.22**	1190.83**	66.29
Relative root weight (mg/mm)	0.066	0.084	0.069	0.083	0.089	0.070	0.013	2.10	0.38	0.59	12.64**	1.13

\*,\*\* Significant at 0.05 and 0.01 levels of probability, respectively.

† Variance components for relative root weight  $\times 10^4$ .

‡ A = additive; D = dominance; AXA = additive  $\times$  additive epistasis; Res. = additive  $\times$  dominance and dominance  $\times$  dominance epistasis.

six generations of each set of crosses for both root traits (Table 7).

In the cross of 3822  $\times$  3851 additive and additive by additive epistasis effects accounted for about one-half of the variation in root length and two-thirds of the variation for relative root weight. It appears that one should be able to select for roots longer than the short parent with a relative root weight similar to the short parent. In the  $F_2$  length was near the long root parent with relative root weight very near the short root parent; however, the large amounts of dominance variation for both traits indicate that one would need to wait until later generations to make selection (Table 7).

In the cross involving 3768 and ST 213, genetic variance estimates from the generation mean analysis showed mostly residual epistasis involved in inheritance of root length and relative root weight. Thus, this would not be a very good cross to use in selecting for long roots with high relative root weight.

Most of the 124 strains evaluated which had high relative root weight also had roots shorter than the six cultivars used as checks in this study. Strains 3777 and 3900 had roots as long as ST 213 with higher relative root weights. These may be useful as strains to study the effects of high relative root weight on seedling diseases. The breeding implications from the diallel analysis and the generation mean analysis were different. This may be related to different parents being involved in each and additionally the generation mean analysis, which makes use of information from six generations, is probably a superior type of analysis for self-pollinated crops like cotton. In attempting to develop plants with long roots with high relative root weight from the cross of 3822  $\times$  3851, selection should not be too vigorous in the early generations because of the relative amounts of additive, dominance, and epistatic effects. Recurrent selection with selection delayed to the  $F_3$  or  $F_4$  generation should be a useful method of breeding to produce the desired improvements in root length and relative root weight.

We speculate that roots that grow fast yet maintain a high relative root weight when grown in the cool germination test would be useful when planting conditions are cool and seedling diseases are a problem. By having long roots with high relative root weights there should be a mechanical type of resistance to

seedling disease; i.e., the plant should have more root tissue to slough off as diseased tissue and yet maintain a viable root system and a healthy plant than would plants with short roots and low relative root weights. We believe the generation mean analyses indicate that genetic potential is available to develop plants with roots which could be used to test this hypothesis concerning seedling diseases. Additionally, parental strains 3777 and 3900 may be useful directly as they have roots equal to ST 213 in length yet have a higher relative root weight.

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