Analysis of Gene Action and Genetic Variability for Enhanced Seedling Traits in Mulberry (*Morus* spp.)

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Original Research Article

ABSTRACT

Information regarding the nature of gene action controlling the yield and yield contributing characters are essential for implementing good breeding program. With this background, a study was conducted to understand the gene action and variance of lines and testers for seedling traits in mulberry. The percent contribution of testers, lines and their interaction to total variance for all the traits clearly revealed that contribution of lines towards total variance was higher than males for all characters. The variance due to lines (females) was significant for germination percentage, seedling height at 60, 90th DAS, number of branches, intermodal distance, but non- significant for the left-over characters studied. The variance due to Testers (males) was significant for Germination percentage, intermodal distance but non-significant for other characters studied. The line × tester interaction variance was very much significant for germination percentage, seedling height at 90th DAS, average leaf weight (g), Leaf area (cm^2) but non- significant for plant height at 60, 120th DAS,

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number of branches, intermodal distance (cm). Variance due to crosses was exceedingly significant for all characters under study except intermodal distance in the present experiment, an effort was made to know the information on the magnitude of GCA, SCA variance and nature of gene action for the trait as a whole. In this experiment also, non-additive genetic component was the major part of genetic variance for most of the growth parameters.

1. INTRODUCTION

Mulberry is a key harvest for the sericulture business as it is the selective wellspring of fed for silkworm. It is an enduring tree belongs to family Moraceae and originated in the lower inclines of the Himalayas [1]. It can develop under different climatic conditions extending from mild to tropical. In India mulberry develops during the time because of positive climatic conditions, making sericulture a full time occupation. Since mulberry leaf production alone expenses more than 60 percent out of the all cost, the financial return in sericulture is generally dictated by the measure of good quality mulberry leaves delivered from a unit region [2].

For developing high yielding varieties in a crop sound breeding programme is essential. Information regarding the nature of gene action controlling the yield and yield contributing characters are essential for implementing good breeding program. In a good hybrid breeding programme important aspect is combining ability and its analysis is of special importance in cross pollinated crops like mulberry because it helps in identifying good parents which can be used for producing hybrids. Selection of parents in terms of performance of hybrid is provided by combining ability studies.

If mulberry leaf production increased, sericulture productivity increases. Leaf yield in mulberry is a complex trait which is mainly influenced by environment and management practices and donated by a number of component traits Tikader and Kamble [3]. For selection of plants average leaf yield alone may not be the good criterion, so it is vital to understand the heritability and genetics of yield components. Capacity of a parent to pass on its pleasing characters to its progeny in crosses is known as combining ability [4] and developing new varieties with high leaf yield and good adaptability helps to achieve this goal. Parents with good combining ability were known to produce greater progenies when united with other parents. The easiest and efficient method to evaluate combining ability in large number of parents is Line × Tester analysis. Parents with superior combining ability can be selected based on GCA for exploitation of hybrid vigour by development of segregating populations [5].

Keeping in vision, the significance of mulberry regarding its foliage esteem, crop improvement research on this yield should be reinforced. Considering all these view points, the current study was held to unravel the genetics of seedling traits.

2. MATERIALS AND METHODS

Studies on "Gene action and combining ability for seedling traits in mulberry (*Morus* spp.)" were conducted at the Department of Sericulture, University of Agricultural sciences, Gandhi Krishi Vignana Kendra Bengaluru - 65. The experimental plot is situated at an elevation of 931 m above mean sea level and has latitude of 12°58¢ N and longitude of 77°37¢ E. In this chapter the material details, the practices adopted in the conduct and statistical analysis of the experiment followed are explained.

2.1 Selection of Parents

For the current investigation seven lines and three testers are used (Table 1), which were selected from the field germplasm existed at the Department of sericulture, UAS, GKVK, Bangalore-65. With all the recommended package of practices viz., fertilizer application and weeding for rainfed mulberry, these parental materials are maintained [6].

Hybridization programme was done using staggered pruning technique lines and testers flower synchronization is done. Pollens collected were transferred to the catkins in the morning hours between 8:00 to 10:00 AM, and catkins are covered immediately. After that harvesting of crossed fruits and seed extraction was done crosswise. Obtained seeds from fruits were washed several times to remove debris from seeds. Then they were sundried and used for

sowing. Evaluation of twenty-one F1 hybrids derive from seven lines and three testers, were the experimental material which were crossed according to line × tester mating design. All the crosses were assessed using randomized complete block design (RCBD) with three replications and 10 plants per treatment per replications were maintained. Then observations were recorded at 60, 90 and 120 days after sowing for plant height (cm). Observations on average leaf weight (g), number of leaves per plant, single leaf area (cm2) and internodal distance (cm) were recorded at 120 days after sowing.

Table 1. List of lines and testers used in L × T study

Sl. No.	Parents
Female parents (Lines)	
1.	*M. laevigata*
2.	*M. multicaulis*
3.	MI-47
4.	BC- 259
5.	*M. cathyana*
6.	MI-494
7.	*M. indica* E-05
Male parents (Testers)	
8.	V1
9.	MI-66
10.	C-776

2.2 Statistical Analysis

2.2.1 Combining ability analysis

General combining ability (GCA) variance of parents and specific combining ability (SCA) variance of different cross combinations were worked out based on the procedures developed by Kempthorne [7] using means of each replication for the characters recorded for twenty one crosses.

From the mean sum of squares, covariance of full sibs and covariance of half sibs were estimated as follows:

H.S. Co-variance of full sibs=(Ml – Me) +(Mt – Me) +(Mlt - Me) / 3r + 6r Cov H.S. – r (l+t) Cov / 3r

Mlt Co- variance of half sibs = Ml + Mt – 2 / r (l + t)

After estimating COV (HS) and COV(FS) the GCA variance of lines and testers and SCA variance of crosses were estimated as here under

GCA variance for lines = Ml – Mlt / Rt

GCA variance for testers = Mt – Mlt / rl

SCA variance for crosses = Mlt – Me / r

Where,

Ml= MSS due to lines (females)
Mt = MSS due to testers (males)
Mlt = MSS due to lines x testers
Me = MSS due to error

2.2.2 Critical Difference (CD)

The critical difference values in each case were computed by multiplying their corresponding SE values with table 't' value at error degrees of freedom at 5 and 1 percent level of significance.

Proportional contribution of lines, testers and line × tester

Contribution of lines (%) = SS (f) a / SS (c) × 100

Contribution of tester (%) = SS (m) b. / SS (c) × 100

Contribution of lines × tester (%) = SS (f × m) c. / SS (c) × 100

Where

SS (c) = Sum of squares due to crosses
SS (f) = Sum of squares due to lines
SS (m) = Sum of squares due to testers
SS (f × m) = Sum of squares due to lines x tester cross

3. RESULTS AND DISCUSSION

The percentage of contribution of testers, lines and their interaction to total varia nce for the traits were given in Table 3. In this research, it was clear that contribution of lines towards total variance was registered higher than males for all characters *viz.,* germination per cent, plant height (cm) at 60, 90, 120th DAS, number of leaves per plant, average leaf weight (g), Single leaf area (cm2), number of branches per plant and Internodal distance (cm). The contribution of females × males interaction towards the total variance was found to be higher than the testers for germination percentage, plant height (cm) at 60, 90,120th DAS, number of leaves per plant, leaves weight(g), single leaf area (cm2), number of branches. On the other side, the contribution of females × males interaction to the

total variance was higher than the lines for average leaf weight(g) and single leaf area (cm^2). The contribution of lines is higher than testers to total variance for all the characters under study. Bhuvana et al. [8] reported that line x tester interaction for contribution of crosses was found to be high for all the traits except number of leaves per plant and number of branches per plant. Among the traits, interactions contributed more to single leaf area (84.43%) and internodal distance (75.06%). Hence, lines and interactions afforded maximum contribution to the total variance. These results are also in accordance with Banerjee et al. [9] who revealed that female parents contributed higher to the total variance of most of the traits in mulberry.

3.1 Analysis of Variance for Combining Ability

3.1.1 Variance due to lines, testers and lines x tester interaction

Variance due to females, males and female × male interaction with respect to all the characters under study are presented in Table 4. Presence of significant higher variability can be observed from the analysis of variance for all characters under study.

The variance due to lines (females) was significant for germination percentage, seedling height at 60, 90th DAS, number of branches, intermodal distance, but non- significant for the left-over characters studied. The variance due to Testers (males) was significant for Germination percentage, intermodal distance but non-

significant for other characters studied. The line × tester interaction variance was very much significant for germination percentage, seedling height at 90th DAS, average leaf weight (g), Leaf area (cm^2) but non- significant for plant height at 60, 120th DAS, number of branches, intermodal distance (cm). Variance due to crosses was exceedingly significant for all characters under study except intermodal distance.

The results illustrate significant variation among the genotypes for some of the traits studied. The sum of squares of genotypes for these characters was further divided into parents and crosses. The variance due to parents and crosses revealed significant differences among themselves indicating genetic variability for the efficient selection. Significance of variance due to lines, testers and between lines *vs* testers indicated the sufficient difference among the parents. These results are in accordance with those of Vijayan et al.*,* [5] Banerjee et al., [9] and Pooja et al., [10]. Results pertaining to analysis of variance for combining ability were in line with Bhuvana et al. [8] who indicated that the mean squares due to testers were highly significant for number of branches per plant, number of leaves per branch, leaf moisture content and moisture retention capacity whereas significant variances among lines were recorded only for fresh leaf weight.

3.1.2 Variance components and nature of gene action

The variances ($σ^2$ GCA, $σ^2$ SCA) and ratio ($σ^2$ GCA/ $σ^2$ SCA) are given in Table 5.

Table 2. ANOVA for Line x Tester Analysis

Source	Degrees of freedom	Mean sumof squares	Expected mean squares
Replication	r-1		
Lines	l-1	M_l	$σ_e$ +r[COV(FS) – 2COV(HS)] +tr COV(HS)
Testers	t-1	M_t	$σ_e$ +r[COV(FS) – 2COV(HS)] +lr COV(HS)
Lines x Testers	(l-1) (t-1)	M_{lt}	$σ_e$ +r[COV(FS)^2 – 2COV(HS)]
Error	(lt-1)(r-1)	M_e	$σ^2_e$
Total	(ltr-1)		

Where,
r = Number of replications
l = Number of female parents (lines)
t = Number of male parents (testers)
COV (FS) = Covariance of full sibs
COV (HS) = Covariance of half sibs

Table 3. Proportional contribution of testers, lines and their interaction to total variance

Sl. No.	Character		Contribution of lines (%)	Contribution of testers (%)	Contribution of lines × testers (%)
1	Germination (%)		53.60	21.96	24.43
2	Plant height (cm) at	60th DAS	54.17	9.73	36.09
		90th DAS	60.76	2.00	37.23
		120th DAS	56.99	2.38	40.61
3	Number of leaves per plant		57.05	0.52	42.41
4	Average leaf weight (g)		47.73	1.04	51.21
5	Single leaf area (cm^2)		37.77	9.73	52.49
6	Number of branches perplant		76.01	0.73	23.25
7	Internodal distance (cm)		46.66	29.25	24.07

DAS - Days After Sowing

For planning the breeding program information regarding the combining ability studies in terms of GCA and SCA variances is needed. Combining ability studies also helps in knowing the gene action for a given trait. Non-additive gene action can be known from the SCA variance, GCA variance reflects the measure of additive gene action of a particular trait. Ratio of non-additive to additive gene action is to be measured to know the predominance of the type of genetic variation for a given character. If the ratio of non-additive to additive gene action is less than one indicates the major role of additive variance in controlling the expression of a character, whereas, more than one indicates the importance of non-additive variance (Gardner, 1963).

In the present experiment, an effort was made to know the information on the magnitude of GCA, SCA variance and nature of gene action for the trait as a whole. In this experiment also, non-additive genetic component was the major part of genetic variance for most of the growth parameters. Present study is in accordance with Banerjee et al. [9] who reported that non-additive genetic variance is more often evident in controlling the inheritance of majority of yield traits than additive components in mulberry. Suresh et al., [11] reported that predominance of non-additive genetic variance offers scope for exploitation of heterosis in mulberry.

For all character's magnitude of SCA variance was higher than that of GCA variance revealed by analysis of variance except for internodal distance (cm), plant height (cm) at 60 DAS, number of branches per plant. GCA and SCA variance ratio was less than one for all characters, except for plant height(cm) at 60 DAS, number of branches per plant, germination

percentage, internodal distance (cm), where variance was higher than unity.

3.2 Germination Percentage

GCA variance for germination percentage was highly significant with greater importance than the SCA variance which is confirmed by combining ability analysis. Additive gene action is governing this character where ratio of GCA/ SCA variance was more than unity. This suggests that recessive gene controls the trait, and this indicates recessive alleles were more abundant than dominant alleles in parents and indicated the importance of additive gene action in the inheritance of this trait.

3.3 Plant Height (cm) at 60, 90 and 120 DAS

In the study the ratio of GCA/SCA variance was less than one and SCA variance was found to be significant depicting the importance of dominant gene effect for plant height at 90th and 120thDAS whereas at 60th day GCA significant and the ratio of GCA/ SCA variance was more than one indicating the importance of additive gene effect in governing the trait. These results are on par with Pooja et al., 2016 who got ratio of variance less thanunity for plant height.

3.4 Number of Leaves Per Plant

In this experiment sca variance is higher than gca variance for number of leaves. Ratio of GCA to SCA is less than one indicating dominance of non- additive gene action in controlling this trait. Same results were obtained by Evelin Kumari [12] obtained variance of less than one for number of leaves per plant in seedlings.

Table 4. Analysis of variance for combining ability for different growth parameters at seedling stage in mulberry

Source	d.f.	Germinationpercentage	Plant height (cm)			No.of leaves / plant	Leaf weight (g)	Single leaf area(cm^2)	No. ofbranches /plant	Internodaldistance (cm)
			60DAS	90 DAS	120 DAS					
Replication	2	14.71	9.48	30.02	86.30	27.92	10.76	7.50	0.016	0.089
Crosses	20	418.50**	20.82*	198.44**	419.90**	199.54**	89.89**	819.52**	6.71**	0.64
Lines	6	747.77*	37.60*	401.93*	797.814	379.53	143.04	1031.86	17.01**	0.99*
Testers	2	919.19*	20.26	39.82	100.24	10.49	9.41	797.41	0.49	1.87**
Line × Tester	12	170.41*	12.52	123.13**	284.22	141.06	76.73**	717.03**	2.60	0.25
Error	140	82.214	9.66	35.62	143.14	75.93	4.62	31.12	1.74	0.44
Total	62	188.51	13.25	87.96	230.58	114.26	32.33	284.68	3.29	0.49

* Significant at 5 %, ** Significant at 1 %, DAS - Days After Sowing

Table 5. Estimates of variance components in respect of different growth parameters at seedling stage in mulberry

Sl. No.	Characters		σ^2 GCA	σ^2 SCA	σ^2GCA/σ^2 SCA
1	Germination %		51.62**	37.11**	1.39
2	Plant height	60 DAS	1.236**	0.712	1.73
	(cm) at	90 DAS	12.91*	32.01**	0.41
		120 DAS	23.33*	61.76**	0.378
3	Number of leaves per plant		9.55	29.78**	0.32
4	Average leaf weight (g)		4.71	23.71**	0.19
5	Single leaf area(cm^2)		58.93	228.8**	0.25
6	Number of branches per plant		0.48**	0.39	1.25
7	Intermodal distance (cm)		0.07**	-0.03	-2.18

3.5 Average Leaf Weight (g)

Analysis of data for combining ability revealed the importance of *sca* variance. The ratio of GCA / SCA is less than one which revealed the importance of non-additive gene action. The results are in accordance with Suresh et al. [11]. In their investigation also non additive component prevails for leaf weight in mulberry.

3.6 Leaf Area (cm^2)

Analysis of data for combining ability revealed the significance of SCA variance. The ratio of GCA / SCA was less than one which revealed predominance of non- additive gene action for fresh leaf weight. This was in accordance with the findings of Suresh et al. [11] in mulberry.

3.7 Number of Branches Per Plant

Analysis of data for combining ability revealed the significance of SCA variance for number of branches. Ratio of GCA / SCA variance recorded was less than unity. This suggested the prevalence of non-additive gene action for the trait. This result was on par with Vijayan et al. [5] non-additive component was important for this trait in mulberry. Pooja et al. [10] also reported non additive gene action for number of branches.

3.8 Internodal Distance (cm)

Combining ability studies revealed significant GCA variance and non-significant SCA variance and the ratio of GCA to SCA is less than one depicting the importance of non-additive gene action for the trait. At the same time variances in line x tester interaction were observed to be highly significant for most of the parameters *viz.,* germination per cent, plant height and single leaf area which revealed that all the mulberry genotypes showed variations with respect to the traits which was reported by Bhuvana et al. [8]. Hence, there is a significant difference among the lines, testers and their interactions.

Similar result was obtained in studies undertaken by Vijayan et al. [5] which confirmed predominant role of SCA variance for this trait and GCA / SCA ratio was less than one for this trait which indicates preponderance of non-additive over additive gene action. Similar result was recorded by Ramachandra et al. [13] in tobacco.

4. CONCLUSION

The percentage of contribution of testers, lines and their interaction to total variance for the traits, it was clear that contribution of lines towards total variance was registered higher than males for all characters. The variance due to lines (females) was significant for germination percentage, seedling height at 60, 90th DAS, number of branches, intermodal distance, but non- significant for the left-over characters studied. The variance due to Testers (males) was significant for Germination percentage, intermodal distance but non-significant for other characters studied. The line × tester interaction variance was very much significant for germination percentage, seedling height at 90th DAS, average leaf weight (g), Leaf area (cm^2) but non- significant for plant height at 60, 120th DAS, number of branches, intermodal distance (cm). Variance due to crosses was exceedingly significant for all characters under study except intermodal distance in the present experiment, an effort was made to know the information on the magnitude of GCA, SCA variance and nature of gene action for the trait as a whole. In this experiment also, non-additive genetic component was the major part of genetic variance for most of the growth parameters.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of this manuscript.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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