Biological Forum – An International Journal 16(8): 215-224(2024)

ISSN No. (Print): 0975-1130
ISSN No. (Online): 2249-3239

Mean Performance and Hybrid Vigour in Mulberry for different Growth and Yield Traits (*Morus* spp.)

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(Received: 03 June 2024; Revised: 24 June 2024; Accepted: 18 July 2024; Published: 14 August 2024)
(Published by Research Trend)

ABSTRACT: To develop a good variety/hybrid through breeding, evaluation of existing genetic variability by crossing with the specific genotypes is required. By understanding the GCA and SCA in the earlier studies, the valuation of existing hybrids of mulberry by application of biometrical methods *viz*., heterosis is essential to compare performance of new hybrids with the commercial and conventional varieties. So also, production of new strains by hybridization. Heterosis analysis revealed that, among hybrids, for mid parent and better parent heterosis most of the selected hybrids recorded significantly positive heterosis for growth and yield characters, indicating hybrid vigour in clones over the parents. Hybrid ME-03 × MI-66 recorded maximum and positive standard heterosis for number of leaves (41.5 and 16.95 %), fresh weight of leaves (130g/30 leaves and 8.15 %), moisture content (74.6 % and 6.42 %), leaf shoot ratio (1.23 and 3.61 %) and specific leaf area (322.93 cm^2/g and 27.12 %). Whereas, hybrid ME-65 × V1 recorded no significant difference over mid, better parent and standard heterosis for leaf yield (g), single leaf area (cm^2), moisture content (%) number of leaves per branch, number of branches per plant and other traits and equally performing as compared to the check variety V1.

Keywords: Heterosis, Mulberry, Hybrids, Silkworm.

INTRODUCTION

Mulberry is the primary food plant of silkworm *Bombyx mori* L. belongs to family Moraceae and its being exploited for commercial purpose**.** Mulberry is the primary food plant of silkworm *Bombyx mori* L. belongs to family Moraceae and its being exploited for commercial purpose. Mulberry leaf productivity is directly supporting the volume of silkworm rearing. Mulberry breeders have been working hard to improve leaf productivity and quality beneficial to increase cocoon productivity per unit area.

Hybrid vigour, the manifest effect of heterosis has been one of the most interesting topics in the science of genetics and plant breeding. Study on hybrid vigour in the beginning were mainly of academic interest. Various hypotheses have been advanced to explain the phenomenon. Theory of heterozygosity (Shull, 1908, Shull, 1911; East, 1908), theory of dominance (Davenport, 1908; Bruce, 1910; Keeble and Pellow 1910) theory of intra-allelic interaction (East, 1936) the super dominance or over dominance hypotheses (Hull, 1945) and physiological hypotheses (Ashby, 1930, Ashby, 1932; Ashby, 1937) are some among the several hypotheses put forth. Consensus is that one or several of these phenomena act alone or in combination in any given situation of heterotic effect.

Heterosis in mulberry has been studied for leaf yield and its component characters. Vijayan *et al*. (1998) reported that heterosis breeding in mulberry is quite possible, though the plant is highly heterozygous. Exploiting heterosis mainly relies on general and specific effects of combining abilities. Studies on combining ability are helpful in evaluating the parental lines and their cross-combination for the estimation of heterosis. To take advantage of commercially viable heterosis, this information helps in the selection of parents based on the performance of the hybrids and clarifies the nature and extent of various types of gene action involved in the expression of morpho-physiological traits.

In mulberry, cross-pollination is the norm rather than the exception. The generation itself of the population and perennial nature are the limitations for the improvement of this crop because of the heterozygous nature of the plant, which causes the segregation of characters in F_1. Open pollinated populations have quite a bit of variety, which allows for significant improvement opportunities while utilising traditional breeding techniques. A sound and effective breeding programme requires genetic variety, which cannot be assessed through biometrical methods, which is a requirement for this crop. The goal of mulberry breeding and selection is primarily focused on the foliage.

Heterosis or hybrid vigour are terms used to describe the increased vigour. A hybrid that is developed by the cross of two dissimilar parents is heterozygous and typically active. The extent to which hybrid vigour is exploited depends on the nature and strength of the gene interactions involved and it is regarded as one of the outstanding achievements of plant breeding. Breeders should practise advanced and modified breeding strategies, to maximise the gain/unit of time (Sarkar *et al.,* 1999). To achieve this, it is necessary to manage the improved population from the beginning and to keep track of the progress made with each succeeding generation. In addition, enough genetic diversity must be preserved to support ongoing long-term breeding programmes (Sarkar *et al.,* 1999).

In order to identify the crosses with a high heterotic potential, the new crosses are contrasted with previously evolved varieties and hybrids. The isolated crosses were assessed for growth and yield traits, of mulberry and silkworm rearing performances. In mulberry, it is established that total shoot length, shoots/plant, longest shoot length, single leaf weight and leaf area have direct bearing on leaf yield potential (Rahman *et al*., 2001; Tikadar and Rao 2002; Puttarama *et al*., 2004; Rahman *et al*., 2004; Tikader *et al*., 2004; Susheelamma *et al*., 2005).

Vijayan *et al*. (1998) reported that heterosis breeding in mulberry and observed that a hybrid Berhampore-1× Kajli is highly promising for future utilization. Large-scale, long-range breeding programmes are required to achieve breeding goals and a sufficiently conserved, broad spectrum genetic variability is a vital prerequisite.Variations in the nutritional composition of mulberry leaves have been reported across different genotypes and environmental conditions (Kumar *et al*., 2016).

Ravi (1991) studied heterosis and stability analysis in mulberry (*Morus* spp.). Significant positive standard heterosis was observed in hybrids Mizusawa/Cattaneo, Sujanpur-1/Philippine Si, Sujanpur-1/Philippine S2, Local/Kokuso 13 Si, over both check varieties for the number of branches per plant. Local/Kokuso 13 S2 outperformed the M5 control and three hybrids outperformed the S-54 control in terms of leaf number per plant. Over both check varieties, the cross Local/Kokuso 13 S2 demonstrated significant positive standard heterosis for leaf to stem ratio by fresh weight. In addition, two hybrids over M5 and three hybrids over S-54 showed significant positive standard heterosis for total leaf dry matter yield per plant.

The study analyzed the genetic variation and comprehensively evaluation of four morphological indexes such as shoot height (SHR), leaf number (LNR), leaf area (LAR), and the total weight of the whole plant after defoliation (BI) in the seedlings of the 14 mulberry hybrids against salt tolerance. Hybrid Anshen × Xinghainei (female: Anshen, male: Xinghainei) found to be best hybrid combination under high salinity stress also perform best specific combining ability for BI (Chen *et al*., 2023). Among the traits, LAR and BI were notably influenced by additive effects from the hybrid combination, suggesting their potential to enhance mulberry resources by breeding and selecting elite germplasms with high salt tolerance.

Bhuvana *et al*., 2020 evaluated the growth and survivability traits in F_1 hybrids for General combining ability (GCA) and Specific combining ability (SCA) plant height, number of leaves per branch, internodal distance and leaf moisture content. Among the fifteen F_1 crosses, MI-0685 × V1 Hybrid was found to perform better than the remaining crosses and MI-0543 × V1 was found to be the best combiners. Hence, these parents and F_1 progenies may be further used in breeding programs for mulberry crop improvement.

Ghosh *et al.* (2008) studied heterosis in mulberry for leaf yield and its attributing characters in hybrid progenies. For leaf yield trait Hybrid No. 73 showed a higher leaf yield exceeding 236.30 per cent compared to the ruling check (100.36 - 236.30 %). For total shoot length, five hybrids showed higher standard heterosis (25.46 to 63.28 %). The improvement observed in hybrid No. 73 across all seven traits aligns with Singh's (2000) view, which attributes such enhancement to the geographical and genetic divergence of the advanced generation improved hybrid parents.

Ghosh *et al*. (2009) studied heterosis for leaf yield and its attributing characters in eight hybrids developed. Among the hybrids, C-2038 recorded a higher number of shoots per plant (10.93), total shoot length (1059.95 cm), unit leaf area (284.06 cm^2), unit leaf weight (4.87 g), leaf yield (55.23 MT/ha/year) and better economic parameters, showing its superiority over check and other test hybrids. Significant positive heterosis ranging from 5.95 to 159.31 per cent over better parent, 13.92 to 159.31 per cent over the mid parent and 1.50 to 27.79 per cent over the standard variety (S-1635) was observed among the crosses for leaf yield.

Doss *et al*. (2011) screened 210 hybrids, among the hybrids CT-44 and CT-11 outperformed the check S1635 by 17.17 and 7.11 per cent respectively. The hybrids CT-44 and CT-11 are yielding only 17.17 and 7.11 per cent higher than the present popular variety S-1635, their yield through the colder months, 7.93 and 8.11 mt per ha respectively, were higher than that of the variety S-1635. Thus, indicated that systematic and concerted efforts can result in achieving specific goals like developing varieties for specific purposes even in tree crops like mulberry.

Andadari *et al*. (2016) tested five mulberry hybrids in Indonesia. Two types of hybrid mulberry, *Morus cathayana* × *Morus amakusaguwa* IV.12 and *Morus cathayana* × *Morus amakusaguwa* IV.10 increased 59.60 and 47.83 per cent silk production and found excellent potential in silkworm feed productivity development.

The ratio of gca and sca variance was less than unity indicating preponderance of non-additive genetic variance and suggests the good prospects of exploiting variation through heterosis breeding. Bishnupur-10, Kajli OPH, C-776 and CF1 -10 were identified as good general combiners and can be utilized for generating desirable recombinants for further selection. Out of 42 crosses, nineteen exhibited significant positive sca effects for yield and could be used for selection of superior hybrid progenies. Nineteen genotypes recorded significantly higher leaf yield (482-501 g/plant/crop) along with better quality over the check variety S1635 (324 g/plant) (Suresh *et al*., 2019).

Evelin Kumari (2018) carried out line× tester analysis to determine combining ability for growth and yield parameters in mulberry. Out of ten crosses, MI-516 × MI-04 (number of leaves per plant, leaf to shoot ratio, leaf yield per plant), MI-139 × C-776 and MI-47 × MI-04 (number of leaves per plant) were good performers with high sca effects.

Bekkamov (2023), found increased weight and volume of the silk gland when fed with the mulberry hybrids that is ranging from 1550-1650 mg and 1.55-1.63 cm^3, respectively.

Improving silk production entails improving the yielding ability of the silkworm *Bombyx mori* L. and host plant. The primary concern of a plant breeder is the genetic improvement of qualitative and quantitative characters of the mulberry plant. The development of high yielding hybrids is only one aspect of mulberry breeders. But ultimately, their efforts will be judged for the qualitative and quantitative advancements seen in silkworm rearing. Although it is thought that leaf nutrients vary from genotype to genotype, it has not yet been proven in all instances that the variation is accurately reflected in cocoon parameters.

The study aims in evaluation of newly developed mulberry hybrids for genetic variation and understanding how different growth and yield traits interact in hybrids can optimize breeding strategies to enhance overall plant performance. This study also explores the morphological basis of heterosis in mulberry to identify the specific traits responsible for enhanced growth and yield.

Only the quality and quantity of mulberry leaves can determine the profitable of sericulture. To compete in the global market, there has been a lot of focus on producing raw silk of superior quality. In order to accomplish this, efforts are being made to boost the production of high-quality mulberry leaves, which have a direct impact on the calibre and output of raw silk.

MATERIAL AND METHODS

The hybrids were developed by Line × Tester mating design during 2019. Saplings were planted in the main field after 90th day for the growth and vigour studies. These hybrids along with the parents were planted in three rows with four plants in each row, with a spacing of 3 ft ×3 ft in the field in RCBD design with three replications at the Department of Sericulture, UAS, GKVK, Bengaluru. These hybrids were maintained with as per the package of practices recommended for rainfed mulberry (Dandin and Giridhar 2014**.** These hybrids were maintained for six months and pruned for the first crop. After pruning hybrids were evaluated on 30, 45and 60th day of pruning for growth and yield parameters.

Table 1: List of mulberry hybrids and their parents used in the study.

Sr. No.	Hybrids	Parents
1.	MI-47 × MI-66	MI-47 (*Morus indica*)
2.	MI-79 × MI-66	MI-79 (*Morus laevigata)*
3.	ME-03 × MI-66	ME-03 (*Morus cathyana)*
4.	ME-146 × MI-66	ME-146 (*Morus indica*)
5.	ME-65 × V1	ME-65 (*Morus alba)*
6.	ME-67 × V1	ME-67 (*Morus alba)*
7.	V1 (Standard check)	MI-66 (*Morus indica*)

Growth parameters. For all the growth and yield parameters five random competitive plants from hybrids were selected and observations were recorded

Shoot height (cm). The height of the main shoot from base of the pruning to tip of the plant was measured and expressed in centimetres. Shoot height of all branches of each hybrid plant was recorded and then average shoot height (cm) was calculated.

Single leaf area (cm^2). Single leaf area was measured by taking an individual leaf from different hybrids and placed in leaf area meter and observations are recorded and expressed in square centimetres.

Number of branches per plant. All the main branches of mulberry hybrid plant were counted and recorded at 30, 45 and 60th days of pruning.

Number of leaves per branch. Total number of leaves was counted in each plant before each harvest and average number of leaves per plant was recorded at 30, 45 and 60th day after pruning.

Internodal distance (cm). Distance between two successive nodes of shoot was measured in scale of the main shoot and expressed in centimetres and average internodal distance was calculated.

Leaf moisture content (%). Fresh leaf weight and dry leaf weight of ten composite sample leaves was taken and moisture content of leaf was calculated by using the following formula and expressed in percentages.

Leaf yield per plant (g). Leaves harvested from different replicated plants and weight of the leaves were recorded. The average leaf yield per plant was estimated and expressed in grams.

HETEROSIS

Estimation of heterosis. The overall mean for each parent or hybrid from the three replications for each character was considered for the estimation of heterosis. The magnitude of heterosis over mid parent (MP), better parent (BP), standard check (SC) was calculated using TNAUSTAT software. The percentage increase or decrease in the mean of the F1 over their respective mid parent, better parent and standard check mean value was calculated by using the following formulae.

1. Heterosis over mid parent (Relative heterosis) = $\frac{\overline{F1} - \overline{MP}}{\overline{MP}} \times 100$

Where, Mid parent ($\overline{MP}$) = $\frac{P1 + P2}{2}$

2. Heterosis over better parent (Heterobeltiosis) = $\frac{\overline{F1} - \overline{BP}}{\overline{BP}} \times 100$

3. Heterosis over standard check (Standard heterosis) = $\frac{\overline{F1} - \overline{SC}}{\overline{SC}} \times 100$

Where, $\overline{F1}$ =Meanperformanceof the F1
P1= Mean performance of the parent one
P2 = Mean performance of the parent two
$\overline{BP}$ = Mean performance of the better parent
$\overline{SC}$ = Mean performance of the standard check
For better parent value (BP) for each character, superior value between the parents in each cross was taken.

Significance of estimates of heterosis: The estimates of relative heterosis, heterobeltiosis and standard heterosis effects were tested for their statistical significance as follows:

SE (MP) = $\sqrt{\frac{3M4}{2r}}$

SE(BP) = $\sqrt{\frac{2M4}{r}}$

SE(SC) = $\sqrt{\frac{2M4}{r}}$

Where,
M4=Error mean sum of squares in ANOVA table of Line xtester analysis
R = number of replications

Overall status of the parents and crosses with respect to heterosis. Since yield and its component characters are correlated either positively or negatively, it is common to find heterosis for a particular parent and hybrid respectively, in the desirable direction for some characters and in the undesirable direction for some characters. The problem of ascertaining the status of a parent and hybrid with respect to heterosis, respectively was determined by following the method suggested by Arunachalam and Bandyopadhyay (1979), which is slightly modified by Mohan Rao (2000).

Statistical analysis. The recorded data was uploaded to the software *viz*., TNAUSTAT for estimating the heterosis among the mulberry hybrids. This package was developed by Manivannan (2014). Results showed variance among the parents and progenies and specific combining ability of the F1 progenies (SCA) and the heterosis over mid parent, better parent and standard check.

Estimation of heterosis over mid and better parent and standard check for growth and yield attributing traits. The mean values of all six mulberry hybrids were compared to those of the midparent, better parent, and standard check variety Victory-1 for each of quantitative traits and expressed as percentages of relative heterosis (mid parent heterosis), heterobeltiosis (better parent heterosis), and standard heterosis (heterosis over check) respectively.

RESULTS AND DISCUSSIONS

Shoot height (cm). In the evaluation of shoot height at 45 days after pruning, the hybrid MI-79 × MI-66 recorded the highest shoot height (115.3 cm), followed by ME-146 × MI-66 (110.9 cm), while the lowest height was observed in ME-67 × V1 (67.7 cm) (Table 3). Heterosis over the standard check V1 ranged from -31.01% to 17.50%, with MI-79 × MI-66 (17.50%) and ME-146 × MI-66 (13.00%) showing significant positive heterosis. Hybrids ME-65 × V1 and ME-03 × MI-66 exhibited non-significant differences compared to the check.

At 60 days after pruning, MI-79 × MI-66 again achieved the maximum shoot height (144.4 cm), followed by ME-146 × MI-66 (137.2 cm), while ME-67 × V1 recorded the lowest (99.6 cm). The standard check V1 measured 132.8 cm in shoot height. Most hybrids demonstrated significant positive heterosis over the better parent, particularly those involving MI-66 as a parent. Better-parent heterosis ranged from -24.99% in ME-67 × V1 to 27.97% in MI-79 × MI-66. Four hybrids displayed significant heterosis over the standard check, with MI-79 × MI-66 showing the highest positive heterosis (8.77%) at 60 days.

The results are in conformity with the earlier study by Ravi (1991) who recorded negative heterosis for plant height over the check. Since plant height is a vegetative character that is not always related to yield, care should be exerted when selecting parents for a heterosis breeding programme. Increased height due to hybrid vigour, on the other hand, can definitely increase the harvest index and, ultimately, the yield. The standard heterosis for plant height ranged from -10.10 to 51.26 per cent and maximum positive heterosis was noticed in hybrid M. cathyana × V1 (51.26 %).

Table 2: Estimates of heterosis over mid parent, better parent and standard check for shoot height at 45 and 60th day in mulberry hybrids.

Sr. No.	Hybrids	Shoot height at 45th day (cm)			Shoot height at 60th day (cm)		
		MPH (%)	BPH (%)	SH (%)	MPH (%)	BPH (%)	SH (%)
1.	MI-47 × MI-66	2.64	-7.82	-20.40**	19.69**	18.23**	-13.85**
2.	MI-79 × MI-66	33.17**	30.34**	17.50**	39.37**	27.97**	8.77*
3.	ME-03 × MI-66	14.53**	14.03**	-1.57	24.51**	15.02**	-3.53
4.	ME-146 × MI-66	28.36**	25.90**	13.00*	36.02**	27.80**	3.36
5.	ME-65 × V1	5.65	-1.60	-1.60	2.68	-8.62*	-8.62*
6.	ME-67 × V1	-22.07**	-31.01**	-31.01**	-13.92**	-24.99**	-24.99**

MPH- Mid Parent Heterosis, BPH-Better Parent Heterosis, SH-Standard Heterosis, *Significance at 5% level, **Significance at 1% level

Table 3: Mean performance of hybrids, parents and standard check for yield attributing characters in mulberry.

Sr. No.	Hybrids	Shoot height (cm)		Number of leaves/branch			Number of branches/plant			Single leaf area (cm^2)	
		At 45th day	At 60th day	At 30th day	At 45th day	At 60th day	At 30th day	At 45th day	At 60th day	At 45th day	At 60th day
1.	**MI-47 × MI-66**	78.1	114.4	24.1	32.5	39.2	5.0	6.8	8.1	166.6	189.3
2.	**MI-79 × MI-66**	115.3	144.4	24.1	29.9	36.6	5.1	7.2	8.6	199.7	226.9
3.	**ME-03 × MI-66**	96.6	128.1	25.4	34.7	41.5	4.3	6.0	7.1	231.0	262.5
4.	**ME-146 × MI-66**	110.9	137.2	24.9	27.0	38.7	3.7	5.6	6.6	202.0	229.4
5.	**ME-65 × V1**	96.6	121.3	23.8	24.8	33.8	4.6	6.5	8.2	209.8	238.3
6.	**ME-67 × V1**	67.7	99.6	18.5	19.2	31.0	3.3	4.5	5.4	246.3	274.3
7.	**V1 (Check)**	98.1	132.8	24.5	29.2	35.5	5.9	8.0	9.6	217.2	246.7
	Parents										
1.	**MI-47**	86.9	109.1	21.1	29.3	32.0	4.1	5.7	7.4	155.3	176.4
2.	**MI-66**	109.1	106.4	20.3	29.0	31.3	4.5	6.3	8.1	150.9	171.4
3.	**MI-79**	114.0	127.3	22.9	28.2	32.4	4.7	6.5	8.5	173.4	197.0
4.	**ME-03**	108.2	125.6	21.3	31.9	35.6	4.7	6.6	8.6	164.0	186.3
5.	**ME-65**	109.1	116.8	20.1	21.4	27.8	4.1	5.8	6.5	185.9	211.2
6.	**ME-67**	97.4	111.2	16.8	20.4	24.4	4.0	5.6	7.3	195.3	221.8
7.	**ME-146**	113.5	121.1	22.4	32.6	34.4	4.5	6.4	8.3	181.1	205.7

Number of leaves/branch. Hybrid ME-03 × MI-66 (25.4) recorded maximum number of leaves followed by ME-146 × MI-66 (24.9) at 30th day of pruning. Whereas other four hybrids recorded non-significant difference (Table 3) and Standard check V1 recorded the average of 24.5 leaves. Standard heterosis was non-significant for five hybrids, implying that they produced an equal number of leaves at 30th day of pruning as the standard variety V1, while ME-67 × V1 noticed negative standard heterosis of 24.46 per cent (Table 4).

Table 4: Estimates of heterosis over mid parent, better parent and standard check for number of leaves at 30, 45 and 60th day in mulberry hybrids.

Sr. No.	Hybrids	Number of leaves/branch at 30th day			Number of leaves/branch at 45th day			Number of leaves/branch at 60th day		
		MPH (%)	BPH (%)	SH (%)	MPH (%)	BPH (%)	SH (%)	MPH (%)	BPH (%)	SH (%)
1.	MI-47 × MI-66	16.49**	14.20	-1.63	11.30**	11.94**	11.20**	18.68**	17.29**	10.44**
2.	MI-79 × MI-66	11.49*	5.09	-1.77	4.49	2.87	2.19	9.97**	8.04*	2.97
3.	ME-03 × MI-66	22.28**	19.41**	3.67	13.96**	19.52**	18.73**	18.95**	11.71**	16.95**
4.	ME-146 × MI-66	16.47**	11.01*	1.36	-12.32**	-6.89	-7.50*	12.85**	7.77*	8.91**
5.	ME-65 × V1	6.58	-3.12	-3.12	-1.91	-15.05**	-8.19*	4.74	-4.79	-4.79
6.	ME-67 × V1	-10.25*	-24.46**	-24.46**	-22.68**	-34.32**	-34.31**	1.84	-12.55**	-12.55**

Hybrid ME-03 × MI-66 had expressed highest percentage of standard heterosis (18.73 %) followed by MI-47 × MI-66 (11.20 %) and other hybrids had negative standard heterosis. Whereas hybridMI-79 × MI-66 doesn't show any significant difference over standard check V1 for number of leaves (Table 5).

Significant higher number of leaves at 60th day of pruning was recorded in hybrid ME-03 × MI-66 (41.5) followed by MI-47 × MI-66 (39.2) where as standard check V1 recorded 35.5 leaves. Lowest number of leaves was registered in hybrid ME-67 × V1 (31.0) (Table 7). Standard heterosis was most prevalent in MI-47 × MI-66, ME-03 × MI-66, and ME-146 × MI-66, with percentages of 10.44, 16.95, and 8.91 respectively. There was no discernible difference between hybrid ME-65 × V1 and a conventional check V1for number of leaves at 60th day of pruning.

The findings agree with earlier study of Ravi (1991) who reported that six hybrids recorded positive standard heterosis over the control variety, indicated that hybrids have a higher leaf production potential than commercial varieties. The current findings were also conformity with the findings of previous research by Ghosh *et al*. (2008).

Number of branches per plant. Table 4 indicates number of branches at 30, 45 and 60 days after pruning (DAP), the standard check variety V1 had the highest number of branches (5.9), followed by hybrid MI-79 × MI-66 (5.1), while hybrid ME-67 × V1 had the lowest (3.3). Hybrids MI-47 × MI-66 and ME-03 × MI-66 showed positive mid-parent heterosis of 28.32% and 22.84%, respectively, while others had non-significant heterosis (Table 4). However, hybrids ME-03 × MI-66, ME-146 × MI-66, and ME-67 × V1 exhibited negative standard heterosis, ranging from -17.77% to -38.01%.

At 60 DAP, V1 maintained the highest branch count (9.6), with MI-79 × MI-66 leading among hybrids (8.1). Hybrid ME-67 × V1 showed significant negative mid-parent heterosis (-30.45%) and standard heterosis (-39.04%), while other hybrids exhibited non-significant heterosis (Table 5).

Table 5: Estimates of heterosis over mid parent, better parent and standard check for number of branches at 30, 45 and 60th day in mulberry hybrids.

Sr. No.	Hybrids	Number of branches/plant at 30th day			Number of branches/plant at 45th day			Number of branches/plant at 60th day		
		MPH (%)	BPH (%)	SH (%)	MPH (%)	BPH (%)	SH (%)	MPH (%)	BPH (%)	SH (%)
1.	MI-47 × MI-66	28.32**	23.15**	-5.12	26.19**	20.54*	-7.37	15.01	9.86	-7.61
2.	MI-79 × MI-66	22.84**	20.19**	-3.23	23.64**	21.01*	-2.88	13.17	10.73	-2.69
3.	ME-03 × MI-66	4.04	1.48	-17.77*	2.08	-0.44	-19.51**	-6.10	-8.43	-18.97*
4.	ME-146 × MI-66	-9.54	-10.19	-29.79**	-1.83	-2.54	-24.01**	-11.97	-12.64	-25.41**
5.	ME-65 × V1	1.48	-13.35	-13.35	3.35	-11.87	-11.87	10.88	-7.23	-7.23
6.	ME-67 × V1	-26.65**	-38.01**	-38.01**	-28.10	-39.30**	-39.37**	-30.45**	-39.04**	-39.04**

MPH- Mid Parent Heterosis, BPH-Better Parent Heterosis, SH-Standard Heterosis, *Significance at 5% level, **Significance at 1% level

Hybrid MI-47 × MI-66 and MI-79 × MI-66 hybrids expressed positive heterosis over the mid parent and the better parent, but no heterosis over standard check. MI-47 × MI-66, MI-79 × MI-66, and ME-65 × V1 recorded same number of branches at 60th day of pruning with a non-significant difference in the standard heterosis, indicating that the quantitative traits were equally expressed. Vijayan *et al.* (1998) found positive midparent and better parent heterosis for the number of primary branches in mulberry, which might be due to heterosis induced by dominant genes and their interactions or complementary gene interactions, as Gravois, (1993) noticed in rice.

Single leaf area (cm^2). Single leaf area in hybrids ranged from 166.6 to 246.3 cm², with the highest recorded in ME-67 × V1 (246.3 cm²), followed by ME-03 × MI-66 (231.0 cm²). The standard check V1 registered a leaf area of 217.2 cm². All hybrids exhibited positive mid-parent heterosis, ranging from 10.11% to 61.45%, and five hybrids showed significant positive heterosis over the better parent, with ME-03 × MI-66 and ME-67 × V1 displaying higher standard heterosis (8.96% and 16.16%, respectively). Whereas at 60th day ME-67 × V1 (274.3 cm²) and ME-03 × MI-66 (262.5 cm²), surpassing the standard check V1 (246.7 cm²). Positive mid-parent heterosis was consistent across hybrids, with ME-03 × MI-66 showing the highest better-parent heterosis (53.11%). Standard heterosis was notably higher in ME-67 × V1 (15.64%) and ME-146 × MI-66 (10.66%) (Table 3 & 6).

These results indicate that hybrids ME-67 × V1 and ME-03 × MI-66 demonstrated superior leaf area traits, suggesting their potential in breeding programs aimed at enhancing leaf yield in mulberry. This supports earlier findings by Vijayashekara (2009), which highlighted significant variability in single leaf area among mulberry hybrids.

Internodal distance (cm). Internodal distance is an important characteristic because it determines leaf yield. The shorter the internodal distance, the more leaves per branch, and thus higher leaf yield. In evaluating internodal distance among six mulberry hybrids, the minimum distance at 45 days after pruning (DAP) was recorded in hybrid ME-65 × V1 (4.2 cm), followed by ME-03 × MI-66 (4.8 cm), while ME-67 × V1 registered the maximum distance (5.8 cm). Hybrids exhibited significant negative mid-parent heterosis, ranging from -10.25% to -21.96%, with ME-65 × V1 showing the most substantial reduction (-10.25%). Significant better-parent heterosis was observed in MI-79 × MI-66 (15.84%) and ME-65 × V1 (-22.09%). Most hybrids displayed negative standard heterosis, with ME-65 × V1 recording the lowest (-22.13%) (Table 3 & 7).

Table 6: Estimates of heterosis over mid parent, better parent and standard check for single leaf area at 45 and 60th day in mulberry hybrids.

Sr. No.	Hybrids	Single leaf area at 45th day (cm^2)			Single leaf area at 60th day (cm^2)		
		MPH(%)	BPH (%)	SH (%)	MPH (%)	BPH (%)	SH (%)
1.	MI-47 × MI-66	19.74**	18.04**	-21.42**	16.78**	18.75**	-20.19**
2.	MI-79 × MI-66	35.49**	26.70**	-5.82	32.43**	24.72**	-4.35
3.	ME-03 × MI-66	61.45**	55.01**	8.96*	57.44**	53.11**	10.66**
4.	ME-146 × MI-66	33.86**	22.71**	-4.75	30.96**	21.75**	-3.26
5.	ME-65 × V1	10.11**	-1.06	-1.06	11.06**	0.49	0.49
6.	ME-67 × V1	26.46**	16.16**	16.16**	25.00**	15.64**	15.64**

Table 7: Estimates of heterosis over mid parent, better parent and standard check for internodal distance at 45 and 60th day in mulberry hybrids.

Sr. No.	Hybrids	Internodal distance (cm) At 45th day			Internodal distance (cm) at 60th day		
		MPH (%)	BPH (%)	SH (%)	MPH (%)	BPH (%)	SH (%)
1.	MI-47 × MI-66	3.03	2.69	-13.50**	3.29	2.60	-15.85**
2.	MI-79 × MI-66	21.96**	15.84**	-3.07	16.97**	13.01**	-7.32**
3.	ME-03 × MI-66	3.49	-1.03	-17.18**	3.84	-0.37	-18.29**
4.	ME-146 × MI-66	5.57	5.57	-11.66**	8.90**	7.79*	-9.76**
5.	ME-65 × V1	-10.25**	-22.09**	-22.13**	-2.82	-15.85**	-15.85**
6.	ME-67 × V1	13.80**	6.75	6.70*	16.04**	8.85**	8.54**

MPH- Mid Parent Heterosis, BPH-Better Parent Heterosis, SH-Standard Heterosis, *Significance at 5% level, **Significance at 1% level

At 60 DAP, ME-03 × MI-66 showed the minimum internodal distance (4.8 cm), and ME-67 × V1 had the maximum (5.9 cm). Mid-parent heterosis varied from -2.82% to 16.97%, with MI-79 × MI-66 exhibiting the highest positive heterosis (16.97%). Negative better-parent heterosis was significant in ME-65 × V1 (-15.85%), while other hybrids, including MI-79 × MI-66 and ME-67 × V1, showed positive heterosis. Standard heterosis was largely negative, with ME-03 × MI-66 showing the most considerable decrease (-18.29%).

These results align with previous studies by Bari *et al.* (1989); Sahu *et al.* (1995); Ghosh *et al.* (2009), which suggest that negative heterosis for internodal distance is desirable as it enhances leaf yield by increasing the number of leaves per unit stem length.

Table 8: Mean performance of hybrids, parents and standard check for yield attributing characters in mulberry.

Sr. No.	Hybrids	Internodal distance (cm)		Moisture content (%)	Leaf yield/plant (g)
		At 45th day	At 60th day		
1.	**MI-47 × MI-66**	4.7	4.6	70.6	939.58
2.	**MI-79 × MI-66**	5.3	5.1	71.1	1182.39
3.	**ME-03 × MI-66**	4.5	4.5	74.6	1254.38
4.	**ME-146 × MI-66**	4.8	4.9	74.8	835.37
5.	**ME-65 × V1**	4.2	4.6	69.3	1080.91
6.	**ME-67 × V1**	5.8	5.9	72.1	537.61
7.	**V1 (Check)**	5.4	5.5	70.7	1298.50
	Parents				
1	**MI-47**	5.0	4.9	71.1	731.78
2	**MI-66**	5.0	4.9	71.2	940.93
3	**MI-79**	4.5	4.6	72.1	1047.14
4	**ME-03**	4.6	4.5	73.6	1233.24
5	**ME-65**	4.4	4.4	70.6	753.87
6	**ME-67**	5.2	5.2	72.4	696.69
7	**ME-146**	5.0	5.0	71.9	1073.00

Moisture content (%). Among the evaluated hybrids, ME-146 × MI-66 (74.8%) and ME-03 × MI-66 (74.6%) showed the highest leaf moisture content at 60 days after pruning, while ME-65 × V1 recorded the lowest (69.3%). Significant positive mid-parent heterosis for moisture content ranged from 4.38% to 15.02%, with ME-146 × MI-66 showing the highest increase. This hybrid also exhibited the maximum positive heterosis over the better parent (14.48%) and check variety (6.78%), indicating superior moisture retention (Table 8, 9).

Leaf moisture content is critical for leaf yield and silkworm health, as higher moisture enhances feed consumption, larval growth, and cocoon yield. The results are consistent with previous studies by Sikdar (1990); Manjula and Kumari (2017), which found that leaf moisture positively influences silkworm performance by improving larval growth and cocoon parameters, particularly in the V1 mulberry variety. Hybrid ME-146 × MI-66 emerges as a promising candidate for improving moisture content in mulberry breeding programs.

Table 9: Estimates of heterosis over mid parent, better parent and standard check for moisture (%) and dry matter content (%) in mulberry hybrids.

Sr. No.	Hybrids	Moisture content (%)			Dry matter content (%)		
		MPH (%)	BPH (%)	SH (%)	MPH (%)	BPH (%)	SH (%)
1.	MI-47 × MI-66	9.13*	9.07**	0.79	12.16**	12.01**	-1.85
2.	MI-79 × MI-66	8.47**	8.47**	1.48	12.17**	10.45	-3.47
3.	ME-03 × MI-66	13.24**	11.38**	6.42**	1.48	-2.78	15.03**
4.	ME-146 × MI-66	15.02**	14.48**	6.78**	-2.60	-3.73	15.87**
5.	ME-65 × V1	4.38**	0.02	0.02	8.29**	2.45	2.45
6.	ME-67 × V1	6.19**	2.95**	2.95**	1.23	-6.89**	-6.89**

MPH- Mid Parent Heterosis, BPH-Better Parent Heterosis, SH-Standard Heterosis, *Significance at 5% level, **Significance at 1% level

Leaf yield per plant (g). Mulberry leaf yield is a complex trait influenced by many characters, and the cumulative effects of this heterosis determine the performance of crosses on leaf yield. Plant breeders prefer crosses with high levels of heterosis for leaf yield over the better parent.

In evaluating leaf yield, the standard check V1 recorded the highest yield per plant (1298.5 g) at 60 days after pruning, while hybrids ME-03 × MI-66 (1254.38 g), ME-65 × V1 (1080.91 g), and MI-79 × MI-66 (1182.39 g) showed no significant difference compared to V1. Significant positive mid-parent heterosis was observed in three hybrids, with MI-79 × MI-66 exhibiting the highest heterosis (30.84%). Similarly, MI-79 × MI-66 recorded significant positive better-parent heterosis (24.21%), while other hybrids showed non-significant results (Table 3 & 6). All hybrids displayed negative or non-significant standard heterosis for leaf yield compared to the check variety. Despite this, hybrids MI-79 × MI-66, ME-03 × MI-66, and ME-65 × V1 produced leaf yields comparable to V1. These findings align with Ghosh *et al.* (2009), who demonstrated the potential for hybrids to surpass commercial varieties, and with Vasal *et al.* (1993), highlighting the importance of progeny testing to identify superior high-yielding hybrids.

Table 10: Estimates of heterosis over mid parent, better parent and standard check for Moisture content (%) and leaf yield per plant (g) in mulberry hybrids.

Sr. No	Hybrids	Moisture content (%)			Leaf yield/plant (g)		
		MPH (%)	BPH (%)	SH (%)	MPH (%)	BPH (%)	SH (%)
1.	MI-47 × MI-66	9.13*	9.07**	0.79	23.58*	9.84	-24.01**
2.	MI-79 × MI-66	8.47**	8.47**	1.48	30.84**	24.21*	-4.37
3.	ME-03 × MI-66	13.24**	11.38**	6.42**	16.23*	-3.73	1.45
4.	ME-146 × MI-66	15.02**	14.48**	6.78**	-8.75	-14.36	-32.44**
5.	ME-65 × V1	4.38**	0.02	0.02	12.49	-12.58	-12.58
6.	ME-67 × V1	6.19**	2.95**	2.95**	-42.50**	-56.52**	-56.52**

MPH- Mid Parent Heterosis, BPH-Better Parent Heterosis, SH-Standard Heterosis, *Significance at 5% level, **Significance at 1% level

CONCLUSIONS

The evaluation of hybrids for growth and yield traits reveals notable hybrid vigour among the tested clones. Hybrid ME-03 × MI-66 exhibited superior performance for several traits, including the number of leaves, fresh leaf weight, moisture content, leaf shoot ratio, and specific leaf area, outperforming the mid-parent, better parent, and standard check V1.

While negative heterosis for internodal distance was prevalent, indicating a reduction in internodal length, hybrid ME-65 × V1 stood out for its consistent performance across various traits. It showed no significant difference in leaf yield, single leaf area, moisture content, number of leaves per branch, number of branches per plant, and other economic traits when compared to mid, better parent, and standard heterosis performing on par with the standard check V1.

In conclusion, the present study analyzed the genetic variation and superior growth and yield characteristics, while ME-65 × V1 shows comparable performance to traditional hybrids. Both hybrids hold promise for enhancing mulberry breeding programs due to their improved economic and yield-related traits. Based on performance of progenies under irrigated condition over season, two hybrids were identified as high yielding over variety V1 which need to be evaluated further following systematic evaluation procedures of mulberry for identify high yielding variety in Mulberry.

FUTURE SCOPE

The hybrids exhibiting desirable traits, such as enhanced leaf yield, moisture content, and leaf shoot ratio, can serve as valuable genetic resources in mulberry breeding programs. They can be utilized as parent lines to develop new hybrids with even better performance, further boosting the productivity and resilience of mulberry plants. Two promising hybrids identified for growth economic traits are to be tested under replicated yield trials along with the commercial checks in different seasons. Hybrids are further need to be tested for pest and disease occurrence and evaluated for chawki rearing.

Acknowledgement. Authors are thankful to the Department of Sericulture, University of Agricultural Sciences, Bangalore, for providing all necessary facilities during the entire research work.

Conflict of Interest. None.

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How to cite this article: Lohithashwa K.M., Chikkalingaiah, Sapna J.S. and Sushmitha C. (2024). Mean Performance and Hybrid Vigour in Mulberry for different Growth and Yield Traits (*Morus* spp.). *Biological Forum – An International Journal, 16*(8): 215-224.