Genetic variability, correlation and path analysis of physiological and yield attributes in mulberry (*Morus spp.*)

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ABSTRACT

Improvement of leaf productivity and quality is the most sustainable and economic approach in sericulture development programme. The experiment was conducted to study the "Genetic variability, Correlation and path coefficients in 982 segregating progenies derived from 57 crosses (utilizing 6 females and 12 male parents), in Gangetic Alluvial plains of West Bengal at the Central Sericultural Research and Training Institute, Berhampore during June 2014 to July 2016. The investigation revealed the high phenotypic and genotypic coefficient of variation for leaf yield per plant, petiole weight, shoot yield, single lamina weight, total shoot length, single leaf area, shoots per plant, total chlorophyll content and inter-nodal distance indicated the existence of greater magnitude of genetic variability in the population. Character association analysis revealed that leaf yield per plant was positive and significantly associated with all the traits studied viz., shoot yield per plant (0.952), total shoot length (0.788) and plant height (0.671), except for leaf moisture and total chlorophyll content etc.. Besides, total shoot length (1.295), shoots per plant (0.619), single lamina weight (0.411) and harvest index (0.205) had a high direct effect on the leaf yield and can be used for indirect selection or as secondary traits in the selection process. Hence, total shoot length, laminar weight, shoots per plant, specific leaf area and harvest index may be considered has a useful surrogates for selection among segregating progenies in mulberry improvement programme.

Keywords : Association, coefficient of variation, direct effect and segregating progenies.

Mulberry belongs to the family Moraceae and its leaves are sole food for the silkworm Bombyx mori. It is a heterozygous perennial species, native of Indo-China and found widely distributed in both the hemispheres (Jolly and Dandin, 1986). Owing to unisexual flowers and cross fertilization, wider range of variation exists in natural populations. It is a tree species maintained mostly as low bush under intensive commercial cultivation. The quantity and quality of leaf have major impact towards the production of a successful cocoon crop (Ravikumar, 1988). Therefore, improvement in mulberry production through development of new varieties with high leaf yield and better adaptability is essential for sustainable growth of sericulture (Vijayan et al., 2012). Genetic variability and correlation coefficients are pre-requisites for improvement of any crop including mulberry as it indicates nature and magnitude of genetic makeup of traits (Vijayan, et al., 1997; Tikader and Roy, 1999). Hybrid progenies derived from two parents show considerable variability owing to heterozygous and heterogeneous nature of mulberry (Ghosh et al., 2009). Moreover, knowledge of heritability and GAM (Genetic advance as percent of mean) indicates the extent of transmissibility of a trait into future generations. Breeding programes to develop high yielding varieties depends on the nature and magnitude of variation in available genotypes or population. Mulberry improvement for leaf yield is possible through selection for desired component traits. Therefore knowledge of the interrelationships among various traits and also their direct and indirect contribution toward yield is necessary, to design appropriate selection criteria in mulberry breeding programme (Vijayan et al., 2012). Correlation coefficient analysis provides information about the degree of relationship between important component traits and when higher numbers of variables are considered in correlation, the interrelationship becomes more complex (Dewey and Lu, 1959). Use of path coefficient analysis would be more appropriate as it reveals direct and indirect associations and identifies the most reliable yield-contributing traits. Evaluation of any crop is a continuous process to evolve new varieties suitable for different agro climatic regions. Hence, the present study was undertaken to know extent of genetic variability and identification of yield-contributing traits to determine the relationship among characters and their association with leaf yield among segregating progenies of mulberry.

MATERIALS AND METHODS

A field experiment was conducted with 982 segregating progenies (developed by crossing 6 females with 12 male parents) at the experimental plots of Central Sericultural Research and Training Institute, Berhampore

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which is situated at an altitude of 19 m above mean sea level, 34° 0' 28" North, 71° 34' 24" East, during June 2014 to July 2016. The soil was Gangetic Alluviul with p^H was 6.9, EC was 0.12 m mhos cm⁻¹, Organic Carbon was 0.56% and humid sub-tropical climate with average annual rainfall was 1377 mm. Seedling of segregating progenies along with the local check variety S-1635 was transplanted in main plot with a spacing of 60cm X 60 cm. The recommended fertilizer dose (NPK: 336:180:112 kg/ha/y) of the region was followed to raise healthy crop (Ghosh et. al., 2011). The plantation was of two years old and maintained at six inches stump height above ground level for bush type cultivation.

Growth and yield parameters : Observations were recorded on three physiological viz., total chlorophyll content (µgcm⁻²), specific leaf area (cm²g⁻¹), leaf moisture content (%) and eleven morphological traits viz., single leaf area(cm²), single lamina weight(g), petiole weight(g), total shoot length(cm), length of the longest shoot (cm), number of shoots per plant, average shoot length(cm), shoot yield, inter-nodal distance(cm), harvest index and leaf yield per plant(g). Total chlorophyll content, leaf area, leaf moisture, leaf lamina weight and petiole weight was recorded for 5th leaf from top of the longest shoot. Total chlorophyll content was estimated by chlorophyll concentration index (CCI) value obtained from chlorophyll concentration meter (CCI-800). Chlorophyll concentration index units was converted into total chlorophyll content (µgcm⁻²) based on conversion formulae "total chlorophyll content ($\mu g \text{ cm}^{-2}$) = (117.1* CCI)/ (148.84 - CCI) given by Cerovic et al., 2012. Moisture content (%) is ratio of difference in fresh leaf weight at harvest and oven dry weight (Tikader and Roy.1999). Single leaf area in square centimeters is measured by using Leaf area meter (3100C LiCor Area Meter). Specific leaf area is determined according to Vile et al., 2005.

Statistical analysis: The experimental data were analyzed statistically following the method of analysis of variance for single factor (Gomez and Gomez, 1984). Mean, Variance components (viz., PV, GV, EV), phenotypic, genotypic and environmental coefficients of variation (viz., PCV, GCV, ECV), were estimated as per Singh and Chaudhary (1979). The broad sense heritability and genetic advance (GA), genetic advance as percent of mean (GAM) were calculated as suggested by Johnson et al. (1955). Correlation analysis was computed using SPSS 10.0 software and the partitioning of correlation coefficient into direct and indirect effects (path analysis) was carried out using the procedure suggested by Dewey and Lu (1959) using TNAUSTAT statistical package version 1.

Source

| SN. | Name | Accession No./Pedigree | Sex | |
|-----|--------------|------------------------|----------|--|
| 1 | M. indica HP | MI - 0099 | 우 | |
| 2 | China White | ME - 0042 | o | |

Table 1: List of parents used in development of hybrid progenies

| SIN. | Iname | Accession No./Pedigree | Sex | Source |
|------|---------------------------|----------------------------|-----|------------------|
| 1 | M. indica HP | MI - 0099 | 우 | India (WB) |
| 2 | China White | ME - 0042 | 우 | China |
| 3 | Chinese F ₁ 10 | - | 우 | India (WB) |
| 4 | MS-30 | - | 우 | India(Mysore) |
| 5 | M. multicaulis | ME - 0006 | 우 | Russia |
| 6 | Kajli OPH | MI - 0068 | 우 | India (WB) |
| 7 | V- 1 | $S30 \times C776$ | 3 | India (Mysore) |
| 8 | Ac.No.1190 | CSRT-MI-0120 | 3 | India(Mysore) |
| 9 | Almora Local | MI - 0015 | 3 | India (UP) |
| 10 | Berhampore-B | MI - 0627 | 3 | India (WB) |
| 11 | Bishnupur-10 | MI - 0117 | 3 | India (WB) |
| 12 | C-776 | English balck x Multiculis | 3 | India (WB) |
| 13 | Charitul | MI - 0169 | 3 | India (J & K) |
| 14 | English Black | ME - 0004 | 3 | France |
| 15 | Kosen | ME - 0066 | 3 | Japan |
| 16 | KPG-1 | MI - 0144 | 3 | India (WB) |
| 17 | MS-7 | CSRT-MI-0069 | 3 | India (Mysore) |
| 18 | Nagaland | MI - 0167 | 3 | India (Nagaland) |

J. Crop and Weed, 13(1)

| | rifudo iorio | leaf | Moisture | | renoie weight | | shoot | ł | tt Shoots ht per plant | • | | | Shoot yield | Inter- nodal | Harvest index |
|---------------------|-----------------------------|---------------------|--------------|----------------------------|------------------|---------------|----------------|--------------|---------------------------|--------------|-------------------------------|--------------------------|--------------------------|------------------|------------------|
| | (µgcm ⁻²) | area (cm^2g^{-1}) | (%) | area (cm ²) | (g) | weight (g) | length (cm) | t (cm) | ~ | le (| length (₁ (cm) | (g.plant ⁻¹) | (g.plant ⁻¹) | distance (cm) | |
| Mean | 15.49 | 261.57 | 77.43 | 110.56 | 0.18 | 1.96 | 674.83 | 3 120.55 | 55 6.57 | | 101.98 | 243.75 | 560.50 | 3.90 | 0.43 |
| Min. | 5.60 | 135.98 | 49.46 | 25.89 | 0.02 | 0.29 | 63.00 | 32.00 | 0 2.00 | | 31.50 | 23.80 | 62.20 | 1.50 | 0.09 |
| Мах. | 32.00 | 409.29 | 92.50 | 268.89 | 0.88 | 5.40 | 1845.00 | 0 214.00 | , , , | | 172.00 | 1064.40 | 2204.00 | 8.00 | 0.88 |
| SD | 4.12 | 37.90 | 4.27 | 43.07 | 0.10 | 0.86 | 265.56 | 5 24.35 | | | 20.32 | 142.08 | 304.32 | 0.85 | 0.08 |
| 5ØBP | 16.97 | 1436.25 | 18.21 | 1855.05 | 0.01 | 0.73 | 70520.63 | 53 592.70 | | - | 413.02 2 | 20186.60 | 92609.99 | 0.72 | 0.01 |
| 5ØBG | 15.90 | 1309.61 | 15.90 | 1676.45 | 0.01 | 0.61 | 65669.97 | 97 488.93 | | | 352.59 1 | 15567.39 | 75489.84 | 0.69 | 0.01 |
| 5ØBE | 1.07 | 126.64 | 2.31 | 178.60 | 0.00 | 0.13 | 4850.67 | 7 103.77 | | | 60.43 | 4619.22 | 17120.15 | 0.03 | 0.00 |
| PCV | 26.60 | 14.49 | 5.51 | 38.96 | 57.42 | 43.74 | 39.35 | 20.20 | | | 19.93 | 58.29 | 54.29 | 21.76 | 18.94 |
| GCV | 25.74 | 13.84 | 5.15 | 37.04 | 51.50 | 39.79 | 37.97 | 18.34 | 4 30.31 | | 18.41 | 51.19 | 49.02 | 21.33 | 17.38 |
| ECV | 6.69 | 4.30 | 1.96 | 12.09 | 25.40 | 18.17 | 10.32 | 8.45 | 5 11.46 | | 7.62 | 27.88 | 23.34 | 4.30 | 7.52 |
| h2(BS) | 93.68 | 91.18 | 87.32 | 90.37 | 80.44 | 82.74 | 93.12 | 82.49 | | | 85.37 | 56.95 | 70.27 | 84.98 | 80.08 |
| GA | 7.95 | 71.19 | 7.68 | 80.18 | 0.17 | 1.46 | 509.42 | | 7 3.84 | | 35.74 | 225.71 | 511.01 | 1.68 | 0.14 |
| GAM | 51.33 | 27.22 | 9.91 | 72.53 | 95.16 | 74.56 | 75.49 | | 2 58.41 | | 35.04 | 92.60 | 91.17 | 43.07 | 32.85 |
| Characters | LS | | X1 | X2 | X3 | X4 | X5 | X6 | X7 | X8 | X9 | X10 | X11 | X12 | X13 |
| Total abla | Total ablanabull (V1) | | 1 000 | | | | | | | | | | | | |
| Specific le | Specific leaf area (X2) | - | -0.326** | 1 000 | | | | | | | | | | | |
| Leaf moisture (X3 | ture (X3) | - - | -0.087 | 0.464^{**} | 1.000 | | | | | | | | | | |
| Single leaf | Single leaf area (X4) | C | | -0.129 | 0.074 | 1.000 | | | | | | | | | |
| Petiole weight (X5) | ight (X5) | C | 0.250^{**} | -0.220** | 0.179 | 0.775* | 1.000 | | | | | | | | |
| Single lam | Single lamina weight (X6) | _ | 0.251^{**} | -0.227^{**} | 0.243^{**} | 0.947* (| 0.829* | 1.000 | | | | | | | |
| Total shoo | Total shoot length (X7) | C | 0.184 | | | | | 0.390^{**} | 1.000 | | | | | | |
| Plant height (X8) | ht (X8) | C | 0.279^{**} | -0.033 | - | * | * | 0.508^{**} | 0.663^{*} | 1.000 | | | | | |
| Shoots per | Shoots per plant (X9) | C | 0.074 | -0.060 | 0.021 | 0.182 (| 0.159 | 0.189 | 0.858^{*} | 0.296^{**} | 1.000 | | | | |
| Average sl | Average shoot length (X10) | | 0.262^{**} | -0.012 | * | - | 0.389** | 0.474^{**} | 0.573^{**} | | 0.115 | 1.000 | | | |
| Shoot yiel | Shoot yield per plant (X11) | | 0.217 | | 0.165 | - | | 0.598^{**} | 0.817* | _ | 0.570^{**} | • 0.644** | 1.000 | | |
| Inter-noda | Inter-nodal distance (X12) | _ | 0.036 | -0.059 | 0.129 | * | * | 0.431^{**} | 0.234^{**} | | 0.125 | 0.260^{**} | 0.379^{**} | * 1.000 | |
| Harvest index(X13) | dex(X13) | | | | | | | 0.128 | 0.062 | 0.045 | 0.021 | | | -0.108 | |
| I asf wiald | Last wald nor alont (V1A) | | 0.201 | -0 303 | 0 167 | 0 575** | 0 500** | 0 605** | 0 788* | 0 671 ** | 0 5/1 ** | ** 0 627** | 0 050* | 0 300** | 0 282** |

Analysis of physiological and yield attributes in mulberry

J. Crop and Weed, 13(1)

| | | | • | | | • | | | | | | |
|----------------------------|--------|------------|------------|--------|--------|--------|--------|------------|------------|--------|--------|--------|
| Characters | X1 | X 2 | X 3 | X4 | X5 | X6 | X7 | X 8 | X 9 | X10 | X11 | X12 |
| Total chlorophyll (X1) | -0.010 | -0.016 | 0.007 | -0.026 | -0.005 | 0.103 | 0.238 | 0.015 | -0.046 | -0.061 | 0.002 | 0.002 |
| Specific leaf area (X2) | 0.003 | 0.049 | -0.038 | 0.016 | 0.005 | -0.093 | -0.075 | -0.002 | 0.037 | 0.003 | -0.004 | -0.003 |
| Leaf moisture (X3) | 0.001 | 0.023 | -0.082 | -0.00 | -0.004 | 0.100 | 0.179 | 0.011 | -0.013 | -0.058 | 0.009 | 0.011 |
| Single leaf area (X4) | -0.002 | -0.006 | -0.006 | -0.122 | -0.016 | 0.389 | 0.475 | 0.027 | -0.113 | -0.104 | 0.029 | 0.025 |
| Petiole weight (X5) | -0.003 | -0.011 | -0.015 | -0.095 | -0.021 | 0.341 | 0.433 | 0.023 | -0.098 | -0.091 | 0.024 | 0.012 |
| Single lamina weight (X6) | -0.003 | -0.011 | -0.020 | -0.116 | -0.017 | 0.411 | 0.505 | 0.028 | -0.117 | -0.111 | 0.029 | 0.027 |
| Total shoot length (X7) | -0.002 | -0.003 | | -0.045 | -0.007 | 0.160 | 1.295 | 0.036 | -0.531 | -0.134 | 0.016 | 0.013 |
| Plant height (X8) | -0.003 | -0.002 | | -0.060 | -0.009 | 0.209 | 0.859 | 0.055 | -0.183 | -0.206 | 0.019 | 0.010 |
| Shoots per plant (X9) | -0.001 | -0.003 | | -0.022 | -0.003 | 0.078 | 1.111 | 0.016 | -0.619 | -0.027 | 0.009 | 0.005 |
| Average shoot length (X10) | -0.003 | -0.001 | -0.020 | -0.054 | -0.008 | 0.195 | 0.742 | 0.048 | -0.071 | -0.234 | 0.018 | 0.015 |
| Inter-nodal distance (X11) | -0.001 | -0.003 | -0.011 | -0.052 | -0.008 | 0.177 | 0.303 | 0.015 | -0.078 | -0.061 | 0.068 | -0.022 |
| Harvest index(X12) | 0.002 | -0.001 | -0.004 | -0.015 | -0.001 | 0.053 | 0.082 | 0.003 | -0.014 | -0.017 | 0.007 | 0.205 |
| Residual effect= 0.196 | | | | | | | | | | | | |

Table 4: Estimates of the direct (bold) and indirect effects of yield attributes on foliage yield

J. Crop and Weed, 13(1)

RESULTS AND DISCUSSION

The parents used to develop the segregating population in this experiment are presented in table 1. The parents were collected from germplasm bank maintained at the same institute. To conquer the problem of low productivity in perennial crops like mulberry, it is pertinent to identify high yielding genotypes among segregation progenies. Identification of these better genotypes, their introduction in clonal evaluation trials coupled with establishment of suitable selection criteria will be helpful for successful mulberry improvement programme.

The results revealed high significant differences among the progenies for all the traits studied, which indicated the existence of sufficient genetic variability and thus providing the scope for selection of progenies to improve the leaf productivity. All the traits under study exhibited high variability as evident from the ranges of mean values and standard deviation. In the progenies under study maximum range of variability (Table 2) was observed for total shoot length (63 to1845 cm), leaf yield plant (23.82 to1064.4 g), specific leaf area (135.9 to 409.3 cm²g⁻¹), single leaf area (25.9 to 268.9 cm²) and length of the longest shoot (32-214 cm). The presence of large amount of variability among the progenies was also reported by earlier worker Doss *et al.* (2006).

The genetic parameters of physiological and leaf yield attributes among the segregating progenies is presented in table 2. The genetic analysis of metric traits is a prerequisite for plant breeding programmes, which can lead to the appropriate planning of plant breeding strategies. The current study suggests that the phenotypic variance (σP) of all traits was higher than the genotypic variance (σ G). Similarly, the phenotypic coefficient of variation (PCV) was also higher than genotypic coefficient of variation (GCV). Mulberry leaf yield being a quantitative trait and observed variability is the sum total of genetic effects as well as the environment. High degree of phenotypic and genotypic coefficient of variation (>20%) were observed for leaf yield plant⁻¹, petiole weight, shoot yield, single lamina weight, total shoot length, single leaf area, shoots plant⁻¹ total chlorophyll content and inter-nodal distance, indicated that these traits are governed by genetic factors and existence of greater magnitude of genetic variability in the population, thus provide ample scope for improvement through selection. The results agree with several workers Tikader and Roy (2001) and Tikader and Dandin (2005). While, moderate values(<20%) were recorded for length of longest shoot, average shoot length, harvest index, specific leaf area and lower values for leaf moisture content, indicated that selection for these traits is less effective.

The influence of environment was observed to minimum for all the traits under study except leaf yield, petiole weight, shoot yield and single lamina weight, indicating the role of environment effect on the expression of these traits and that selection based on these traits alone is not effective. It also suggests that selection on these traits may be effective in future evaluation trials *viz.*, primary or final yield trial. These results agree with the observations made by and Tikader and Kamble (2008), Doss *et al.* (2012).

The selection efficiency was higher when the parameters had higher heritability. High broad sense heritability values for all traits studied ranged from 57 per cent for leaf yield to 91 per cent. All traits had high heritability estimates (80 % to 94%) except for leaf yield and shoot yield, suggesting influence of fixable additive gene effects for inheritance of these traits (Ajayi, et al., 2014). Therefore, they can be scored by their phenotypic performance (Khatun, et al., 2015) and selection for these traits may lead to fast genetic improvement. For heritability estimates to be reliable, it must be in conjunction with high genetic advance for a reliable index for selection of traits. Earlier studies in mulberry also stated that metric traits with high h² and GAM responded better to simple phenotypic selection due to their additive gene action (Doss et al., 2012). Thus a trait possessing high heritability along with high GAM will be useful for selection in segregation population. High heritability coupled with high GAM (>80%, >20%) was observed for almost all studied traits except for leaf moisture content, indicating the prevalence of additive gene action in the expression of these traits and effective progress in improvement through selection could be achieved for leaf yield. These results are in agreement with the findings of Doss (2006), Tikader and Kamble(2008), Maji (2009) Keshava Murthy et al. (2010) in mulberry.

Phenotypic correlations are estimated directly from values measured in the field and only the genetic portion of it is used to guide breeding programs. Correlation studies enable to quantify indirect gains due to selection on correlated traits, and also to evaluate the complexity of the traits (Tiwari & Upadhyay, 2011). Correlations are measures of the intensity of interrelationships between traits, whereby selection for a trait results in progress for other positively associated traits. Pearson correlation analysis among yield and its contributing traits are shown in table 2. Significant positive phenotypic correlations to leaf yield were recorded for the all traits under study except for leaf moisture content and specific leaf area, indicated that the selection for these will result in increase in leaf yield. Various authors reported positive association of leaf yield with other traits in mulberry viz., Vijayan et al., 1997, Tikadar and Roy (2003), Rahman et al., 2006, Doss et al., 2012. Significant negative correlation was observed between total chlorophyll content with specific leaf area (-0.326**) and leaf moisture content (-0.087). Specific leaf area also had significant negative correlation with petiole weight (-0.220**), single laminar weight (-0.227**) and leaf yield (-0.303**). Marenco et al., 2009 observed a negative relationship between SPAD values based total chlorophyll content with specific leaf area and the leaf water content in Amazonian tree species. Unit leaf lamina weight had significant positive correlations with unit leaf area (0.947^{**}) and petiole weight (0.829^{**}) . Where as in earlier studies of Rahman et al. (2006) positive correlations were observed between leaf weight and leaf area.

The path coefficient analysis (Table 4) provides a more realistic evidence of the interrelationship, as it partitions into direct as well as indirect effects via various yield contributing traits. Path coefficient analysis showed that total shoot length had maximum direct effect (1.295) followed by single lamina weight (0.411), harvest index (0.205), inter-nodal (0.068), plant height (0.055) and distance and specific leaf area (0.049) on leaf yield plant⁻¹. The greatest direct positive effect of total shoot length on leaf yield may be attributed to its influence on numbers of leaves indicating that selection for this trait should be done considering its positive indirect effects. Shoots plant⁻¹ (-0.619), average shoot length (-0.234), single leaf area (-0.122) and leaf moisture (-0.082) showed negative direct effect on leaf yield and supported with the similar findings of Doss, 2012.

Direct selection for the leaf yield might be more effective if indirect selection via yield contributing traits with high heritability is considered. Shoots plant⁻¹ (1.111), plant height (0.859), average shoot length (0.742) and single leaf area (0.475) had high positive indirect effect via total shoot length but low direct effects on leaf yield, Thus for developing high yielding mulberry genotypes emphasis should be placed on shoots plant⁻¹, plant height and single leaf area. Therefore, selection for tall plants with more shoots, larger thick leaf and uniform height shoots is very important to improve leaf yield in mulberry.

In conclusion it can be said that genotypic and phenotypic coefficient of variation was high for, leaf yield plant⁻¹, petiole weight, unit leaf lamina weight, total shoot length, unit leaf area, shoots plant⁻¹ and total chlorophyll content thus indicating the presence of variability. Moreover high heritability along with high genetic gain as a percentage of mean and their significant correlation with leaf biomass suggested that total shoot length, shoots per plant and unit leaf laminar weight can be considered as selection criterion for mulberry improvement.

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J. Crop and Weed, *13*(*1*)