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Combining ability analysis of seven silk yield attributes in silkworm, *Bombyx mori* L.

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Abstract. The combining ability effects of five silkworm (*Bombyx mori* L.) strains and their 20 F_1's including reciprocals were analysed in a 5×5 diallel cross for seven silk yield attributes, viz. effective rate of rearing (ERR), a measure of survival, cocoon yield, cocoon weight, shell weight, raw silk percentage, silk filament length/cocoon and silk reelability. Higher values of specific combining ability (SCA) than the corresponding general combining ability (GCA) for all the attributes except ERR and cocoon yield are indicative of non-additive gene action. The parental strain JC2P was the best general combiner for all the attributes, except cocoon yield. The highest general combining ability effect for cocoon yield was shown by N4. The highest desirable or positive SCA effects resulted from N4 × SH2 (all the attributes except shell weight), JC2P × 14M (all the attributes except cocoon weight and shell weight), 14M × SH2 (all the attributes except ERR), N4 × SPJ1 (all the attributes except shell weight and raw silk percentage). Only one reciprocal, namely SPJ1 × N4, showed positive effects for all the attributes except silk reelability. The improvement of cocoon yield through mass selection followed by intermating and the use of the parental strain JC2P in future cross breeding are discussed.

Key words: *Bombyx mori*, combining ability, silk yield.

Commercial exploitation of mulberry caterpillars (*Bombyx mori*) commonly known as silkworm has resulted in the production of 72,879 tones of raw silk (CURRIE 1991). World silk production has doubled during the last 30 years inspite of artificial fibres replacing silk for the same uses (CHERRY 1987). India is the second largest producer of silk thread after China (BHARGAVA et al. 1992). Some European countries like France, Italy and Spain which used

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to have a fairly large scale of sericulture are no longer engaged in mulberry planting and silkworm rearing. In India, the sericulture or silk production is now identified as a major agro-based industry which has tremendous potential of increasing income of jobless farmers of rural areas.

Silk is a natural fibre made by silkworms. There are about 2,000 different strains of silkworm in the world which differ in their silk yield attributes both quantitatively and qualitatively (THIAGARAJAN et al. 1993a). The standard breeding methodology for the improvement of silk yield attributes has been limited to examination of racial/strains differences and selection from induced variability with very few genetic investigations in silkworm (PETKOV 1975, 1978, 1981, PERSHAD et al. 1986a, b, DATTA, PERSHAD 1987, BHARGAVA et al. 1992, CHATTERJEE, DATTA 1993, THIAGARAJAN et al. 1993b). The diallel cross system (GRIFFING 1956) is one of the best approaches for assessing the nicking ability of parents that aids in selecting the parents which when crossed would give rise to more desirable segregants.

Though combining ability has been extensively used in plant crop breeding (SMITH et al. 1973, TRIPATHI et al. 1979, JAIMINI, MATHUR 1980, NAWAR et al. 1980, CHAWLA, GUPTA 1982, SRIVASTAVA et al. 1986), it has not been effectively utilized for silkworm improvement. The purpose of this investigation was to study the type of gene action involved in the inheritance of silk yield attributes and to identify superior strains/crosses for exploitation in the silkworm breeding programme.

Materials and methods

Rearing of silkworms

Five promising and genetically diverse parental strains of silkworm (*Bombyx mori* L.), namely N4, 14M, SPJ1, JC2P (all of Japanese origin) and SH2 (of Indian origin) constituted the experimental material of the present study. These strains have been maintained in the germplasm collection of Regional Sericultural Research Station, Coonoor, since 1987.

Crosses in all possible combinations including reciprocals were made. Thus, 20 F_1's along with their five parents were reared during May-June 1992 in a randomised block design. The hatched silkworms were reared under standard rearing conditions (temperature 23-28°C; relative humidity 70-90% and 12/12 h dark/light cycle), by the method described in an earlier paper (BHARGAVA et al. 1993). Before starting the experiment, the rearing place and equipment were disinfected with 2% formalin spray and 5% bleaching powder solution. The silkworm eggs were masked with black cloth a day before hatching and then exposed to light for uniform hatching. Fresh and tender bits of mulberry leaf were fed to the newly hatched larvae. After 30 min. the larvae were brushed and taped onto the rearing bed inside the trays (Brushing: silkworm eggs will be either on egg sheet or in loose form. Brushing is the process of separating newly hatched larvae gently and carefully from the empty egg shells or egg sheets and transferring them to the rearing trays with the help of a smooth brush). The larvae were fed four times a day till their maturity. Feeding was stopped during ecdysis. Regular cleaning of silkworms' beds was carried out with the cleaning nets to keep the beds hygienic. Adequate spacing was also provided to worms for proper population density throughout the rearing period. Suitable leaves were fed according to the age of worms. Mature worms ready for spinning the cocoons were mounted. The silk cocoon crop was harvested after the completion of pupation. Leaf of 'MR2' variety of mulberry was fed to silkworms and about 250 kg leaf was utilized to rear 10,000 worms. For each strain/hybrid, a minimum of 10 layings (total number of eggs laid by ten mother moths) was brushed en masse and after the IIIrd ecdysis, the larvae were distributed into five replications, each with 500 larvae reared in 60×90 cm standard wooden rearing trays. For spinning of the cocoons plastic mountages were used. The positions of the trays were changed regularly 3-4 times a day to reduce the effect of environmental factors.

Estimation of silk yield attributes and combining ability

Observations were made on seven silk yield attributes:
1 – effective rate of rearing (ERR), i.e. a measure of survival expressed in percentage. ERR was estimated from the number of cocoons formed by 500 larvae in each replication.
2 – the weight of a cocoon (g).
3 – cocoon shell weight (g). The mean weight of ten male and ten female cocoons and cocoon shells for each replication was considered as the weight of a cocoon and cocoon shell.
4 – cocoon yield per 10,000 larvae brushed. This is noted as weight of cocoons harvested from 10,000 larvae brushed.
5 – raw silk percentage (silk filament is recovered from the outer cocoon shell with the reeling machine in the form of a long filament or fibre. Raw silk percentage denotes the percentage of raw silk reeled from cocoons. For example, 12% raw silk percentage means 12 kg of raw silk can be reeled from 100 kg of cocoons).

6 – silk filament length/cocoon (the length of silk fibre reeled from the cocoon shell).
7 – silk reelability (the percentage of silk filament length reeled easily without any break).

The average values for three silk yield attributes, viz. raw silk percentage, silk filament length/cocoon and silk reelability were obtained from the reeling results of cocoons. Data recorded on all the seven silk yield attributes were subjected to the combining ability analysis using Model-1 and Method-1 of GRIFFING (1956).

Process followed for reeling of cocoons

After harvesting, a total of 400 g cocoons were randomly taken from each replication for reeling silk thread. All the cocoons were dried in electrical hot air oven for 6 h (first 2 h at 90°C; next 2 h at 70°C and last 2 h at 60°C) to achieve 40-42 driage percentage. The dried cocoons were conditioned under room temperature (23-28°C) in cocoon storage racks for a minimum of two days. Dried and conditioned cocoons were cooked in a three pan cooking unit. Before cooking the cocoons were subjected to 60 s soaking at 50°C. The duration of cooking and temperature maintained in the 1st, 2nd and 3rd pans were 150 s and 90°C, 60 s and 65°C and, 120 s and 95°C, respectively. After this treatment a cold water shower was provided in the 3rd pan for 30 s followed by restore steam supply for 30 s. Again cold water was sprayed to the 3rd pan to bring down the temperature to 60°C followed by hand brushing with the help of a soft brush. The cooked cocoons ready for reeling were reeled with Japanese multiend reeling machine following the standard technique (three reeling ends with eight cocoons in each end) as described by SONWALKAR (1985). A constant reeling speed (90 m/min.) was maintained and the temperature of the reeling basin was kept between 40-45°C.

Results

Analysis of variance for combining ability

Significant (0.05 level) to highly significant (0.01 level) differences were observed for general combining ability, specific combining ability and reciprocal defferences for all seven silk yield attributes. However, general combining ability for cocoon weight and specific combining ability for effective rate of rearing were found to be non-significant (Table 1). From the estimates

Table 1. Analysis of variance of combining ability and estimates of variance components for seven silk yield attributes in *Bombyx mori* L.

Source of variation	df	Silk yield attributes						
		ERR	cocoon yield	cocoon weight	shell weight	raw silk percentage	silk filament length/ cocoon	silk reelability
GCA	4	55.022*	4.584*	0.002	0.0008*	1.864*	7115*	18.297*
SCA	10	4.812	2.892*	0.014*	0.0007*	3.191*	10540*	68.605*
Reciprocal differences	10	20.449*	3.812*	0.005*	0.0007*	6.578*	15003*	12.955*
Error	97	4.156	1.106	0.001	0.0001	0.602	573	5.214
GCA variance (σ_g^2)		5.024	0.178	–0.001	0.0000	–0.120	–295	–4.729
SCA variance (σ_s^2)		0.390	1.063	0.007	0.0004	1.541	5933	37.733
$2\sigma_g^2/(2\sigma_g^2+\sigma_s^2)$		0.963	0.251	–0.231	0.06	0.185	–0.110	–0.334

* Significant at 1% level

Table 2. Estimates of the general combining ability effects for seven silk yield attributes in *Bombyx mori* L.

Parents	Silk yield attributes						
	ERR	cocoon yield	cocoon weight	shell weight	raw silk recovery	silk thread length/ cocoon	silk reelability
N4	–3.581	0.612	0.021	0.008*	–0.342	9.4	1.092
SPJ1	1.257*	–0.406	–0.002	–0.007	–0.510	20.4**	–0.457
JC2P	2.740**	–0.144	0.003	0.012**	0.538**	15.3*	0.406
14M	–0.133	0.018	–0.004	–0.005	0.277	–45.9	1.095
SH2	–0.283	–0.080	–0.017	0.007	0.037	0.8	–2.134
S.E. (g_i)	0.576	0.297	0.011	0.003	0.386	6.770	1.209
C.D.$_{0.05}$ (g_i)	1.141	0.588	0.021	0.006	0.441	13.399	1.380
C.D.$_{0.01}$ (g_i)	1.514	0.780	0.028	0.008	0.585	17.773	1.832
S.E.$_{0.05}$ ($g_i - g_j$)	0.912	0.470	0.017	0.005	0.347	10.705	1.021
C.D.$_{0.05}$ ($g_i - g_j$)	1.804	0.931	0.033	0.010	0.396	33.742	1.165

* Significant at 5% level; ** Significant at 1% level.

of general combining ability variance (σ_g^2) and specific combining ability variance (σ_s^2) the ratio $2\sigma_g^2/(2\sigma_g^2+\sigma_s^2)$ was worked out as described by BAKER (1978) for all seven silk yield attributes. None of the attributes except ERR (0.963) showed near unity the above ratio.

General combining ability effects

Estimates of the general combining ability (GCA) effects of five parents for all seven silk yield attributes are presented in Table 2. The parental strain JC2P had the best general combining ability for all silk yield attributes, except the cocoon yield. The strain N4 showed significant positive GCA effects for all silk yield attributes except the ERR and raw silk percentage.

Specific combining ability effects

Estimates of the specific combining ability (SCA) effects for 10 F_1's are presented in Table 3. Positive SCA effects were expressed by six out of ten crosses, namely N4 × SH2 (all silk yield attributes except shell weight), 14M

Table 3. Estimates of the specific combining ability effects for seven silk yield attributes in *Bombyx mori* L.

Hybrids	Silk yield attributes						
	ERR	cocoon yield	cocoon weight	shell weight	raw silk percentage	silk filament length/ cocoon	silk reela-bility
N4 × SPJ1	10.75	2.282	0.037	–0.007	–0.625	122.1*	1.260
N4 × JC2P	–2.18	–1.097	0.106	0.045*	1.436	70.2*	4.662
N4 × 14M	–8.87	–2.210	0.012	–0.001	–1.952	–78.6	–2.056
N4 × SH2	6.66*	0.841	0.072	–0.006	0.963	13.7	0.607
SPJ1 × JC2P	–13.86	–0.492	0.005	–0.008	0.119	–55.3	–5.529
SPJ1 × 14M	–3.20	0.495	–0.007	0.011	0.226	11.9	–4.023
SPJ1 × SH2	–5.68	–2.461	–0.004	0.003	0.436	–52.3	6.266*
JC2P × 14M	0.74	0.003	–0.101	–0.013	1.522	8.0	2.899
JC2P × SH2	4.90	0.999	–0.079	–0.026	–1.463	–26.7	–1.052
14M × SH2	–2.08	0.385	0.053	0.009	1.244	99.5	1.589
C.D.$_{0.05}$(s_{ij})	2.35	1.213	0.044	0.013	0.895	27.6	2.634
C.D.$_{0.05}$ ($s_{ii} - s_{ji}$)	5.05	2.606	0.094	0.028	1.923	59.3	5.648

* Significant at 5% level

× SH2 (all the attributes except ERR), N4 × SPJ1 (all the attributes except shell weight and raw silk percentage), JC2P × 14M (all the attributes except cocoon weight and shell weight); N4 × JC2P (all the attributes except ERR and cocoon yield), SPJ21 × 14M (all the attributes except ERR, cocoon weight and silk reelability).

Reciprocal effects

Only two reciprocals showed positive effects for most of silk yield attributes. These are SPJ1 × N4 (all the attributes except silk reelability), and SH2 × 14M (all the attributes except ERR and silk reelability).

Discussion

The main purpose of this study was to identify superior silkworm strains which could be utilized as parents in hybridization programme. The choice of parents for hybridization is based on the performance of parents per se, the performance of F_1's and combining ability effects. The present study showed that the per se performance of parents was not an indication of their combining ability. Similar results were obtained by PETKOV (1975) and PETKOV et al. (1984) on the combining ability of some Bulgarian and Ukrainian silkworm breeds. As evident from the obtained results, on the one hand, the parental strains (SPJ1, 14M and SH2) were negative for most of the attributes and, on the other, 6 out of 10 F_1 hybrids (N4×SH2; 14M×SH2; N4×SPJ1, JC2P×14M, 4N×JC2P and SPJ1×14M) showed positive SCA effects. Hence, the prospects of exploiting heterosis in their above listed crosses is possible. A comparison of GCA effect of parental strains indicated the superiority of JC2P for most of the silk yield attributes except cocoon yield.

The data for most of the silk yield attributes, viz. cocoon weight, shell weight, raw silk percentage, silk filament length/cocoon and silk reelability indicated that the non-additive genetic effects were larger than additive genetic effects. Also in contrast to the earlier reports (KRISHNASWAMI et al. 1964, SENGUPTA et al. 1974, BHARGAVA et al. 1992) the above results indicate predominance of additive gene action for expression of ERR and cocoon yield as seen from the higher GCA than SCA for these two attributes. However, the role of non-additive gene action cannot be totally ruled out for the cocoon yield as the ratio obtained by BAKER'S (1978) formula was less than unity for the cocoon yield and other silk yield attributes except ERR indicating non-additive gene action. Therefore, a simple selection programme which may involve infusion of desirable genes in the population like mass selection followed by intermating as suggested by REDDEN and JENSEN (1974) may prove to be useful in improving cocoon yield.

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