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Effects of selection on cocoon filament length in divergently selected lines of the silkworm, *Bombyx mori*

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To examine the effects of selection and the degree of manifestation of heterosis, we compared the polyvoltine populations of *Bombyx mori* selected for long or short cocoon filament lengths for 11 generations, and performed their *inter se* matings and crosses of selected lines with unrelated bivoltine tester populations. We measured the selected trait, viz. the filament length, as well as other unselected traits such as survival rate, single cocoon weight, cocoon shell weight, cocoon shell ratio and filament size. The genetic improvement brought about for filament length by virtue of selection was found to be reflected in the hybrids. On an average, genes for short filament length were found to be dominant over those of long filament length. Significant difference in reciprocal crosses was observed for the selected trait in the interline matings. Matings of the lines with long or short filament lengths with the unrelated tester population reflected the filament length of the selected population. Among the unselected traits, heterosis was more extensive for the survival rate followed by the single cocoon weight and cocoon filament size in both interline matings and the crosses of the selected lines with unrelated tester population.

Key words: cocoon filament length, heterosis, selection of lines, *Bombyx mori*

A high level of heterosis for various quantitative characters is found to occur in the crosses of highly inbred lines and geographically divergent populations of the silkworm, *Bombyx mori* (HARADA, 1957, 1961; KRISHNASWAMI *et al.*, 1964; HIROBE, 1968; YOKOYAMA, 1979; GAMO and HIRABAYASHI 1983; NAGARAJU, 1990). It has also been demonstrated in various plants and animal species that the lines derived from the same population with different selection histories are known to reveal heterosis when they are crossed (FALCONER, 1986). Practical demonstrations have also come from poultry for selection of egg weight and body weight (HAGGER, 1985; DUNNINGTON *et al.*, 1990), mouse for body weight (BHUVAN KUMAR *et al.*, 1985) and *Drosophila melanogaster* for bristle number (ROBERTSON, 1960). In *B. mori* selection for various quantitative characters results in change in their mean to a varying degree (KURASAWA, 1968; GAMO and ICHIBA,

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1971; MIYAHARA, 1978). The selection for one character is found to produce correlated changes in the other quantitative characters of economic importance (KOBARI and FUJIMOTO, 1966; MIYAHARA, 1978). In this connection of interest is the problem as to how the amount of heterosis for the selected and unselected traits is manifested depending upon the selection history of population.

In a polyvoltine base population (non-hibernating strain that breeds throughout the year) divergent selection exercised for long and short filament lengths for 11 generations resulted in two populations with respective mean filament lengths of 876±11 m and 704±7 m (NAGARAJU, 1990). The correlated changes in size and weight of the silk filament and weight of the cocoon also occurred. For example, the line selected for longer cocoon filament registered longer filament with a correlated decrease in its size while opposite was the case in the line selected for shorter cocoon filament.

The experimental population derived from the selection experiment thus provides an excellent genetic background for studying the effects of artificial selection on the genetic differentiation of the selected populations of this economic insect. Hence the present investigation was carried out to: (a) assess the level of heterosis in the interline crosses and their reciprocals and (b) assess the degree of heterosis in the crosses of the selected lines with unselected tester line.

Materials and Methods

The lines "LFL" and "SFL" selected for 11 generations for long and short filament lengths, respectively, were used in the present study. These lines were derived from a common polyvoltine silkworm populations (NAGARAJU, 1990). The common base population was synthesized by crossing 4 highly inbred polyvoltine lines, viz. MW1, MW2, Kollegal jawan and MHMP(W) (NAGARAJU *et al.*, 1988). As an unrelated tester line, NB_{18}, a highly inbred line that has passed 80 generations since bred from a Japanese double cross, (Kokko × Seihako) × (N124 × C124), was used (KRISHNASWAMI, 1978). This particular line was chosen for the present investigation since it is known to produce highly heterotic hybrids when used as a male parent to cross with the females of polyvoltine strains by providing a new genetic background (NAGARAJU *et al.*, 1987). Since the reciprocal crosses polyvoltine male × bivoltine female are known to show striking reciprocal effects due to sex linked early maturing genes (TAZIMA, 1988; MURAKAMI, 1989), they were avoided in the present study.

From each of the parental populations, LFL and SFL, 10 males and 10 females were randomly picked and their reciprocal crosses (LFL × SFL and SFL × LFL) were prepared. Ten females each from LFL and SFL lines were crossed to 10 males of the bivoltine inbred line NB_{18}. Mating period of 3 hr was allowed, followed by 24 hr duration for oviposition. At least 50 eggs from each of the egg layings obtained from the 10 pair matings were pooled to generate 3 replicates in each cross. Similarly, 3 replicates from each of the parental lines, LFL and SFL, were raised from 10 pair matings. The interline crosses, their reciprocal crosses, line × tester crosses and their parental lines were reared simultaneously. Data were collected for the survival rate (live pupae recovered from the total number of larvae retained in each replicate). Cocoon characters such as single cocoon weight, cocoon shell weight and cocoon shell ratio were recorded in parallel with cocoon fiber characters such as cocoon filament length and size.

Heterosis for each character was measured as percentage of deviation of the hybrid mean from the parental mean. Analysis of variance was carried out to elicit differences between parental populations, between hybrid populations, between parental and hybrid populations and between direct and reciprocal interline matings. The statistical contrasts (Table 1) among populations were analyzed by the method according to DUNNINGTON *et al.*, (1990).

Results

The short-long difference for the selected trait accounted for 172 m (Table 2). The significant ($p<0.01$) difference between the two parental populations was found for the se-

Table 1. Contrasts analysed among poulations.

1. Parents	
Comparison of LFL line with SFL line	LFL vs. SFL
Comparison of Parents with interline matings	LFL vs. SFL×LFL and LFL × SFL SFL vs. SFL×LFL and LFL × SFL
2. Interline matings	
Heterosis	$\frac{LFL+SFL}{2}$ vs. LFL×SFL and SFL×LFL
Reciprocal effects	LFL×SFL vs. LFL×SFL
3. Crosses with tester line ♂ (T)	
Comparison of LFL and SFL ♀ when mated to T	LFL× T vs. SFL×T
Comparison of LFL × T cross with SFL line	LFL vs. LFL×T
Comparison of SFL × T cross with SFL line	SFL vs. SFL×T

LFL, line selected for longer filament length; SFL, line selected for shorter filament length; T, tester line (NB_{18}).

Table 2. Heterosis for various characters between divergently selected lines.

Characters	Lines		Interline matings		Heterosis %	
	LFL	SFL	LFL×SFL	SFL×LFL	LFL×SFL	SFL×LFL
Filament length (m)	876	704	783	717	−0.89	−9.2
Cocoon filament size (denier)	2.00	2.85	2.19	2.2	3.29	3.76
Single cocoon weight (g)	1.37	1.34	1.42	1.35	4.17	0.80
Single shell weight (g)	0.21	0.20	0.23	0.22	20.70	4.83
Cocoon shell ratio (%)	15.63	14.79	16.50	16.08	8.48	5.72
Survival rate (%)	56.25	68.06	73.12	74.97	17.64	20.06

lected trait, cocoon filament length (Table 3). For the other traits, the line SFL had higher mean for the cocoon filament size and survival rate, while the LFL line showed favorable mean for the single cocoon weight and cocoon shell weight, although differences were not significant.

The striking effect of differences in length of filament accrued in the parental line was seen in the hybrids of interline matings. Values of heterosis expressed as percentages of midparental values are presented in Table 3. The hybrid mean in both direct and reciprocal crosses fell short of the midparental values. Results indicate that the heterosis found here is evidence for directional dominance of alleles at loci involved in determining the character under study. Alleles from the SFL line were on an average dominant for the filament length and size while those from the LFL line were dominant for the cocoon shell weight and single cocoon weight. The hybrids of LFL and SFL and their reciprocals were not significantly different from each other for any trait other than the selected one, the filament length (Table 3).

The hybrids of LFL and SFL dams with tester (NB_{18}) sires showed interesting results

Table 3. Extract of analysis of variance for the characters measured in parental lines and their interline matings.

Source	df	Mean sum of squares				
		Survival rate	Single cocoon wt.	Single shell wt.	Shell ratio	Filament length
Parents	1	210.0417	0.0014	0.0004	3.5267	44204.17**
Hybrids	1	4.86	0.0067	0.0003	5.2267	6600.17**
Parents × hybrids	1	423.6408*	0.0016	0.0011*	3.6299	4880.34**
Error	36	42.18	0.0033	0.0002	0.9242	359.00

** and * indicate $P<0.01$ and $P<0.05$, respectively.

Table 4. Heterosis for various characters between divergently selected lines and tester.

Characters	Lines			Crosses		Heterosis (%)	
	LFL	SFL	Tester	LFL× Tester	SFL× Tester	LFL× Tester	SFL× Tester
Filament length (m)	876	704	870	1,000	883	14.55	12.20
Cocoon filament size (denier)	2.00	2.85	2.60	2.68	2.80	16.24	15.67
Single cocoon weight (g)	1.37	1.34	1.76	1.72	1.68	12.93	11.48
Single shell weight (g)	0.21	0.20	0.35	0.35	0.32	25.71	17.61
Cocoon shell ratio (%)	15.63	14.79	20.86	20.48	19.05	12.83	7.44
Survival rate (%)	56.25	68.06	47.50	66.40	72.08	23.81	25.14

(Table 4). The hybrid of LFL × tester recorded a higher mean with a difference of 117 m as compared to that of SFL × tester. Difference between the two hybrids was highly significant ($p<0.01$) for this trait (Table 5). For other traits, heavier single cocoon and cocoon shell weights and thinner filament size of the LFL line and lighter single cocoon and cocoon shell weights of the SFL line were evident in their respective hybrids, although analysis of variance did not reveal any significant differences. Both the hybrids revealed significant heterotic effects for all the traits over their parental strains.

Discussion

The striking effect on the length of filament in interline matings and crosses of the selected lines with an unrelated tester parent is consistent with the differences observed in the parental stocks with different selection histories, where the SFL line secreted shorter and thicker cocoon filament and formed smaller cocoon while the opposite was the case in the LFL line. The results show that genetically distinct populations with different mean values could be obtained in the silkworm by virtue of selection for an important economic trait like the length of cocoon filament. Genetic differences observed in the lines selected divergently from the same base population and heterotic effects for the selected and unselected traits corroborate the theoretical contention (FALCONER, 1986) that sub-division of base population through inbreeding results in lines with different genic values. The directional dominance for the short silk filament length probably reflects a history of past directional selection towards increasing fitness as contended by ROBERTSON (1955). The results are not surprising considering the fact that the polyvoltine silkworms from which the lines are derived were adopted towards secreting shorter filament. In an evolutionary context, almost all the polyvoltine strains distributed in the tropical countries are endowed with shorter and thicker filament and smaller cocoon, thus revealing positive correlation between voltinism and other quantitative traits (TAZIMA, 1988; MURAKAMI, 1989).

The significant difference between direct and reciprocal interline crosses indicates the involvement of sex linked genes which are known to cause differences in quantitative traits in reciprocal crosses in the silkworm (NAGATOMO, 1942; MOROHOSHI, 1949; MURAKAMI and OHTSUKI, 1989). Relative to midparent values both interline matings and the hybrids of selected lines and unrelated tester parent revealed significant levels of heterosis for the

Table 5. Extract of analysis of variance measured in LFL and SFL lines and their crosses with an unrelated tester line.

Source	df	Mean sum of squares				
		Survival rate	Single cocoon wt.	Single shell wt.	Shell ratio	Filament length
Parents	2	213.7817	0.0637 * *	0.0132 * *	21.1850 * *	19005.50 * *
Hybrids	1	39.0625	0.0018	0.0011	2.1025	13572.25 * *
Parents × hybrids	1	359.6602 *	0.1369 * *	0.00116 *	18.4815 *	37350.15 * *
Error	45	44.946	0.0017	0.0003	1.3260	413.10

* * and * indicate $P<0.01$ and $P<0.05$, respectively.

survival rate. These results corroborate earlier findings (HARADA, 1961; YOKOYAMA, 1979; GAMO and HIRABAYASHI, 1983). It is expected that the low heritability trait such as survival rate (GAMO and HIRABAYASHI, 1983) displays extensive heterosis and has become focal trait for exploitation of heterosis in the silkworm.

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JAVAREGOWADA NAGARAJU・TUMULURI PAVAN KUMAR: カイコにおける繭糸長の選抜効果

多化性蚕について繭糸長を長短の両方向に11世代にわたって選抜し，選抜した系統間での交配並びに検定用の２化性系統との交配によって選抜効果とヘテローシスの発現を追究した。 選抜した繭糸長および無選抜の生存率，繭重，繭層重，繭層歩合，繭糸繊度をそくていして定量化したところ，繭糸長の選抜によってもたらされる遺伝的な改良は交雑種に反映することが明らかとなった。短糸長の性質は長糸長に対して優性であった。選抜した系統間の相互交配では繭糸長が有意に相違した。長糸長と短糸長の系統を検定用２化性系統に交配すると，選抜集団の繭糸長が反映された。また，選抜した系統間および２化性の検定系統との交配において無選抜の形質についてヘテローシスを調べた結果，生存率で最も大きく現れ，ついで繭重，繭糸繊度で認められた。