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Genetic Improvement of Some Traits in Four Strains of Silkworm, *Bombyx mori* L.

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A breeding plan was carried out on four commercial strains of silkworm (*Bombyx mori* L.) 101, 102, 103 and 104 to improve some important traits. Genetic gain or response to selection (ΔG), heritability of cocoon shell weight (CSW) and specific combining ability effects were estimated to determine the strains that can be improved. Strain 101 had lowest heritabitity, ΔG and viability. Strain 102 was acceptable in selection response but its viability was low. Therefore these two strains were not suitable for more selection. As a result, only lines 103 and 104 were chosen for further improvement. Intra population selection based on independent culling level method practiced from third to sixth generation for both productive and viability traits simultaneously. While CSW and CW had increasingly enhanced during primary generations, they went slightly up after third generation. According to negative genetic correlation, viability decreased during primary generations, but after third generation that paid attention to balanced development of both productive and viability traits, viability increased so that the pupation rate reached to 91% in 103 and 97% in 104 for last generation (G_8).

Key words: Selection, Heritability, Independent culling levels, Outbreeding, Genetic improvement

Introduction

In some countries new commercial silkworm varieties and hybrids were continuously produced and replaced with previous varieties and hybrids. In the past half century in china, almost every ten years, Chinese silkworm varieties have been upgraded (Anying *et al.*, 2004; Kang *et al.*, 2004). The progress of India, the second largest silk producer in the world, is largely due to the introduction of several bivoltine strains to Indian sericulture (Raju and Krishnamurthy, 1993). On the other hand, some countries like Iran can not evolve new breeds continuously because of limited genetic resources. Therefore they must employ other methods of breeding such as selection methods in order to genetic improvement.

Cocoon shell weight (CSW), cocoon weight (CW) and shell percentage (SP) as important traits of silkworm have high heritability and are affected by additive gene action, therefore they respond better to selection than those that have low heritability (Petkov and Nguyenvan, 1987). Heritability (h^2) estimates of CSW is medium to high (0.2 – 0.7) and has positive genetic correlation with some traits such as cocoon weight, cocoon shell percentage, larval weight, filament length, raw silk percentage and egg weight (Sohn *et al.*, 1987; Singh *et al.*, 1998; Jayaswal *et al.*, 2000).

Improving cocoon and silk filament characters is the most important breeding goal. The selection of one character is found to result in correlated changes on other economic characters. As productive traits and viability are negatively correlated, viability must be considered in selection program (Singh *et al.*, 1998; Sekharappa *et al.*, 1999; Reddy *et al.*, 2004). In silkworm breeding, independent culling levels can be used as a multiple trait selection (Hosseini Moghaddam *et al.*, 2002; Greiss *et al.*, 2004).

This study was conducted to evaluate possibility of improving economic traits of four strains of silkworm and then improving of desirable traits.

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Materials and Methods

Four strains of silkworm including two Japanese (101,103) and two Chinese (102,104) strains were studied. In G_0 generation 200 cocoons from each strains were randomly sampled. Forty-eight disease free layings were sampled from the harvested cocoons of each strain and half of each allocated into selection (S) and control (C) lines. Therefore each selection and control lines consisted of 24 families (batches).

Tandem selection method practiced in each selection line in the first step of program(Hallavar *et al.*, 1988; Hosseini Moghaddam *et al.*, 2002). Ten percent (selection pressure) of cocoons of each sex (female, male) with the heaviest CSW were selected (selected cocoons) and their moths were mated randomly. In other words, mating program was based on outbreeding, therefore four outbred populations was established in G_1.

In control lines no selection was carried out on cocoon characters and their moths were mated randomly. Data collected from three generations in control lines were used for estimation of h^2 of CSW by variance components of full-sib data of single pair mating (Becker, 1984; Chapman, 1985). h^2 of CSW was estimated using Harvery software (1990). In addition Realized heritability of CSW was estimated by method of Pirchner (1983). Response to selection (ΔG) estimated from different means of selected and control lines (Becker, 1984).

Also specific combining ability of these strains estimated by diallel crosses method. This method gives the required parameters for explanation of genetic backgrounds and choosing the best parent for hybridization (Sohn *et al.*, 1987; Hosseini Moghaddam *et al.*, 2000). The Griffing's (1956) approach of diallel analysis is the most reliable and precise methods to get information about combining ability.

According to genetic parameters (h^2, ΔG and combining ability) only 103 and 104 strains chose for further improvement on the second step.

Intra population selection was practiced from G_2 to G_6 for productive (CSW and CW) and viability (larval survival and pupation rate) traits simultaneously. The uniformity of hatching and silkworm growth was considered in selection program and families with uncoordinated growth were rejected. Four families (batches) with better viability than others were retained and then cocoons with shell weight and cocoon weight over the average level were selected for reproducing. This procedure is the same as independent culling levels that is a method of multiple traits selection (Chapman, 1985; Hallauar and Miranda, 1988). In G_7 generation selection for cocoon characters wasn't done to evaluate performance of two strains. Rearing technology and recording methods were conducted according to standard method (ESCAP, 1993; Hosseini Moghaddam, 2004).

Results

Table 1 presents the mean of CSW and CW in control and

Table 1. Mean of cocoon shell weight, cocoon weight and response to selection (ΔG) in G_1 and G_2 generations

Strains	CSW (mg)				CW (g)				ΔG (mg)	
	G_1		G_2		G_1		G_2		G_1	G_2
	C	S	C	S	C	S	C	S		
101	375.0	379.4	404.3	416.0	1.59	1.61	1.69	1.70	4.4	11.7
102	312.1	322.8	345.2	376.7	1.41	1.49	1.49	1.59	10.7	31.5
103	378.8	392.5	401.0	426.1	1.63	1.69	1.69	1.75	137	25.1
104	360.2	374.2	380.8	407.8	1.60	1.66	1.64	1.79	14	27.0

C = Control line, S = Selection line.

Table 2. Estimates of heritability (h^2) and realized heritability (h^2_R)

Strains	h^2 (SE)				h^2_R	
	Female	n	Male	n	Female	Male
101	0.280(0.069)	869	0.276(0.067)	952	0.175	0.154
102	0.502(0.088)	1201	0.465(0.084)	1200	0.540	0.390
103	0.470(0.085)	1201	0.394(0.077)	1222	0.281	0.287
104	0.413(0.078)	1235	0.367(0.073)	1250	0.340	0.399

n: number of observations.

Table 3. Pupation rate and larval mortality in G_1 and G_2 generations

Strains	Pupation rate (%)				Larval mortality (%)			
	G_1		G_2		G_1		G_2	
	C	S	C	S	C	S	C	S
101	78	74	70	65	2.6	5	4.1	5.6
102	84	83	85	84	1.9	1.8	1.9	1.9
103	81	79	75	69	3.5	6	2.1	2.8
104	95	92	91	87	2	2.2	1.6	4.6

C = Control line, S = Selection line.

selection lines and also response to selection (genetic gain). Total genetic gain was 16.1 mg, 42.2 mg, 38.1 mg and 41 mg for 101, 102, 103 and 104 strains respectively. The h^2 of CSW is presented in Table 2 by two different methods of estimations. It is clear from Tables 1, 2 and 3 that in comparison with others, in strain 101 h^2, ΔG and viability were very low. Strains 102 had sufficient variation for selection but viability was low as compared to other Chinese strain (104). Also the uniformity of larval growth was not acceptable. Therefore 101 and 102 strains had not competence for further improvement in this program. On the other hand, h^2 and ΔG of strains 103 and 104 showed that selection could be done in subsequent generations.

Estimated values of Special Combining Ability effect (SCA) are presented in Table 4. Comparison of SCA related to crosses between Japanese strains with Chinese strains showed that 103 × 104 had high positive significant SCA for both SW and CW. Also comparison of heterosis determined that the 103 × 104 hybrid had the most heterosis for productive traits (Hosseini Moghaddam, 2000). Therefore 103 and 104 strains were the best parents for hybridization. As a result these two lines were selected for more improvement on the second step.

Table 4. Estimates of SCA and heterosis (HET) in different crosses of silkworm

Crosses	SW		CW	
	SCA	HET	SCA	HET
J101 × j101	-0.040	-------	-0.189	-----
J101 × j102	0.033	0.052	0.167	0.28
J101 × j103	-0.014	0.016	-0.078	0.06
J101 × j104	0.020	0.044	0.10	0.23
J102 × c102	-0.043	------	-0.180	-----
J102 × c103	0.028	0.065	0.118	0.28
J102 × c104	0.019	0.014	-0.105	0.02
J103 × c103	-0.049	-------	-0.190	-----
J103 × c104	0.035	0.077	0.150	0.30
J104 × c104	0.037	------	-0.145	------

In the next generations emphasis was laid on balanced development of all economic characters by multiple traits selection. The mean values of selection lines and selected cocoons for CSW is given in Table 5 from zero (G_0) to eight (G_8) generation. Selection for CSW in the primary generations increased this trait and other traits correlated to it. In later generations, equal emphasis was given to all traits. Selection for productive and viability traits and also uniformity of growth, modify the improvement of all traits. As showed in Table 5 and 6, moderate improvement was obtained for CSW and CW after third generation both in spring rearing season and autumn rearing season. Also pupation rate increased gradually, because of selecting families with less viability (larval mortality and pupation rate). Pupation rate that decreased in primary generations, increased gradually so that it was 91% and 97% for 103 and 104 respectively in the last generation.

Discussion

In the last decades several new silkworm bivoltine breeds were evolved as parents of hybrids for production of silkworm egg. Sekharappa *et al.* (1999) evolved superior bivoltine races having superior cocoon shell percentage and better survival. Raju and Krishnamurthy (1993) programmed a breeding plan for isolating two bivoltine breeds with higher viability and silk productivity. Reddy *et al.* (2004) attempted to evolve productive bivoltine breeds for commercial exploitation. Selection of segregated traits and inbreeding are the specifications of this systematic breeding approach in silkworm (Haque, 1990; Raju and Krishnamurthy, 1993; Kang *et al.*, 2004; Zhao *et al.*, 2004).

Because of inbreeding difficulties such as inbreeding depression, mating plan at present study was outbreding. Petkov (1989) and Hosseini Moghaddam *et al.* (2002) reported about outbred populations in silkworm. Results showed outbred lines do not have difficulties of inbred

Table 5. Mean of CSW for selection lines and selected individuals (cocoons) by sex and strains in G0-G8

Year	Rearing season	Generation	Sex	101		102		103		104	
				Selection line	Selected cocoons	Selection line	Selected cocoons	Selection line	Selected cocoons	Selection line	Selected cocoons
1995	Autumn	G_0	F[a]	0.325	0.373	0.296	0.339	0.319	0.389	0.311	0.363
			M[a]	0.312	0.361	0.274	0.320	0.315	0.377	0.294	0.344
1996	Spring	G_1	F	0.381	0.428	0.330	0.376	0.397	0.457	0.380	0.444
			M	0.378	0.412	0.316	0.367	0.388	0.443	0.369	0.427
	Autumn	G_2	F	0.428	0.451	0.392	0.411	0.435	0.469	0.422	0.455
			M	0.403	0.424	0.361	0.378	0.417	0.457	0.392	0.426
1997	Spring	G_3	F	0.458	-	0.445	-	0.484	0.511	0.475	0.507
			M	0.411	-	0.417	-	0.461	0.492	0.446	0.479
	Autumn	G_4	F	-	-	-	-	0.341	0.370	0.330	0.355
			M	-	-	-	-	0.341	0.362	0.320	0.348
1998	Spring	G_5	F	-	-	-	-	0.435	0.463	0.416	0.436
			M	-	-	-	-	0.409	0.433	0.391	0.412
1999	Autumn	G_6	F	-	-	-	-	0.446	0.471	0.385	0.411
			M	-	-	-	-	0.456	0.461	0.369	0.392
2000	Spring	G_7	F	-	-	-	-	0.494	-	0.407	-
			F	-	-	-	-	0.458	-	0.381	-
2001	Spring	G_8	F	-	-	-	-	0.502	-	0.411	-
			M					0.470	-	0.399	-

[a], F = female, M= male.
*selection was not done in G7.

Table 6. Mean of cocoon weight and pupation rate of 103 and 104 strains in selection line

Year	Rearing season	Generation	Sex	Cocoon weight (gr)		Pupation rate (%)*	
				103	104	103	104
1995	Autumn	G0	F[a]	1.72	1.72	78	93
			M[a]	1.45	1.37		
1996	Spring	G1	F	1.85	1.81	79	92
			M	1.54	1.51		
	Autumn	G2	F	1.93	2	69	87
			M	1.56	1.57		
1997	Spring	G3	F	2.25	2.2	75	88
			M	1.78	1.76		
	Autumn	G4	F	1.67	1.62	73	85
			M	1.37	1.32		
1998	Spring	G5	F	2.06	1.88	80	92
			M	1.6	1.48		
1999	Autumn	G6	F	2.18	1.94	87	94
			M	1.81	1.55		
2000	Spring	G7	F	2.23	2.01	89	96
			M	1.76	1.62		
2001	Spring	G8	F	2.2	2.05	91	97
			M	1.8	1.65		

[a], F = Female, M = Male.
*, Amount of pupation rate is batch average.

lines and can be introduced as improved lines.

Usually the method of selection in silkworm breeding is independent culling levels, but sometime other methods were utilized such as tandem and selection index methods (Escap, 1993; Hosseini Moghaddam *et al*., 2002; Greiss *et al*., 2004). While Selection index required heritability values, independent culling levels required experience of breeder. Greiss *et al.* (2004) showed independent culling is able to ensure almost the same average index value of the best-selected individuals and by using equal selection pressure, it does not need heritability estimates. Greiss *et al.* (2004) studied difference of results of selection based on breeder experience and computer. They found no significance in the average selection index between breeders and computer.

The selection of 103 and 104 strains was according to genetic parameters by statistical analysis while simple calculations and experience of breeders was used for independent culling level on the second step. For improving the traits of 101 & 102, the outcrossing with exotic races or repeated backcrossing was suggested (Raju and Krishnamurthy, 1993; Hosseini Moghaddam *et al*., 2002). At present, the improved strains with the name of S103 and S104 were used by Iran Sericulture Research Center.

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