

Identification of Breeding Resource Material for the Development of Thermo-Tolerant Breeds in the silkworm, *Bombyx mori*

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(Received 7 March 2001; Accepted 20 April 2001)

Screening of fifteen bivoltine silkworm breeds of *Bombyx mori* Linn at a temperature of $31 \pm 1^\circ\text{C}$ and relative humidity of $85 \pm 5\%$ resulted in the identification of eight thermo-tolerant breeds. The survival and cocoon shell ratio of the tolerant breeds ranged from 72.7 to 78.7% and 20.0 to 20.1% respectively. The tolerant breeds comprised of four oval breeds and four dumb-bell breeds. Eight foundation crosses prepared by crossing the oval and dumb-bell parents among themselves were screened at a temperature of $31 \pm 1^\circ\text{C}$ and relative humidity of $85 \pm 5\%$. The performance of the foundation crosses on 11 economic characters were analysed by employing Multiple Trait Evaluation Index method. Four foundation crosses which scored average index value > 50 were selected as breeding parents and breeding initiated for the evolution of thermo-tolerant bivoltine silkworm breeds. The methodology and the results of the foundation crosses reared both at $31 \pm 1^\circ\text{C}$ and at $25 \pm 1^\circ\text{C}$ temperatures, are discussed.

Key words : Thermo-tolerant, Bivoltine silkworm breeds, Multiple trait evaluation index, silkworm breeding, *Bombyx mori*

Introduction

Commercial exploitation of multivoltine \times bivoltine silkworm *Bombyx mori* Linn., in India led to quantitative increase in the silk yield compared to the pure races or their hybrids. The multivoltine \times bivoltine silk no doubt

meets the domestic demand but the silk quality is poor and also not internationally recognised. High quality, internationally recognised silk can be achieved only by rearing univoltine/bivoltine races. But bivoltine silkworm rearing is practised only by 5 to 10% farmers in India. Due to temperature fluctuations, poor management conditions and poor quality mulberry leaves, bivoltine crop losses are often witnessed with farmers. Hence, there is a need to evolve bivoltine silkworm breeds for the tropical climate in India. A similar climatic condition exists in the tropical part of China, and they have succeeded in evolving bivoltine silkworm breeds suited to their tropical fluctuating climate. Therefore, in the present study an attempt has been made to develop thermo-tolerant bivoltine silkworm breeds of *B. mori* in India based on the Chinese methodology (Shao *et al.*, 1987).

Materials and Methods

Summer rearing performance of 150 silkworm breeds were short-listed based on their pupation rate and shell ratio. A total of forty silkworm breeds indicated survival rate of 80.0 to 96.0% and shell ratio of 18.0 to 24.30% (Table 1). These were further short-listed by using Multiple trait Evaluation Index method (E.I.) (Mano *et al.*, 1993). In selection of a breed, it is necessary to give importance to all the economically important characters contributing to productivity. E.I method is utilised as a tool by giving importance to all the traits of economic importance. Fifteen silkworm breeds showed an average index value of 50 and more than 50 index value in the four economic traits studied. These 15 breeds comprised of seven oval breeds and eight dumb-bell breeds. They are pure exotic breeds *viz.*, C124, Tokai, Dong 306, C3, A and evolved breeds *viz.*, CDC2 and CCS. These have plain larval body which build oval cocoons. The dumb-bell breeds

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Table 1. Rearing performance of the silkworm stock breeds

(Average of 3 years)

Sl. No	Breed	Survival No.	Cocoon Wt. (g)	Shell Wt. (g)	Shell Ratio %	Evaluation Index Value				Avg E.I
1	Tokai*	9600	1.392	0.321	23.1	68	37	52	70	57
2	935 E*	8840	1.651	0.322	19.5	52	55	52	46	51
3	16 A	8533	1.805	0.342	18.7	45	66	59	41	53
4	916A1*	8823	1.903	0.379	19.9	52	73	73	49	62
5	JA2	8823	1.803	0.333	18.5	46	66	56	39	52
6	44FM*	9449	1.688	0.357	21.1	72	58	65	57	63
7	CPP1	8000	1.548	0.282	18.2	34	48	38	38	40
8	Hu204	8333	1.649	0.306	18.6	41	55	46	40	46
9	KPGA	9007	1.526	0.295	19.3	56	46	43	45	48
10	CC1SL	8250	1.531	0.334	21.8	39	47	56	62	51
11	NN6D	8333	1.470	0.269	18.3	41	42	33	39	39
12	CAC	8366	1.553	0.289	18.6	42	50	40	40	43
13	KDC	8466	1.766	0.332	18.8	44	63	56	42	51
14	DD3*	9100	1.739	0.327	18.8	58	61	54	42	54
15	CDC2*	9433	1.920	0.362	18.9	65	74	66	42	62
16	ND	8400	1.490	0.282	18.9	43	44	38	42	42
17	36 PC	8625	1.500	0.302	20.2	47	44	45	51	47
18	N x N140	9000	1.503	0.278	18.5	55	46	36	39	44
19	KPGB	8978	1.562	0.309	19.8	55	49	48	48	50
20	C124*	9222	1.578	0.311	19.7	60	50	48	48	52
21	SHOWA	8022	1.481	0.304	20.5	35	43	46	53	44
22	644	8505	1.406	0.253	18.0	45	38	28	36	37
23	SH2	8626	1.598	0.332	20.8	47	51	56	55	52
24	SPC1	8126	1.680	0.331	19.7	37	57	55	48	49
25	NB2P	8108	1.485	0.306	20.6	36	43	46	54	45
26	912B2*	8962	1.590	0.324	20.8	55	49	53	55	53
27	CCS*	8999	1.501	0.308	20.5	55	50	47	53	51
28	NB4D2*	8773	1.586	0.322	20.3	51	50	52	52	51
29	BL	9416	1.394	0.254	18.2	64	37	28	38	42
30	Dong 306*	8967	1.565	0.313	20.0	55	49	49	50	51
31	P5	9233	1.543	0.296	19.2	60	47	43	44	49
32	44BM	9219	1.534	0.308	20.1	60	47	47	50	51
33	935 A*	9017	1.592	0.338	21.2	56	51	58	58	56
34	NJ1	8014	1.627	0.332	20.4	34	53	56	52	49
35	SPJ2	8564	1.661	0.355	21.4	46	56	64	59	56
36	M190	8332	1.596	0.342	21.4	41	51	59	59	53
37	M56	8149	1.769	0.357	20.1	37	63	65	51	54
38	A*	8800	1.311	0.311	24.3	51	31	48	78	52
39	C3*	8967	1.448	0.341	23.5	55	41	59	73	57
40	J2*	9430	1.320	0.295	22.3	71	32	43	65	53

viz., 916A1, DD3, 912B2, NB4D2, 935A have plain larval body where as 935 E, 44FM and J2 have marked larval body which build dumb-bell cocoons. These breeds have

been evolved at Central Sericultural Research & Training Institute, Mysore (Table 2).

Eggs from 10-15 dfls were pooled from each breed

Table 2. Rearing performance of the fifteen silkworm stock breeds

(Average of 3 years)

Sl. No	Breed	Survival No.	Cocoon Wt. (g)	Shell Wt. (g)	Shell Ratio %	Evaluation Index Value				Avg E.I
1	Tokai*	9600	1.392	0.321	23.1	68	37	52	70	57
2	935 E*	8840	1.651	0.322	19.5	52	55	52	46	51
3	916A1*	8823	1.903	0.379	19.9	52	73	73	49	62
4	44FM*	9449	1.688	0.357	21.1	72	58	65	57	63
5	DD3*	9100	1.739	0.327	18.8	58	61	54	42	54
6	CDC2*	9433	1.920	0.362	18.9	65	74	66	42	62
7	C124*	9222	1.578	0.311	19.7	60	50	48	48	52
8	912B2*	8962	1.590	0.324	20.8	55	49	53	55	53
9	CCS*	8999	1.501	0.308	20.5	55	50	47	53	51
10	NB4D2*	8773	1.586	0.322	20.3	51	50	52	52	51
11	Dong 306*	8967	1.565	0.313	20.0	55	49	49	50	51
12	935 A*	9017	1.592	0.338	21.2	56	51	58	58	56
13	A*	8800	1.311	0.311	24.3	51	31	48	78	52
14	C3*	8967	1.448	0.341	23.5	55	41	59	73	57
15	J2*	9430	1.320	0.295	22.3	71	32	43	65	53

and brushed in three replications at a temperature of $31 \pm 1^\circ\text{C}$ and relative humidity of $85 \pm 5\%$ for screening. After III moult, five replications of 100 larvae in each replication were retained and reared upto cocoon stage. After harvesting of cocoons the number of live pupae were recorded. The breed which showed the highest number of live cocoons were considered as the tolerant breed. Cocoons from all the five replications of tolerant breeds were pooled and layings were prepared. The performance of the breeds at $31 \pm 1^\circ\text{C}$ is shown in Tables 3 and 4.

The screening of the breeds was carried out for three cycles to study the consistency of tolerance at high temperature. From the repeated screening of fifteen breeds at a temperature of $31 \pm 1^\circ\text{C}$ and relative humidity of $85 \pm 5\%$ resulted in the identification of eight tolerant breeds. The tolerant breeds comprised of four oval and four dumb-bell breeds. These breeds were utilised and foundation crosses were prepared. The oval parents were crossed among themselves and four foundation crosses were prepared. Similarly the dumb-bell breeds were crossed among themselves and four foundation crosses

Table 3. Performance of the oval breeds at $31 \pm 1^\circ\text{C}$ and RH $85 \pm 5\%$

Sl. No	Breed	Survival No.	Cocoon Wt. (g)	Shell Wt. (g)	Shell Ratio (%)	Total larval period (hrs)	V age period (hrs)
1	A	7500 (8934)	1.376 (1.536)	0.268 (0.321)	19.50 (20.89)	20.00 (21.02)	5.00 (5.22)
2	Tokai	7266 (9231)	1.330 (1.587)	0.263 (0.336)	19.7 (21.2)	20.08 (22.00)	5.14 (6.04)
3	C 124	8000 (9066)	1.331 (1.515)	0.262 (0.334)	19.7 (22.00)	20.08 (21.10)	5.14 (6.04)
4	C3	7866 (9105)	1.434 (1.594)	0.284 (0.344)	19.8 (21.6)	20.08 (22.00)	4.22 (6.00)
5	CCS	2933 (8979)	0.936 (1.436)	0.180 (0.286)	19.20 (19.9)	20.10 (21.08)	5.14 (6.00)
6	CDC2	3217 (8729)	0.879 (1.798)	0.166 (0.389)	18.9 (21.65)	20.08 (21.06)	5.14 (6.04)
7	Dong 306	1829 (8872)	1.192 (1.589)	0.236 (0.328)	19.84 (20.67)	20.06 (21.04)	5.12 (5.22)

*Figures in parentheses indicate the performance at $25 \pm 1^\circ\text{C}$ and RH $75 \pm 5\%$.

Table 4. Performance of the dumb-bell breeds at $31 \pm 1^\circ\text{C}$ and RH $85 \pm 5\%$

Sl. No	Breed	Survival No.	Cocoon Wt. (g)	Shell Wt. (g)	Shell Ratio (%)	Total larval period (hrs)	V age period (hrs)
1	912B2	7300 (9170) *	1.434 (1.679)	0.284 (0.368)	19.80 (21.9)	20.00 (22.00)	5.12 (6.00)
2	935 A	7206 (9333)	1.309 (1.586)	0.251 (0.354)	19.2 (22.3)	21.00 (22.06)	5.12 (6.12)
3	935 E	7800 (8984)	1.375 (1.579)	0.287 (0.336)	20.9 (21.3)	20.22 (22.00)	5.14 (5.22)
4	916 A1	7700 (9226)	1.306 (1.785)	0.278 (0.391)	21.3 (21.9)	21.00 (22.06)	5.12 (6.22)
5	44 FM	1900 (9134)	1.102 (1.605)	0.209 (0.350)	19.00 (21.8)	21.06 (22.12)	5.14 (6.12)
6	J2	1200 (8993)	0.992 (1.769)	0.187 (0.384)	18.9 (21.7)	21.14 (22.12)	5.12 (6.06)
7	NB4D2	2000 (9327)	1.170 (1.579)	0.236 (0.330)	20.2 (20.9)	21.12 (22.08)	5.22 (6.06)
8	DD3	1300 (9173)	1.191 (1.549)	0.236 (0.322)	19.8 (20.8)	21.10 (22.00)	5.18 (6.06)

*Figures in parentheses indicate the performance at $25 \pm 1^\circ\text{C}$ and RH $75 \pm 5\%$.

were prepared. A total of eight foundation crosses were prepared and screened at a temperature of $31 \pm 1^\circ\text{C}$ and relative humidity of $85 \pm 5\%$.

Data were recorded on the rearing and reeling characters viz., 1) total larval duration (hrs), 2) V instar duration (hrs), 3) cocoon yield by number, 4) cocoon yield by weight (kg), 5) cocoon weight (g), 6) cocoon shell weight (g), 7) cocoon shell ratio (%), 8) filament length (m), 9) denier, 10) raw silk % and 11) reelability (%).

From each hybrid combination, 60 cocoons were reeled in Japanese multiend reeling machine. An evaluation index (e.i) was calculated for each character by the formula mentioned below:

$$e.i. = \frac{A-B}{C} \times 10 + 50$$

Where A = Value of a particular hybrid
 B = Mean value of all the hybrids
 C = Standard deviation of all the hybrids
 10 = Standard unit
 50 = Fixed value

Results

Mean values and standard deviation for each trait in all the eight foundation crosses were calculated. For each trait the evaluation index (E.I) was calculated. The E.I calculated for 11 traits were used in calculation of 'Evaluation Index for each foundation cross. Different foundation crosses reared at temperature of $25 \pm 1^\circ\text{C}$ and rel-

ative humidity of $70 \pm 5\%$ indicated higher values for different economic traits. 1) A \times Tokai for shorter larval period (514 hrs), 2) Tokai \times C124 and A \times Tokai for shorter V instar period (126 hrs), 3) A \times Tokai for yield/10,000 larvae by number (9,800), 4) 912 B2 \times 935 A for yield/10,000 larvae by weight (16.53 kg), 5) C3 \times A for single cocoon weight (1.665 g), 6) Tokai \times C124 for single shell weight (0.357 g), 7) A \times Tokai for shell ratio (22.02%), 8) 916 A1 \times 935 A for average filament length (996.30 m), 9) A \times C3 for denier (2.18), 10) C3 \times A for raw silk % (13.50%), 11) 916 A1 \times 935 A for reelability (98.93%), (Table 5).

The results of the foundation crosses reared at high temperature of $31 \pm 1^\circ\text{C}$ and relative humidity of $85 \pm 5\%$ are as follows: 1) 916 A2 \times 912B2, Tokai \times C124 for shorter larval period (504 hrs), 2) A \times Tokai, 912B2 \times 935A for shorter V instar period (120 hrs), 3) A \times Tokai for yield/10,000 larvae by number (8315), 4) Tokai \times C124 for yield/10,000 larvae by weight (10.87 g), 5) Tokai \times C124 for single cocoon weight (1.291 g), 6) 916 A1 \times 935 A for single shell weight (0.268 g), 7) 916 A1 \times 935 A for shell ratio (20.87%), 8) 916 A1 \times 935 A for average filament length (995.04 m), 9) Tokai \times C124 for denier (2.09), 10) C3 \times A for raw silk % (11.29%), 11) 916 A1 \times 935 A for reelability (90.20%) (Table 7).

Although there are 21 characters contributing to silk yield (Thiagarajan *et al.*, 1993 a, b), in this study it is essential to select foundation crosses with the combinations of all the economic characters contributing to silk yield. Therefore, evaluation index method was employed

Table 5. Rearing performance of the foundation crosses at room temperature

Sl. no.	Hybrid	Larval period (hrs)	V age period (hrs)	Yield/10000 Larvae		Single cocoon weight (g)	Single shell weight (g)	Shell ratio (%)	average filament length (m)	Denier	Raw silk (%)	Reel-ability (%)
				No.	Wt (kg)							
1	935Ax916A1	558.00	144.00	9500.00	15.20	1.582	0.334	21.11	994.10	2.55	10.10	85.65
2	916A2x912B2	528.00	144.00	9100.00	13.47	1.627	0.349	21.45	936.80	2.32	13.50	92.20
3	916A1x935A	552.00	144.00	9666.00	15.60	1.583	0.345	21.79	996.30	2.30	12.29	98.93
4	C3xA	528.00	150.00	9000.00	14.99	1.665	0.350	21.02	960.30	2.39	13.89	89.98
5	AxTokai	514.00	126.00	9800.00	14.00	1.544	0.340	22.02	891.70	2.32	10.38	83.02
6	912B2x935A	528.00	130.00	9600.00	16.53	1.428	0.297	20.80	889.80	2.33	10.71	85.21
7	AxC3	528.00	138.00	9000.00	13.80	1.652	0.347	21.00	864.90	2.18	12.00	89.71
8	TokaixC124	528.00	126.00	9733.00	14.67	1.664	0.357	21.45	754.30	2.28	11.30	81.69
	Mean	530.20	137.20	9479.80	14.76	1.573	0.331	21.05	911.03	2.29	11.67	87.40
	S.D.	13.96	8.82	303.25	0.86	0.084	0.024	0.67	107.35	0.13	1.20	5.04

Table 6. Evaluation index values of the foundation crosses at room temperature

Sl. no.	Hybrid	Yield/10,000 Larvae		Single cocoon weight (g)	Single shell weight (g)	Shell ratio (%)	average filament length (m)	Denier	Raw silk (%)	Reel-ability (%)	AVG. EI
		No.	Wt (kg)								
1	935Ax916A1	50.67	55.12	51.11	51.10	50.93	56.53	70.29	36.90	46.54	53.17
2	916A2x912B2	37.48	35.01	56.47	57.43	56.00	51.19	52.06	65.20	65.20	52.89
3	916A1x935A	56.14	59.76	51.23	55.74	61.16	56.55	50.48	55.13	55.13	56.74
4	C3xA	34.18	52.63	61.00	57.85	49.55	53.38	57.61	68.44	68.44	55.90
5	AxTokai	60.56	41.21	46.58	53.63	64.57	46.99	52.06	39.23	39.23	49.34
6	912B2x935A	53.96	70.58	32.75	35.48	46.23	46.81	52.85	41.98	41.98	46.96
7	AxC3	34.18	38.89	59.45	56.58	49.31	44.49	40.97	52.71	52.71	47.70
8	TokaixC124	58.35	48.93	60.88	60.80	56.06	34.19	48.89	46.89	46.89	51.32

Table 7. Rearing performance of the foundation crosses at 31±1°C and RH 85±5%

Sl. no.	Hybrid	Larval period (hrs)	V age period (hrs)	Yield/10000 Larvae		Single cocoon weight (g)	Single shell weight (g)	Shell ratio (%)	average filament length (m)	Denier	Raw silk (%)	Reel-ability (%)
				No.	Wt (kg)							
1	935Ax916A1	534.00	138.00	8000.00	8.30	1.134	0.236	20.81	937.09	2.50	9.80	85.00
2	916A2x912B2	504.00	130.00	8123.00	8.61	1.170	0.235	20.09	736.10	2.38	9.70	89.00
3	916A1x935A	514.00	138.00	8033.00	9.56	1.284	0.268	20.87	995.04	2.40	9.13	90.20
4	C3xA	528.00	144.00	8000.00	10.80	1.210	0.245	20.21	860.30	2.45	11.29	87.80
5	AxTokai	510.00	120.00	8315.00	9.06	1.134	0.235	20.73	748.28	2.32	9.62	80.30
6	912B2x935A	510.00	120.00	7500.00	8.78	1.170	0.235	20.08	758.19	2.33	9.33	85.00
7	AxC3	510.00	126.00	7900.00	9.87	1.246	0.249	19.98	719.20	2.38	10.40	89.31
8	TokaixC124	504.00	126.00	8300.00	10.87	1.291	0.258	19.98	686.40	2.09	9.33	88.88
	Mean	513.20	130.00	7866.63	9.53	1.20	0.25	20.35	793.68	2.31	9.73	86.94
	S.D.	9.68	8.49	280.06	0.82	0.06	0.01	1.21	100.67	0.14	0.63	3.89

giving due importance to all the economic traits contributing to silk yield in selection of foundation crosses. In the present study the foundation crosses reared at 25 ± 1°C

and relative humidity of 70 ± 5% which recorded an average index value >50 are as following : 916 A1 × 935 A (56.74), C3 × A (55.90), Tokai × C124 (51.32) and 935 A

Table 8. Evaluation index values of the foundation crosses at high temperature

Sl. no.	Hybrid	Yield/10,000 Larvae		Single cocoon weight (g)	Single shell weight (g)	Shell ratio (%)	average filament length (m)	Denier	Raw silk (%)	Reel-ability (%)	AVG. EI
		No.	Wt (kg)								
1	935Ax916A1	54.76	35.10	39.91	49.48	60.03	64.25	63.24	51.08	45.71	51.51
2	916A2x912B2	33.34	38.88	45.83	58.91	61.94	44.28	54.79	49.49	58.86	49.59
3	916A1x935A	55.94	50.41	64.58	56.73	43.30	70.00	56.20	40.47	62.80	55.60
4	C3xA	54.76	65.46	52.33	39.69	37.85	56.62	59.72	74.67	54.91	55.11
5	AxTokai	36.91	44.34	39.86	49.24	58.70	45.49	50.56	48.23	30.26	44.84
6	912B2x935A	36.91	40.85	45.89	58.91	61.91	46.47	51.27	43.64	45.71	47.95
7	AxC3	51.19	54.15	58.33	42.95	35.91	42.60	54.79	60.58	59.88	51.15
8	TokaixC124	65.47	66.27	65.73	68.33	52.64	39.34	34.37	43.64	58.46	54.92

× 916 A1 (53.17), (Table 6) and at $31 \pm 1^\circ\text{C}$ the foundation crosses which scored above the average evaluation index value are: 916 A1 × 935 A (55.60), C3 × A (54.11), Tokai × C124 (54.92) and 935 A × 916 A1 (51.51) (Table 8).

Discussion

China has succeeded in evolving thermo-tolerant bivoltine silkworm breeds suitable for spring, summer and autumn season (Shao *et al.*, 1987; Shao, 1989 He *et al.*, 1989; He *et al.*, 1990; He *et al.*, 1991). Chen (1994) and Zhang *et al.* (1994) bred silkworm varieties suitable for summer-autumn season by following the technique of hybridisation, rearing of silkworm at a temperature of 29 - 32°C, relative humidity of 85% and QTL selection in the early breeding.

Xu *et al.* (1990) bred silkworm varieties by employing cross breeding, back cross breeding, line isolation and reeling of fresh cocoons suitable for spring season. Baoyu *et al.* (1996) bred silkworm varieties by hybridisation, directional breeding under high temperature, high humidity for summer season. Shao *et al.* (1990) and Yong *et al.* (1996) developed silkworm varieties with high cocoon and silk quality suitable for autumn season. Datta *et al.* (2000) bred robust bivoltine silkworm hybrid CSR18 × CSR19 tolerant at $36 \pm 1^\circ\text{C}$, Suresh *et al.* (1999) studied the performance of robust and productive bivoltine hybrids of silkworm under high temperature of $36 \pm 1^\circ\text{C}$ and high humidity conditions of $80 \pm 5\%$. Krishna *et al.* (1996) bred two silkworm breeds *viz.*, NP2 and SP2 tolerant to high temperature suitable for the tropical climate. Ramesh *et al.* (1996) utilized multivoltine and bivoltine races as breeding resource material and isolated four strains. The hybrid of these strains showed high survival with moderate productivity suitable for rearing in the tropics.

In the present study the breeding resource material was identified by screening the silkworm breeds at a temperature of $31 \pm 1^\circ\text{C}$ and relative humidity of $85 \pm 5\%$. The tolerant parents were crossed and foundation crosses were prepared and breeding initiated. This is in conformity with the Chinese technique (rearing of silkworm at a temperature of 29 - 32°C and relative humidity of 85% and QTL selection in the early breeding).

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