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Evaluation of genetic potential of the polyvoltine silkworm (*Bombyx mori* L.) germplasm and identification of parents for breeding programme*

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Abstract: In the present study, polyvoltine germplasm stock of Andhra Pradesh State Sericulture Research and Development, Institute (APSSRDI) was evaluated for its performance based on quantitative and qualitative traits. Twenty-one oval and 10 peanut cocoon shaped lines were reared in different seasons of the year. Since the polyvoltines are non-diapausing, six generations were reared and evaluated for various economically important traits based on evaluation index and sub-ordinate function statistical methods. Ten top ranked lines obtained by using both the methods were identified as potential parental strains. Among oval lines, APM14, APM11, APM18, APMW9, and APM19, and among peanut lines APMD5, APMD1, APMD3, APMD9 and APMD8 were selected as base material. The identified high yielding lines will be used in various breeding programmes as initial parents for the synthesis of superior polyvoltine breeds/hybrids.

Key words: Polyvoltine, Silkworm, Germplasm, Performance, Evaluation
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INTRODUCTION

Silkworm is not only a commercially important insect, it is also found to be an important laboratory tool. It is estimated that more than 3000 silkworm strains are available all over the world due to various ongoing breeding programmes (Nagaraju, 2002; Thangavelu *et al*., 2003). These silkworm varieties include univoltines, bivoltine and polyvoltines. Univoltines and bivoltines are qualitatively and quantitatively superior races whereas polyvoltines are relatively inferior in both the traits but superior in their survival and hardiness. But in tropical countries like India, polyvoltine silkworm strains play important role in the production of silk, since they are well accustomed to the tropical climatic conditions. Hence, maintenance of polyvoltine silkworm genetic resources has become very important for meeting the desired objectives of the breeder for immediate or long-term utilization in silkworm breeding programmes. But, it is necessary to maintain them in their original form for their rational use in different breeding and other research purposes (Mukarjee *et al.*, 1999; Basavaraja *et al*., 2003; Thangavelu *et al*., 2003; Yamaguchi, 2003). In addition to maintenance, systematic study of resource material is also very important, not only for classification and characterization of varieties but also for the selection of promising parents to initiate various breeding programmes. Evaluation of genetic material also helps in identification of lines with special characters like longer filament length, fine denier, fluoride resistance, disease resistance, etc. (Li *et al*., 2001). Availability of diverse genetic stock, gives ample choice for the breeder in selection of initial parents of his desire.

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Even half of a good silkworm egg laying from a good genetic stock can potentially transform the sericulture scenario to a greater extent (Chandrashekaraiah and Ramesh Babu, 2003).

Polyvoltine silkworm gene bank of APSSRDI, Hindupur consists of geographically isolated races and breeds, obtained from indigenous and exotic origin. In the present study, an attempt was made to evaluate and characterize polyvoltine germplasm and inbred lines based on evaluation index and sub-ordinate function statistical methods, frequently used for evaluating breeds/hybrids. Germplasm evaluation was conducted to ascertain the genetic potential of various silkworm lines/strains for commercial exploitation. Since sericulture is practiced in diverse agro-climatic zones, systematic evaluation is needed for the proper utilization of the available lines. The information generated will be useful for future breeding programmes.

MATERIALS AND METHODS

For the present study, 21 oval cocoon types (viz. APMW12, APM25, APMW10, APMW7, APM11, APMW9, APMW8, APM21, APM24, APMG1, APM27, APM4, APM14, APM3, APM20, APM22, APM28, APM19, APM18, APM26, APM23) and 10-peanut type (APMD1, APMD2, APMD3, APMD4, APMD5, APMD6, APMD7, APMD8, APMD9, APMD10) polyvoltine germplasm lines with genetically varied characters were selected. The lines were reared under standard rearing conditions (Krishnaswami, 1975; 1983) for six generations in various seasons of the year. The young larvae (1st~3rd instars) were reared at 27~28 °C with 85%~90% relative humidity and the late age larvae (4th and 5th instars) were maintained at 24~26 °C with a relative humidity of 70%~80%. Each breed was maintained in three replications. At the beginning of 4th instar, 300 larvae were counted from each line and retained for further studies. Rearing was carried out under hygienic conditions. At the end of 5th instar, the spinning larvae were collected manually and mounted in plastic collapsible mountages. During the rearing period, larvae and cocoons were assessed for the yield parameters like fecundity, cocoon yield, pupation rate, cocoon weight, shell weight, shell ratio, filament length, raw silk (%), reelability (%) and neatness using the following formulae:

Cocoon yield/10000 larvae=(weight of cocoons obtained (kg))×10 000/(larvae retained after 3rd moult (300));

Pupation rate (%)=(No. of good cocoons+(No. of double cocoons×2))×100/(larvae retained after 3 rd moult–uzi infestated cocoons);

Cocoon weight (g)=(wt. of 25 male (g)+25 female cocoons (g))/50;

Cocoon shell weight (g)=(wt. of 25 male (g)+25 female cocoon shells (g))/50;

Cocoon shell ratio (SR, %)=(single cocoon shell weight (g))×100/(single cocoon weight (g));

Silk filament length (m)=revolutions of aprovette× circumference of wheel (m).

These data were analyzed further by using the following evaluation index method and sub-ordinate function method. Based on these values obtained, ranks were given for each breed accordingly.

Evaluation index method

Evaluation index value (EI) for silkworm breed performance was calculated by using the following formula (Mano *et al*., 1993).

$$EI=(A-B)/C\times 10+50,$$

where, A is mean of the particular trait; B is overall mean of particular trait; C is standard deviation; 50 is constant.

Sub-ordinate function method

Sub-ordinate function is calculated by utilizing the following formula (Gower, 1971).

$$X_u=(X_i-X_{\min})/(X_{\max}-X_{\min}),$$

where, X_u is sub-ordinate function; X_i is measurement of trait of tested breed; $X_{\min}$ is minimum value of the trait among all the tested breeds; $X_{\max}$ is maximum value of the trait among all the tested breeds.

RESULTS AND DISCUSSION

The germplasm lines were broadly divided into two groups based on the shape of the cocoons.

Oval lines

Among 21 oval shaped (cocoon) lines, larvae of almost all the strains were found plain except the line APM19 that showed larval marks on the dorsal surface of their body. However these lines have produced cocoons of different colours like white (APMW12, APMW10, APMW7, APM11, APMW9, APMW8, APM21, APM24), greenish yellow (APMG1, APM27, APM4, APM14, APM3), yellow (APM20, APM22, APM28), cream colour (APM25) and flesh colour (APM19, APM18, APM26, APM23) cocoons and the cocoon grains were found to be fine to medium and coarse (Table 1).

Table 1 Certain phenotypic traits of the lines studied

SL. No	Lines	Larval markings	Cocoon colour	Cocoon grains
Oval lines				
1	APMW12	Plain	White	Medium
2	APM25	Plain	Cream	Coarse
3	APM10	Plain	White	Coarse
4	APM14	Plain	Gr. yellow	Fine
5	APM11	Plain	White	Fine
6	APMW9	Plain	White	Medium
7	APMW8	Plain	White	Fine
8	APM21	Plain	White	Coarse
9	APM24	Plain	White	Medium
10	APMG1	Plain	Gr. yellow	Medium/fine
11	APM27	Plain	Gr. yellow	Medium/fine
12	APM4	Plain	Gr. yellow	Fine
13	APMW7	Plain	White	Coarse
14	APM3	Plain	Gr. yellow	Fine
15	APM20	Plain	Yellow	Medium
16	APM22	Plain	Yellow	Medium
17	APM28	Plain	Yellow	Medium
18	APM19	Marked	Flesh	Coarse
19	APM18	Plain	Flesh	Fine
20	APM26	Plain	Flesh	Fine
21	APM23	Plain	Flesh	Medium
Peanut lines				
1	APMD1	Plain	White	Medium
2	APMD2	Plain	White	Coarse
3	APMD3	Plain	White	Medium
4	APMD4	Marked	White	Medium
5	APMD5	Marked	Yellow	Fine
6	APMD6	Marked	Yellow	Fine
7	APMD7	Plain	Flesh	Fine
8	APMD8	Marked	Flesh	Fine
9	APMD9	Plain	Flesh	Fine
10	APMD10	Marked	Flesh	Medium

The fecundity (number of eggs laid by a single female silk moth) among oval lines ranged between 438 (APMG1 and APM26) and 567 (APM14) and average fecundity was found to be 474. Lowest cocoon yield (yield/10000 larvae by weight) was recoded in APMG1 (8.836 kg) and highest was in APM28 (12.261 kg) with an average yield of 10.85 kg. In all the lines, pupation (survival) was recorded more than 80% except in APMG1 (72.55%) and APM11 (76.73%) and the average survival rate was 86.4%. Single cocoon weight was found to be highest in APM26 (1.412 g) and lowest in APMW12 (1.199 g) with an average weight of 1.312 g. Maximum cocoon shell ratio was recorded in APM11 (18.58%), minimum in APM25 (15.60%) while average cocoon shell ratio was 16.27%. Highest silk filament length (m) was recorded in APM11 (996 m), lowest in APM25 (604 m). Raw silk (%) was maximal in APM11 (13.03%) and minimum in APM20 (10.48%). In all the oval lines, silk reelability was observed to be above 70%. Highest neatness was recorded in APM14 (83.2) followed by APMW7 (82.0) and APM25 (81.2) whereas lowest was recorded in APM20 (73.0) followed by APM28 (76.2) and APM26 (77.2) (Table 2).

Peanut lines (dumbbell lines)

Among ten peanut shaped lines studied, the larvae of 5 lines (APMD1, APMD2, APMD3, APMD7 and APMD9) were found to be plain while the remaining five (APMD4, APMD5, APMD6, APMD8 and APMD10) were marked. Four lines (APMD1, APMD2, APMD3 and APMD4) yielded white colour cocoons, 2 lines yielded yellow colour cocoons with remaining 4 lines (APMD7, APMD8, APMD9 and APMD10) found to be flesh colour cocoons. The cocoon grains were found to be fine to medium and coarse (Table 1).

In the peanut lines, low fecundity was recorded in APMD9 (441) followed by APMD1 (443) and high fecundity was found in APMD6 (505) followed by APMD2 (487). Yield (yield/10000 larvae), was low in APMD4 (10.30 kg) and high in APMD2 (11.66 kg) followed by APMD6 (11.64 kg). Almost all the lines showed more than 80% pupation rate. Maximum cocoon weight was recorded in APMD8 (1.368 g) followed by APMD2 (1.358 g) whereas minimum was recorded in APMD7 (1.247 g). Highest cocoon shell ratio was recorded in APMD4 (17.39%) followed by APMD8 (16.99%) whereas lowest was recorded in APMD1 (14.90%), followed by APMD10 (15.28%). Maximum filament length was recorded in APMD6 (905 m), minimum measured in APMD1 (610 m). Raw silk percentage was found to be highest in APMD4 (11.95%), lowest in APMD1 (10.02%). Almost all the stock recorded more than 70% reelability and neatness (Table 3).

Table 2 Mean performance of oval type multivoltine silkworm breeds

SL. No.	Breed	Fecundity (No.)	Yield/10000 larvae (kg)	Pupation rate (%)	Cocoon weight (g)	Shell weight (g)	Shell ratio (%)	Filament length (m)	Raw silk (%)	Reelability (%)	Neatness (P)
1	APMW12	484	10.379	89.63	1.199	19.03	15.69	670	10.63	74	79.2
2	APM25	477	10.347	88.07	1.226	19.28	15.60	604	10.87	76	81.2
3	APM10	479	10.669	90.33	1.258	20.22	15.94	619	10.77	74	79.0
4	APM14	567	11.377	87.90	1.332	22.85	16.91	950	12.22	78	83.2
5	APM11	465	8.840	76.73	1.346	25.28	18.58	996	13.03	76	80.5
6	APMW9	490	11.455	87.67	1.364	22.98	16.63	873	11.37	74	79.3
7	APMW8	476	11.025	89.84	1.276	20.60	15.96	766	10.87	74	79.3
8	APM21	492	10.619	86.75	1.301	21.87	16.60	801	11.52	75	80.4
9	APM24	470	11.316	84.80	1.330	22.67	17.02	968	11.76	75	80.0
10	APMG1	438	8.836	72.55	1.275	21.55	16.85	803	11.62	75	79.8
11	APM27	440	10.836	88.34	1.244	20.15	16.10	665	10.82	73	78.3
12	APM4	451	11.090	89.19	1.282	21.37	16.56	711	11.12	73	78.2
13	APMW7	475	10.642	88.35	1.230	20.23	16.42	839	11.64	77	82.0
14	APM3	477	10.730	86.08	1.328	21.32	15.99	712	10.95	75	79.7
15	APM20	470	11.414	87.20	1.351	21.58	16.09	729	10.48	71	73.0
16	APM22	493	11.180	88.35	1.337	20.97	15.69	789	10.66	74	79.3
17	APM28	441	12.261	91.22	1.387	21.70	15.63	736	10.70	75	76.2
18	APM19	456	11.056	81.74	1.386	21.75	15.67	719	10.62	74	79.2
19	APM18	481	11.231	85.76	1.390	22.63	16.18	783	11.05	75	79.5
20	APM26	438	11.358	84.53	1.412	22.70	15.95	778	11.02	75	77.2
21	APM23	487	11.054	89.21	1.290	20.25	15.65	749	10.81	75	80.3
	Mean	473.6	10.843	86.391	1.312	21.48	16.27	777.9	11.17	74.8	79.3
	SD	28.1	0.793	4.523	0.060	1.44	0.70	114.0	0.62	1.4	2.1
	Min.	438	8.836	72.55	1.199	19.03	15.60	604	10.48	71	73.0
	Max.	567	12.261	91.22	1.412	25.28	18.58	996	13.03	78	83.2

Table 3 Mean performance of peanut type polyvoltine germplasm

SL. No.	Breed	Fecundity (No.)	Yield weight (kg)	Pupation rate (%)	Cocoon weight (g)	Shell weight (g)	Shell ratio (%)	Filament length (m)	Raw silk (%)	Reelability (%)	Neatness (P)
1	APMD1	443	10.84	89.26	1.270	19.02	14.90	610	10.02	74	79
2	APMD2	487	11.66	88.91	1.358	22.13	16.12	740	11.04	75	78
3	APMD3	470	10.74	84.64	1.309	22.20	16.83	702	11.45	74	79
4	APMD4	451	10.30	87.05	1.268	22.23	17.39	866	11.95	75	80
5	APMD5	454	10.45	83.51	1.324	22.67	16.95	660	11.48	74	79
6	APMD6	505	11.64	89.02	1.345	21.65	16.03	905	10.93	74	79
7	APMD7	454	10.65	87.82	1.247	20.65	16.53	655	11.37	75	80
8	APMD8	460	10.75	81.50	1.368	23.27	16.99	696	11.64	74	79
9	APMD9	441	10.99	88.20	1.278	20.23	15.79	622	10.76	75	80
10	APMD10	445	10.60	86.23	1.275	19.53	15.28	616	10.37	74	79
	Mean	461	10.86	86.61	1.304	21.36	16.28	707	11.10	74	79
	SD	21	0.46	2.63	0.043	1.42	0.80	103	0.59	0.3	0.5
	Min.	441	10.30	81.50	1.247	19.02	14.90	610	10.02	74	78
	Max.	505	11.66	89.26	1.368	23.27	17.39	905	11.95	75	80

Analysis of data

Based on rearing performance, quantitative traits of lines were assessed on different parameters like fecundity, yield/10000 larvae (kg), pupation rate (%), cocoon weight, cocoon shell weight, shell ratio, filament length, raw silk (%), reelability and neatness using the evaluation index and sub-ordinate function methods and the details are as follows.

Among oval lines, as per the evaluation method, the lines APM14 (64.2), APM11 (57.4), APM18 (55.9), APMW9 (54.8), APM19 (52.7), APMG1 (52.3), APM4 (52.1), APMW7 (50.2) showed higher evaluation index values (above 50 EI value) and APM28 (43.5), APMW12 (43.6) showed lower index (Table 4). Whereas six peanut lines viz. APMD5 (57.6), APMD1 (54.5), APMD3 (53.8), APMD9 (52.7), APMD8 (50.9) and APMD2 (50.1) showed higher performance index (above 50 EI) (Table 5).

The other important method used in line evaluation is sub-ordinate function method (Gower, 1971). As per this method, among oval lines, APM14 (7.711), APM11 (6.363) and APM18 (6.030), have shown higher index values whereas APMW12 (2.989) and APM28 (3.092) showed lower index values (Table 4)

Table 4 Ranking of polyvoltine germplasm breeds (oval type)

SL. No.	Breed	Evaluation index method		Sub-ordinate function method	
		Value	Rank	Value	Rank
1	APM14	64.2	1	7.711	1
2	APM11	57.4	2	6.363	2
3	APM18	55.9	3	6.030	3
4	APMW9	54.8	4	5.757	4
5	APM19	52.7	5	5.156	5
6	APMG1	52.3	6	4.976	7
7	APM4	52.1	7	5.144	6
8	APMW7	50.2	8	4.687	8
9	APM20	49.6	9	4.500	10
10	APM21	49.4	10	4.553	9
11	APM3	49.2	11	4.323	11
12	APM22	48.6	12	4.256	12
13	APMW8	48.3	13	4.177	13
14	APM23	47.3	14	4.011	14
15	APM24	47.0	15	3.953	15
16	APM25	45.8	16	3.414	17
17	APMW10	45.6	17	3.515	16
18	APM26	44.4	18	3.358	18
19	APM27	43.7	19	3.266	19
20	APMW12	43.6	20	2.989	21
21	APM28	43.5	21	3.092	20

and in the case of peanut lines, APMD5 (7.605), APMD1 (6.626), APMD3 (6.193), APMD9 (5.885) and APMD8 (5.156) ranked high (Table 5).

Table 5 Ranking of multivoltine germplasm breeds (peanut type)

SL. No.	Breed	Evaluation index method		Subordinate function method	
		Value	Rank	Value	Rank
1	APMD5	57.6	1	7.605	1
2	APMD1	54.5	2	6.626	2
3	APMD3	53.8	3	6.193	3
4	APMD9	52.7	4	5.885	4
5	APMD8	50.9	5	5.156	5
6	APMD2	50.1	6	5.097	6
7	APMD10	47.7	7	4.223	7
8	APMD4	47.2	8	4.170	8
9	APMD6	43.6	9	2.898	9
10	APMD7	42.0	10	2.455	10

After evaluation by both the statistical methods (evaluation index method and sub-ordinate function method) five oval type lines (APM14, APM11, APM18, APMW9, APM19) and five peanut shaped lines (APMD5, APMD1, APMD3, APMD9, APMD8) that stood within five ranks were identified as potential parents for further breeding programme. Constant efforts are being made to develop productive polyvoltine silkworm hybrids suitable for sericulture in tropical agro-climatic conditions of peninsular India because, more than ninety percent of the raw silk is still coming from polyvoltine silkworm hybrids (polyvoltine×bivoltine cross breeds) only. Therefore, maintenance of polyvoltine resource material and their effective utilization has become very important. Most of the quantitative traits of commercial importance in silkworm are under complicated polygenic control under the influence of the environment. For synthesizing the potential polyvoltine cross breeds, usually, the high yielding traits of bivoltine varieties and fitness traits of polyvoltine strains are hybridized as proper selection of potential and homozygous parents is very important. Effective utilization of selected germplasm also plays an important role in saving the time of the breeder in the synthesis of new hybrids. Keeping the need in view, the germplasm strains have been reared consecutively for several generations and their quantitative traits were evaluated using two reliable statistical methods, i.e.

evaluation index method and sub-ordinate function method to assess the performance of the inbred lines. Earlier many breeders (Mano *et al*., 1993; Gower, 1971; Ramesh Babu *et al*., 2001, Rao *et al*., 2004) analyzed their breeds by adopting the above methods either individually or together. The breeds, which have been selected through these methods could be effectively used in further breeding programmes as potential parents for synthesizing superior polyvoltine silkworm hybrids that are suitable for culture under tropical climatic conditions. Proper choice of suitable parents plays a pivotal role in achieving the targets in scheduled time.

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