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Shabir Ahmad Bhat
Temperate Sericulture Research
Institute, SKUAST-Kashmir,
Jammu and Kashmir, India

Ravi Kant
Temperate Sericulture Research
Institute, SKUAST-Kashmir,
Jammu and Kashmir, India

Naveena Nazim
Temperate Sericulture Research
Institute, SKUAST-Kashmir,
Jammu and Kashmir, India

Mohd Rafiq Bhat
Temperate Sericulture Research
Institute, SKUAST-Kashmir,
Jammu and Kashmir, India

Noor-ul-Din
Temperate Sericulture Research
Institute, SKUAST-Kashmir,
Jammu and Kashmir, India

Munazah Yaqoob
Division of Entomology,
SKUAST-Kashmir, Jammu and
Kashmir, India

Nageena Nazir
Division of Agri-Statistics,
SKUAST-Kashmir, Jammu and
Kashmir, India

Correspondence
Shabir Ahmad Bhat
Temperate Sericulture Research
Institute, SKUAST-Kashmir,
Jammu and Kashmir, India

Genetic variability analysis of some bivoltine silkworm (*Bombyx mori* L.) breeds

Shabir Ahmad Bhat, Ravi Kant, Naveena Nazim, Mohd Rafiq Bhat, Noor-ul-Din, Munazah Yaqoob and Nageena Nazir

Abstract

Nineteen bivoltine silkworm breeds of varied eco-geographical origin were analysed for genetic variability for eleven economic traits. The study revealed significant differences among the breeds in traits studied. On the basis of clustering, breeds could be grouped into seven clusters and the number of breeds in different clusters varied. Cluster 1st comprised maximum (five) breeds and the cluster 7th had only one breed. Breeds spinning oval and constricted cocoons fell in the same cluster which clearly showed that breeds exhibiting phenotypic divergence may not be genetically diverse. The clustering pattern also depicted a mixed trend, silkworm breeds having different geographic origin were included in the same cluster and breeds having same geographic origin fell in different clusters this revealed that geographic and genetic diversity have no association. Crossing of breeds from most distant clusters may yield heterotic crosses and transgressive segregants in advanced generations.

Keywords: bivoltine, cluster, geographical, quantitative, silkworm, trait, variability

Introduction

The success of any breeding programme depends on the selection of appropriate genetic resources, their judicious utilization in appropriate and desirable combinations and also, choice of mating systems to generate ample genetic variability for facilitating enough scope for selection. Therefore, it is of paramount importance that utmost care is taken in verifying and analyzing the genetic worth of parents to be utilized as breeding resource material. Further, selection of appropriate resource materials for any improvement programme is very essential and aims at a derivation of better constellation of gene complexes which suits to express good phenotypic values on a wide range of traits by amalgamating distinct and different gene pools. Conventional breeding methods are directed not only for the evolution of new breeds but also for the identification of the promising hybrid combinations for commercial exploitation based on the heterosis expressed in F₁ hybrids. Many silkworm breeders, Trag *et al.* (1992) [18], Malik *et al.* (2001) [9], Dandin *et al.* (2005) [4], Singh *et al.* (2006) [15] and Malik *et al.* (2010) [8] emphasized the need for better understanding of genetic diversity of parental strains to be utilized in the breeding programme by the systematic evaluation. Therefore it's imperative to evaluate diverse genotypes and to identify the distinct and different gene pools existing in a group of resource materials to be utilized in breeding programmes. The present study was, therefore, undertaken to assess the genetic diversity of some of the available and promising silk worm breeds to identify diverse parental stocks which could be utilized either as the parents of heterotic crosses or involved in future breeding programmes for the extraction of new and more productive silkworm breeds

Material and methods

Nineteen silkworm breeds of diverse eco- geographical origin viz. SK-1, SK-3, SK-6, SK-13, SK-22, SK-24, SK-28, SK-30, CSR-2, CSR-4, CSR-18, CSR-19, CSR-26, KSO, KH-Badami, M-106, DUN-6 and DUN-22 formed basis for the present study. The disease free layings (DFLS) of these selected breeds were obtained from the Germplasm Bank maintained at Temperate Sericulture Research Institute (TSRI), Mirgund, Sher-e-Kashmir University of Agricultural Sciences and Technology, of Kashmir (SKUAST-K) and Central Sericulture Research and Training Institute (CSR & TI) Central Silk Board (Govt. of India) Pampore.

The disease free layings of these breeds were incubated sbrushed and reared using standard rearing method (Anonymous, 2003) [2] during summer (July-August) season of 2015. The experiment was laid out in Completely Randomized Design (CRD) with three replications for each treatment. Each replication comprised 200 larvae (after third moult). The data pertaining to eleven metric traits viz. fecundity, hatching percentage, average larvae weight, fifth instar

larval duration, total larval duration, cocoon yield/10,000 larvae by number, cocoon yield/10,000 larvae by weight, single cocoon weight, single shell weight, shell ratio and pupation rate were recorded and subjected to analysis of variance and cluster analysis using R-software (3.2.1) package.

Results

Analysis of variance (Table-1) for eleven metric traits indicated presence of significant variability in the study materials. No specific breed performed best in all the traits studied. However, SK-6 outperformed all other breeds in nine of the eleven parameters under study. Whereas, SK-1, SK-28 and CSR-2 each performed best in one trait only (Table 2). On the basis of cluster means and D² values, these breeds could be grouped into seven clusters and the number of breeds in each cluster varied from one to five (Fig. 1 and Table 3). Cluster 1st, 2nd, 3rd, 4th, 5th, 6th and 7th included five, two, three, three, two, three and one silkworm breeds respectively. Cluster 1st comprised five silkworm breeds viz. SK-1, CSR-2, CSR-4, CSR-6 and KH-Badami, out of these two are temperate and three are topical in origin. Cluster 2nd comprised two breeds viz., SK-3 and CSR-19 which are temperate and tropical in origin respectively. Cluster 3rd comprised three silkworm breeds viz., SK-6, SK-28 and SK-30 which are high yielding temperate breeds developed by this institute. Cluster 4th also comprised three breeds viz. SK-13, SK-22 and DUN-6, out of which former two are temperate in origin and later one is sub-tropical in origin. Cluster 5th included two breeds viz. SK-24 and KSO, Out of these SK-24

is temperate in origin and KSO is subtropical in origin. Cluster 6th comprised three silkworm breeds viz. CSR-18, CSR-26 and DUN-22 among which first two are tropical in origin whereas, DUN22 is subtropical in origin. Cluster 7th was represented by only one breed namely M-106 which is temperate in origin. The ranking of clusters was calculated on the basis of overall performance of cluster with respect to economic parameters and cluster 3rd showed best performance among all the clusters and ranked 1st (Table 3). Cluster mean for different traits of these silkworms are presented in table 4. The fecundity ranged from 420-529, hatching percentage from 76.19-88.85, larval weight 36.78-41.48g, 5th instar larval duration 198.66-211h, total larval duration 688.00-677.77h, cocoon yield by number 6769-8793, cocoon yield by weight 11.21-15.04kg, single cocoon weight 1.59-17.20g, single shell weight 0.25-0.34g, SR ratio 15.15-19.76% and pupation 71.00-90.33%.

Out of eleven characters studied the maximum positive and significant associations were observed between single shell weight and shell ratio (0.9493) followed by hatching percentage and average larval weight (0.8830), pupation rate and average larval weight (0.8534), single cocoon weight and single shell weight (0.8484), pupation rate and hatching (0.8174), fecundity and pupation rate (0.7745), single cocoon weight and shell ratio (0.7503) respectively. However, negative correlations were observed between cocoon yield/10000 larva (by weight) and fifth instar larval duration (-0.687) followed by shell ratio and total larval duration (-0.547), total larval duration and hatching (-0.5059) and total larval duration and fecundity (-0.4899) (Table 5).

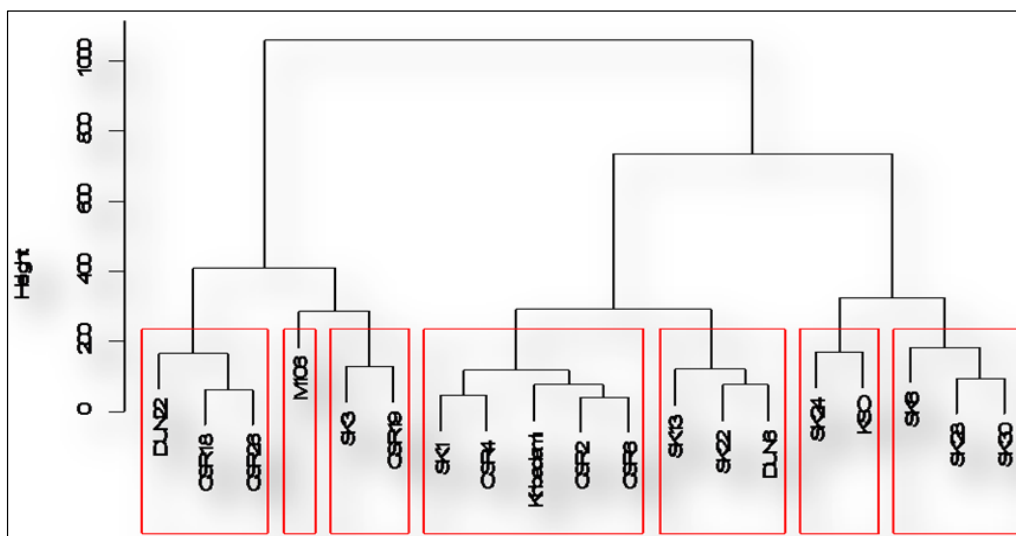


Fig 1: Dendrogram depicting the clustering pattern

Table 1: Mean performance of various silkworm breeds

Breeds	Fecundity (no.)	Hatching (%)	Larval weight (g)	5 th instar larval duration (h)	Total larval duration (h)	Cocoon yield by number	Cocoon yield by weight (Kg)	Single cocoon weight (g)	Single shell weight (g)	Shell ratio (%)	Pupation rate (%)
SK1	468	77.00	38.60	200	676.00	7736.33	11.80	1.87	0.39	20.85	81.00
SK3	503	76.00	36.80	204	678.00	7096.66	11.76	1.60	0.30	18.75	74.00
SK6	524	94.00	42.70	192	672.00	8914.33	15.75	1.86	0.40	21.50	92.30
SK13	509	86.64	40.36	204	678.00	8193.33	12.71	1.63	0.28	17.17	85.00
SK22	495	87.33	39.57	204	678.00	8038.66	13.02	1.71	0.34	19.58	86.30
SK24	456	88.35	41.13	198	678.66	8556.70	14.68	1.71	0.31	18.12	82.00
SK28	534	90.24	40.60	202	676.00	8687.00	14.43	1.66	0.31	18.67	91.30
SK30	529	82.33	41.16	202	685.33	8780.00	14.94	1.65	0.31	18.78	87.30
M-106	420	76.58	37.10	202	682.00	6769.33	13.65	1.65	0.25	15.15	71.00
KH-Badami	459	78.81	38.20	198	678.00	7920.66	14.85	1.59	0.24	15.09	79.00
CSR2	477	81.36	39.20	196	686.00	7829.33	13.93	1.47	0.19	12.92	80.00
CSR4	472	81.43	39.76	192	686.00	7778.33	13.35	1.67	0.29	17.36	83.00
CSR6	471	76.63	35.32	195	675.00	7866.34	13.76	1.77	0.34	19.20	77.30

CSR18	428	72.00	35.33	210	686.00	7283.00	11.24	1.71	0.25	14.61	74.30
CSR19	425	72.93	36.76	218	698.00	6999.00	10.67	1.63	0.27	16.56	76.00
CSR26	461	79.26	37.20	194	674.00	7329.67	12.87	1.69	0.32	18.93	71.00
KSO	515	86.83	40.12	196	676.00	8398.33	14.19	1.74	0.32	18.39	85.70
DUN6	498	80.66	39.00	196	676.00	8111.66	13.83	1.54	0.27	17.53	80.30
DUN22	475	77.31	38.77	204	682.00	7466.67	13.11	1.38	0.20	14.49	82.70
Mean	480	81.35	38.82	200.36	680.05	7476.54	13.39	1.65	0.29	17.57	81.06
SD	34.48	6.06	2.02	6.42	6.12	1711.30	1.34	0.12	0.05	2.29	6.15
C.D P≤0.05	3.932	0.575	0.332	0.173	0.213	5.410	0.071	0.001 [^]	0.001	0.064	0.6311

Table 2: Parameter-wise status of silkworm breeds

S. No	Silkworm breeds	Parameter	Trait status
1	SK-28	Fecundity (No.)	534.66
2	SK-6	Hatching (%)	94
3	SK-6 and CSR-4	Fifth instar larval duration (hrs)	192
4	SK-6	Total larval duration (hrs)	672
5	SK-6	Average larval weight (g)	42.70
6	SK-6	Cocoon yield by number	8914.33
7	SK-6	Cocoon yield by weight (kg)	15.75
8	SK-6	Pupation rate (%)	92.30
9	SK-1	Single cocoon weight (g)	1.87
10	SK-6	Single shell weight (g)	0.40
11	SK-6	Shell ratio (%)	21.50

Table 3: Cluster based classification and performance of silkworm breeds

Cluster number	Number of treatments/ breeds	Treatments/silkworm breeds	Ranking
1 st	5	SK1, CSR2, CSR4, CSR6 and KH-Badami.	5
2 nd	2	SK3 and CSR19	6
3 rd	3	SK6, SK28 and SK30	1
4 th	3	SK13, SK22 and DUN6	3
5 th	2	SK24 and KSO	2
6 th	3	CSR18, CSR26 and DUN22	4
7 th	1	M106	7

Table 4: Cluster mean for different characters of nineteen silkworm breeds

Cluster No.	Fecundity (No.)	Hatching (%)	Larval weight (g)	5 th instar larval duration (hrs.)	Total larval Duration (hrs.)	Cocoon yield by number	Cocoon yield by weight (kg)	Single cocoon weight (g)	Single shell weight (g)	Shell ratio (%)	Pupation rate (%)
1	469.66	79.04	38.21	196.20	680.20	7826.20	13.54	1.67	0.27	16.16	80.06
2	464.32	74.46	36.78	211.00	688.00	7047.83	11.21	1.62	0.28	17.28	75.33
3	529.33	88.85	41.48	198.66	677.77	8793.77	15.04	1.72	0.34	19.76	90.33
4	500.89	84.87	39.64	201.33	677.33	8114.55	13.19	1.62	0.29	17.90	83.88
5	485.83	87.59	40.62	197.00	677.33	8477.51	14.44	1.72	0.32	18.60	83.83
6	455.23	76.19	37.10	202.66	680.66	7359.78	12.41	1.59	0.26	16.35	75.99
7	420.00	76.58	37.10	202.00	682.00	6769.34	13.65	1.65	0.25	15.15	71.00

Table 5: Correlation matrix among economic parameters

Fecundity	Hatching	Average larval weight	5 th instar larval duration	Total larval duration	Cocoon yield by No.	Cocoon yield by Wt.	Single cocoon weight	Single shell weight	Shell ratio	Pupation rate	
Fecundity	1.000										
Hatching %	0.706	1.000									
Average larval weight	0.679	0.883	1.000								
5 th instar larval duration	-0.353	-0.458	-0.379	1.000							
Total larval duration	-0.489	-0.505	-0.267	0.658	1.000						
Cocoon yield by number	0.447	0.017	0.015	-0.068	-0.099	1.000					
Cocoon yield by weight	0.502	0.698	0.668	-0.687	-0.475	0.030	1.000				
Single cocoon weight	0.071	0.267	0.107	-0.204	-0.369	0.014	0.055	1.000			
Single shell weight	0.399	0.487	0.409	-0.242	-0.488	0.069	0.134	0.848	1.000		
Shell ratio	0.494	0.488	0.379	-0.277	-0.547	0.044	0.179	0.949	0.750	1.000	
Pupation rate	0.774	0.817	0.853	-0.217	-0.246	0.278	0.554	0.410	0.180	0.419	1.000

Discussions

Success of any breeding programme is dependent on the degree of genetic variability of the available genetic resources and the selection of suitable and genetically diverse parental lines to build heterotic combinations. Determining genetic divergence among the available breeding materials facilitate

this. Genetic divergence is one of the criterion for selection of parents to generate crosses which segregate in later generations into genotypes transcending the performance of better parents (Timoty, 1963) [17]. In present study nineteen silkworm breeds were evaluated to obtain fair and precise information about their comparative performance and genetic

divergence. Results of various economic parameters showed significant difference among the breeds which revealed their genetic potential as well as variability of the breeds. These breeds have immense potentiality as breeding material as they displayed adequate variability for economic traits. Higher the variability more is the scope for obtaining higher amount of heterotic potential and also more variability is expected to surface in segregating generations of such crosses.

Selection of a desirable parent from genetic resources based on their *per se* performance is difficult (Tazima, 1964)^[6], so a unique method based on the genetic divergence *i.e.* clustering was found very useful in choosing the initial parents (Mahalanobis, 1936)^[7]. Many workers (Roa, 2016; Ahsan *et al.* 2000; Farooq *et al.* 2009; Maqbool *et al.* 2015; Rao, 1952; Jolly *et al.* 1989 and Chatterjee and Dutta, 1992)^[13, 1, 5, 11, 14, 6, 3] used clustering for choosing the initial parents in silkworm breeding. Genetic divergence analysis therefore was attempted to identify suitable parents among 19 silkworm breeds by assessing the data recorded from 11 economic traits and were grouped into 7 clusters based on the relative magnitude. The distribution pattern of clusters indicated that the cluster 1st had the largest comprising of five breeds followed by 3rd, 4th, 6th with three breeds each, the 2nd and 5th cluster with two breeds each and the 7th cluster was comprised of only one breed. An interesting observation is that in cluster 3rd all the three breeds evolved by Temperate Sericulture Research Institute Mirgund were clustered together and this cluster showed better performance among all the clusters studied and ranked 1st. These clusters consisted of breeds with both oval and constricted cocoons which clearly showed that breeds having different cocoon shapes may have some similarities with regard to quantitative parameters. The present study is in conformity with earlier findings of Roa (2016)^[13] who reported silkworm races included in a cluster comprised of both oval and spindle shaped cocoons. The clustering pattern in present investigation showed a mixed trend, the silkworm breeds from different geographical origin were included in the same cluster while as breeds from same geographical origin fell in different cluster which reveals that geography has no association with the genetic divergence. In a similar study Jolly *et al.* (1989)^[6], Farooq *et al.* (2009)^[5] reported that silkworm breeds originating from different geographical origin formed same group and those originating from the same region grouped into different cluster. The maximum inter cluster distances suggested wide diversity among the breeds between the groups and the promising breeds from these clusters could be utilized as parents and crossing between these breeds would result in heterotic expression for yield component traits.

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