International Journal of Fauna and Biological Studies
Available online at www.faunajournal.com

ISSN 2347-2677
www.faunajournal.com
IJFBS 2020; 7(4): 172-177
Received: 10-05-2020
Accepted: 12-06-2020

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Multiple trait evaluation index of bivoltine hybrid of silkworm csr2×csr4 under sub-tropical conditions

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Abstract
Feeding is the most important factor in silkworm rearing as it has a direct impact on growth and development of worms. Silkworm requires five hourly feeds a day but the rearers of J&K state feed the worms two times only which results in low cocoon yield. The present study was conducted on authorized commercial silkworm hybrid CSR2 x CSR4, comprising of four treatments viz. single feeding (24 hrs. gap), 2 feedings (12 hrs. gap), 3 feedings (8 hrs. gap) and 4 feedings (6 hrs. gap). Seventeen commercial characters were studied. Data generated was statistically analyzed and also evaluated cumulatively by Manos' Evaluation Index (E.I.) method.

Keywords: *Bombyxmori* L., feeding, mano´s evaluation index, hybrid, economic traits, yield

Introduction
India is a developing country where agriculture supports nearly 70 per cent of the rural population presently the challenge before Indian agriculture is to feed the second most populous nation in the world encounting for about 17 per cent of the world's population supported by only 2.4 per cent of the land area. Inspite of the fact that agriculture mainstay of rural per yet GDP in agriculture continuous to fall which could be attributed to a couple of factors and one among them is continued declined in productivity levels, disproportionate involvement of women in various agriculturally important ventures. In order to contribute to receding income of the farmers, adoption of multidisciplinary income augmenting ventures is of paramount importance which will not only go in a long way to increase the better livelihood earnings but will definitely improve the socio economic status of farmers. One such farmer which not only is women friendly but also fetches quick returns to the farmers comparatively at a greater ease, is sericulture- the practice of silkworm *Bombyxmori L.* rearing for production of cocoons which finally yields splendid fabric of high elegance and durability. In our country sericulture is not only a tradition but also a living culture. This is the only country in the world where all the four types of commercially available silks namely; mulberry, Tassar, Eri and Muga are produced. India also has a distinct advantage of practicing sericulture throughout the year yielding 4-6 crops under its tropical climatic conditions. Sericulture in Jammu and Kashmir has played a vital role in the economy as it provides a source of livelihood to a sizable section of the society. Owing to the highly conducive climatic conditions, Jammu and Kashmir is famous for the production of quality bivoltine silk of optimum quality. It is an income generating agro-enterprise in the mid hilly regions increasing rural women employment and generating their income. This is a potential sector of allied agriculture to raise economic status of the farming community as subsidiary occupation and also generates foreign exchange for the country. Its success mainly depends on the various factors including successful implementation of technological and managerial tools along with high yielding best-suited mulberry and silkworm varieties (Thapa and Shrestha, 1999; Ghimire, 2000) [17, 5]. Evaluation of genetic resources is an essential prerequisite for their effective utilization in order to gauge the extent of variability among genotypes. Silkworms have been evaluated in many environment and agro climatic conditions in order to identify the season and region specific breeds for utilization (Malik *et al*., 2002) [11]. The necessity for identification of season/region specific breeds/ hybrids arises due to variation in quantitative characters during different environmental conditions. The present study was proposed to make a comparative evaluation of feed rationing and its impact on economic characters, and an attempt was made to evaluate the silkworm hybrid combination (CSR2 × CSR4) based on Evaluation index method developed by Mano *et al*. (1993) [12].

Materials and Methods

The seed of silkworm hybrid combination (CSR2 × CSR4) was procured from RSRS, Dehradun during spring- 2017.The experimental research material study comprised of four feeding frequency fed to the worms i.e. one time per day (24 hours), two times per day (12 hours), three times per day (8 hours) and four times per day (5 hours).The experiment was laid out in Completely Randomized Block Design (CRD) with four replications. Three disease free layings of hybrid was incubated for each treatment and reared as per the standard techniques of Dandin *et al.* (2003) [2]. The worms of the three disease free layings were brushed enmass and reared upto third moult. After third moult, the worms were reared in three replicates, taking a population size of 200 worms per replicate at random for each treatment. Ripe worms were picked for seriposition and spinning was conducted on collapsible plastic mountages. The cocoons were harvested on sixth day after mounting. Cocoon samples were taken and stifled in hot air oven at 90 ^{0}C-60 ^{0}C for six hours for reeling purpose. Randomly selected cocoon samples were reeled for post cocoon parameters at Demonstration cum Technical Centre of Central Silk Board, MiranSahib, Jammu. The observations were recorded for different chatacters at egg, larval, cocoon and post-cocoon stage.

3.1.1 Fecundity

It is the total number of eggs laid by a single mother moth and was calculated by counting the total number of eggs laid by a female moth. Average of three layings in each replicate was recorded for analysis purpose.

3.1.2 Hatching percentage

It is the number of larvae hatched out from total eggs laid by a mother moth and was recorded as an average of three layings in each replicate. It was calculated by the following formula:

$$\frac{\text{Number of eggs hatched}}{\text{Total number of eggs per laying}} \times 100$$

3.1.3 Brushing Percentage

It is the total number of larvae brushed out from each disease free laying for each replicate and was recorded in percentage. It was calculated by the following formula:

$$\frac{\text{Number of larvae brushed}}{\text{Total number of eggs per laying}} \times 100$$

3.2.1 IV age larval duration

It is the duration of larvae from III moult out upto initiation of IV moult and was recorded in days and hours for each replicate.

3.2.2 V age larval duration

It is the duration of larvae from IV moult out upto pre-spinning and was recorded in days and hours for each replicate.

3.2.3 Total larval life

It was recorded as an average of total larval life in days and hours from brushing to pre-spinning stage including moulting duration in each instar of each replicate.

3.2.4 Weight of 10 mature larvae (g)

Ten mature larvae were picked randomly from each replicate from 4 to 6 day of fifth instar and weighed using digital balance. The maximum larval weight was recorded in each replicate.

3.2.5 Larval survival percentage

The larval survival percentage represents the number of worms surviving during rearing up to pre spinning stage and was calculated by using the following formula:

$$\frac{\text{Number of larvae surviving at prespinning stage}}{\text{Total number of larvae retained after III moult}} \times 100$$

3.3.1Cocoon yield/10000 larvae

3.3.1.1 By weight (kg)

This parameter was recorded as an average weight of cocoons harvested in kg and converted for 10,000 larvae and was worked out by using the by following formulae:

$$\text{By weight} = \frac{\text{Cocoon yield in kg}}{\text{Total number of larvae retained after III moult}} \times 10{,}000$$

3.3.1.2 By number:

It was recorded as an average number of cocoons harvested and converted for 10,000 larvae and was worked out by using the by following formulae:

$$\text{By number} = \frac{\text{Cocoon yield by number}}{\text{Total number of larvae retained after III moult}} \times 10{,}000$$

3.3.2 Pupation percentage

This parameter represents the average number of live pupae obtained in each replicate and is represented in percentage. It was calculated by using the following formula:

$$\frac{\text{Number of live pupae in harvested cocoon}}{\text{Total number of larvae retained after III moult}} \times 100$$

3.3.3 Good cocoon percentage

Good cocoons were sorted out and counted in each replicate. Average number of reelable cocoons obtained was recorded. It was calculated by using the following formula:

$$\frac{\text{Number of good cocoons harvested}}{\text{Total number of larvae retained after III moult}} \times 100$$

3.3.4 Double cocoon percentage

This represented the average number of double cocoons obtained in each replicate and was determined by using the following formula:

$$\frac{\text{Number of double cocoons harvested}}{\text{Total number of larvae retained after III moult}} \times 100$$

3.3.5 Flimsy cocoon percentage

This parameter depicts the average number of flimsy cocoons counted in each replicate and was computed by using the following formula:

$$\frac{\text{Number of flimsy cocoons harvested}}{\text{Total number of larvae retained after III moult}} \times 100$$

3.3.6 Dead cocoon percentage

This represented the average number of cocoons with dead pupae obtained in each replicate and was calculated by using the following formula:

$$\frac{\text{Number of dead cocoons harvested}}{\text{Total number of larvae retained after III moult}} \times 100$$

3.3.7 Single cocoon weight (g)

Twenty five male and twenty five female cocoons were randomly selected and weighed on digital balance to determine the average cocoon weight by using the following formula:

$$\frac{\text{Weight of 25 male(g)} + \text{25 female cocoon (g)}}{50}$$

3.3.8 Single shell weight (g)

Same twenty five male and twenty five female cocoon shells from each replicate were weighed on digital balance to determine average single shell weight. The formula applied was;

$$\frac{\text{Weight of 25 male(g)} + \text{25 female cocoon shells (g)}}{50}$$

3.3.9 Shell ratio percentage:

It is the average ratio of twenty five male and twenty five female cocoon shell to that of average cocoon weight of same cocoons per replicate and was calculated by using the following formula:

$$\frac{\text{Average weight (g)of 25cocoon shells of each sex}}{\text{Average weight of same cocoons of each sex}} \times 100$$

3.4.1 Total Filament length (m)

Filament length indicates the total reelable length of silk filament obtained from a single cocoon in meters. It is the average length of the silk reeled from a single cocoon.

$$\text{TFL} = \frac{\text{Length of raw silk reeled (m)} \times \text{Number of cocoons maintained per end}}{\text{Number of reeled cocoons} *}$$

*Number of reeled cocoons = Number of cocoons taken for testing – Number of new unreelable cocoons/Number of converted carry over cocoons

3.4.2. Non-breakable filament length (m)

It is a length at which cocoon filament breaks and is replaced by another cocoon. It was recorded as per the following formula:

$$\text{NBFL} = \frac{\text{Length of silk filament reeled} \times \text{No. of cocoons maintained per end}}{\text{No of reeling ends} *}$$

*Indicates number of castings + Number of carry over cocoons – Number of converted carry over cocoon.

3.4.3. Filament size (d)

It was determined by using filament reeled from ten cocoons from each replicate and was calculated by using the following formula:

$$\frac{\text{Weight (g)of raw silk reeled}}{\text{Length (m)of silk reeled} \times \text{No. of cocoons maintained per end}} \text{ x } 9000 \text{ (m)}$$

Evaluation Index

The data was also subjected to Evaluation of Mano *et al.* (1993) [12] performance index which gives a single value measure of the multiple trait performance. The E.I. values were obtained by using the following formula:

$$\text{E. I.} = \frac{\text{A}-\text{B}}{\text{C}} \times 10 + 50$$

Where,

A = Value obtained for a particular trait of the hybrid combinations.

B = Mean value of a particular trait of all the hybrid combinations.

C = Standard deviation of a particular trait.

10 = Standard unit

50 = Fixed value

Results and Discussion

Evaluation indices (EI) assessment is the multiple performance of a population for selection/ short-listing of the genotypes/hybrid combinations by taking into consideration all the economic traits. The data recorded from different traits viz., fecundity, hatching, larval duration, cocoon weight, shell weight, pupal weight, shell ratio, filament length and cocoon yield by number and by weight was subjected to multiple trait evaluation as per the procedure outlined by Mano *et al.* (1993) [12]. Based on the performance the individual indices were calculated for each of the 14parameters. In silkworm *Bombyxmori* L. large numbers of breeds were tested and promising ones were selected based on the economic traits (Lakshmi *et al*., 2012; Gowda *et al*., 2013; Mandal *et al*., 2016 and Gowda *et al*., 2017) [10, 3, 13, 4]. Evaluation index is one such method that increases the precision of selection of breed among an array of breeds by a common index giving due weightage to all the yield component traits (Bhargava *et al*., 1994) [1]. The results obtained during the investigation are presented as under:

Egg Stage

The experimental trial was started after IIIrd moult. However, upto IIIrdage mass rearing was conducted after brushing randomly 4 dfls (disease free layings) for each treatment and average fecundity, hatching percentage and brushing percentage of four treatments was calculated. The E.I. value > 50 was recorded for fecundity with four feeding frequencies was 64.79. The E.I. values for hatching percentage obtained were as 63.61 and 51.48 for single and 2 feeding frequencies respectively. Similarly, E.I. values for brushing percentage recorded was as 61.92 and 54.73 for single and two feedings per day respectively. The average E.I. for fecundity, hatching percentage and brushing percentage observed in single feed treatment was highest at 57.75followed by 4 feeding frequency treatment with EI value at 51.13 surpassing the benchmark of >50 (Table 1).

Table 1: Evaluation Index (E.I.) value of CSR2 X CSR4 bivoltine silkworm hybrid for egg traits.

Frequency of Feeds/day	Fecundity (Nos.)	Hatching percentage	Brushing percentage	Total	Avg. E.I.
1	47.74	63.61	61.92	173.27	57.75
2	43.38	51.48	54.73	149.59	49.86
3	44.73	44.04	39.29	128.06	42.68
4	64.79	41.06	44.56	153.41	51.13

Larval Stage

At larval stage the observations were recorded for IVthand Vth larval age and weight 10 mature larvae. The E.I. value for weight of 10 mature larvae in different feeding frequency treatments recorded was maximum in case of 4 feeding frequency at 56.62 followed by three (55.35) and two feeding frequency (52.83). However, for of larval survival the E.I. value was maximum in case of 2 feeding treatment (57.31) followed by 4 feeding treatment (54.55) and 2 feeding frequency treatment recorded E.I value of 52.90(Table 2). The average E.I. value for weight of 10 mature larvae and larval survival percentage was higher in 4 feeding frequency treatment (55.58) followed by 2 feeding treatment (55.07) and for 3 (54.12) surpassing the benchmark of >50.

Table 2: Evaluation Index (E.I.) values of CSR2 X CSR4 bivoltine silkworm hybrid for larval traits

Frequency of Feeds/day	Weight of 10 mature larvae (g)	Larval Survival (%)	Total	Average
1	35.18	35.24	70.42	35.21
2	52.83	57.31	110.14	55.07
3	55.35	52.90	108.25	54.12
4	56.62	54.55	111.17	55.58

Cocoon Stage

The following observations on cocoon parameters were recorded for different feeding frequency treatments on experimental material of silkworm hybrid CSR2 x CSR4. Silkworm cocoon is the important and economic product of rearing. The manifestations for different parameters of this stage recorded are as under**.** The E.I. analysis for five cocoon yielding parameters *viz.*, cocoon yield per 10,000 larvae by weight, and by number, pupation percentage, single cocoon weight, single shell weight and shell ratio percentage were recorded.

The cocoon yield per 10,000 larvae (by wt.) recorded highest E.I. value of 57.96 for 4 feeding frequency (55.04), for 3 feeding (51.55) for 2 feeding frequencies and least (35.54) in single feed frequency schedule (Table 3).

Maximum E.I. value 56.43 for cocoon yield per 10,000 larvae (by no.) was recorded in 4 feeding frequency treatment followed by 3 feeding frequency regimes (55.24). While as, 2 feeding frequency stood at E.I value of 53.17. Lowest E.I. value of 35.13 was recorded in case of single feed treatment. Maximum good cocoon percentage with E.I value of 57.14 was scored by 4 feeding treatment frequency followed by 3 feeding (54.70) and for 2 feeding (52.93). Lowest E.I. value of 35.22 was observed in case of single feed treatment (Table 3).

For pupation rate, maximum E.I. value of 56.39 followed by (55.10) and (53.39) were recorded for 4, 3 and 2 feeding frequencies respectively. Single feed treatment recorded E.I value of 35.11 and remained at lowest rank. Double cocoon is an undesirable character and maximum E.I. value of 60.27 for this parameter was depicted by 2 feeding frequency followed by 3 feeding treatment (55.72) and 4 feedings (46.63). Lowest E.I. value of 37.54 was recorded in single feed treatment. Flimsy cocoon are also an undesirable character. Higher percentage of flimsy cocoons results in higher cocoon loss. In the present study, single feed treatment exhibited higher flimsy cocoon percentage and it stood with E.I. value of 64.96followed by 2 and 3 feeding frequency (45.76), (45.53) respectively while as least E.I. value of 43.77 was found in 4 feeding frequency (Table 3).

Single cocoon weight isimportant from yielding/reeling point of view. In 4 feeding treatment highest E.I value of 59.33 closely followed by 3 and 2 feeding treatment frequency scored E.I values of (54) and (50) respectively. Least value of (36.00) was recorded in case of single feed treatment. The E.I. value for single shell weight are presented in Table 3 and reveals that 4 and 3 feeding frequency scored maximum E.I. value of 60.00 and 56.66 respectively followed by 2 feeding system with E.I. at 50. Least value of 36.66 E.I. was recorded in case of single feed treatment. Shell ratiobeing most important parameter depicts quality and quantity of actual silk content of a cocoon. Maximum E.I. value of 64.54 was observed in 4 feeding frequency treatment followed by 2 and single feedings which remain at bench mark of 50.

Table 3: Evaluation Index (E.I.) values of (CSR2 X CSR4) of bivoltine silkworm hybrid for cocoon traits

Frequency of Feeds/day	Cocoon yield By Wt.	Cocoon yield By No.	Good cocoon percentage	Pupation Percentage	Double cocoon percentage	Flimsy cocoon Percentage	Single cocoon weight	Single shell weight	Shell ratio %	Total	Avg. E.I.
1.	35.54	35.13	35.22	35.11	37.54	64.96	36	36.66	50	366.16	45.77
2.	51.55	53.17	52.93	53.39	60.27	45.76	50	50	50	467.07	58.38
3.	55.04	55.24	54.70	55.10	55.72	45.53	54	56.66	38.18	470.17	58.77
4.	57.96	56.43	57.14	56.39	46.63	43.77	59.33	60.00	64.54	502.79	62.84

Post Cocoon Stage

Following observation were recorded for different parameters of post cocoon characters.

The Evaluation Index (E.I) value for total filament length was maximum in 4 and 3 feeding frequency as 59.60 and 55.86 respectively followed by 47.44 for 2 feeding frequency.

Whereas, minimum E.I. value of 37.08 was scored by single feed system. Four and three feeding frequency treatments scored maximum non-breakable filament length with E.I. values of 61.64 and 54.88 followed by 2 feedings (43.11). Single feed frequency recorded E.I value of 40.34.

Filament size denotes the thickness/ thinness of the filament. The E.I. value of 46.78was recorded in 4 feeding frequency, 56.42 for 3 feeding, 60 for 2 feeding and 37.50 in single feed system. Among four different feeding treatments the average E.I. value ranged between 55.72 (3 feedings) to 56.00 (4 feedings) as indicated in the (Table 4).

The differential expression of hybrids in the present study is in conformity with the observations of several workers Masarat 2014 [14]; Kumaresan 2007 [6]; Kumaresan 2000 [8] and Mukherjee 2000 [15]. This is largely due to the variable gene frequencies at different loci in different silkworm strains which make them to respond differently to changing environmental conditions Nanjundaswamy 1997 [16] and Kalpana 1992 [7]. It is thus understood that the performance of a race or a breed is mainly dependent on the combined action of hereditary potential of its population and the extent to which such potential is permitted to express in the environment to which it is exposed.In silkworm (*Bombyxmori*L.), selection for various quantitative characters results in change in their mean to a varying degree and the selection for one character is found to produce correlated change in other quantitative characters of economic importance (Kobori and Fujimoto, 1966) [9].

Table 4: Evaluation Index (E.I.) values of (CSR2 X CSR4) bivoltine silkworm hybrid for post cocoon traits

Frequency of Feeds/day	Total filament length (m)	Non-breakable filament length (m)	Filament size Denier	Total	Average
1	37.08	40.34	37.5	114.92	38.30
2	47.44	43.11	60	150.55	50.18
3	55.86	54.88	56.42	167.16	55.72
4	59.60	61.64	46.78	168.02	56.00

Among four feeding frequency treatments for all important commercial cocoon parameters the average E.I. ranged between 55.79 (4 feedings) to 52.24(3 feedings) as indicated in the Table 5.

Table 5: Evaluation Index (E.I.) values of (CSR2 X CSR4) bivoltine silkworm hybrid commercial traits

Feeding Frequency		T-1	T-2	T-3	T-4
		24 hrs gap	12 hrs gap	8 hrs gap	6 hrs gap
Fecundity		47.74	43.38	44.73	64.79
Hatching		63.61	51.48	44.04	41.06
Brushing		61.92	54.73	39.29	44.56
Cocoon yield per 10,000 larvae	Wt.	35.54	51.55	55.04	57.96
	No.	35.13	53.17	55.24	56.43
Good cocoon		35.22	52.93	54.70	57.14
Double cocoon		37.54	60.27	55.72	46.63
Flimsy cocoon		64.96	45.76	45.53	43.77
Pupation		35.11	53.39	55.10	56.39
Single cocoon weight		36	50	54	59.33
Single shell weight		36.66	50	56.66	60
Shell ratio		50	50	38.18	64.54
Total filament length		37.08	47.44	55.86	59.60
Non-breakable filament length		40.34	43.11	54.88	61.64
Filament size		37.50	60	56.42	46.78
Total		654.35	767.21	765.36	820.62
Avg. E.I.		46.73	54.80	54.66	58.61

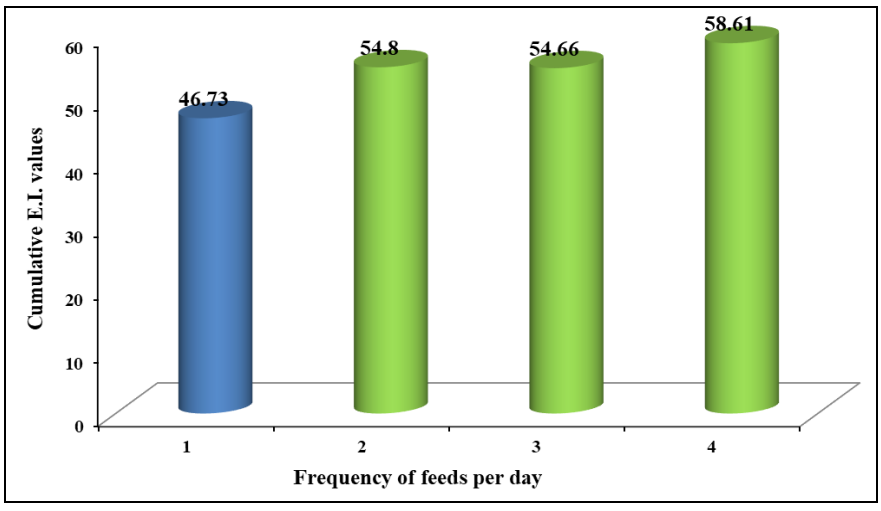

Fig 1: Cumulative Evaluation Index (E.I.) values of bivoltine silkworm hybrid (CSR2 x CSR4) on different feeding schedules per day for commercial traits.

Conclusion

In the present investigation, the fourteen commercial quantitative and qualitative traits revealed broad variability between different feeding frequencies. This may be attributed to the adaptability of silkworm larvae to different feeding frequencies during the rearing of the worms. The present investigation indicates that rearing of silkworm, with feeding frequency of four and three times, is suitable for commercial rearing of bivoltine hybrid.

Acknowledgements

The author is grateful to the Head of Department Division of Sericulture SKUAST-Jammu, for providing facilities and support for carrying out research.

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