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Research Article

EVALUATION OF NUTRITIONAL INDICES AND CONVERSION EFFICIENCY IN MULTIVOLTINE, CROSS-BREEDS AND POPULAR BIVOLTINE DOUBLE HYBRID OF MULBERRY SILKWORM *BOMBYX MORI*

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ABSTRACT

In sericulture, the nutritional ability of mulberry varieties and conversional traits of silkworm breeds have an important role in the improvement of silk yield. The present study aims to evaluate silk conversion efficiency in a few breeds and hybrids. The silkworm *Bombyx mori* breeds and hybrids *viz.*, MVI, PMxCSR2, MV1xS8 and FC1xFC2 were assessed during the fifth instar, with PM as control during the years 2019 and 2020. The study focused on the nutritional indices and conversion traits of silkworms by feeding V1 mulberry leaves, reared under the same environmental conditions. Significant variations were observed among silkworm breeds and hybrids. The test breed and hybrids were found superior to the control PM, which recorded an ingestion of 2.33g and a conversion ability of 8.65%. The bivoltine double hybrid FC1xFC2 showed high food ingestion (3.25g) and conversion (18.18%), followed by the cross breed MV1xS8 with moderate ingestion (3.17g) and conversion (16.63%). The values for ingesta and conversion in PMxCSR2 were 3.43 g and 8.92%, respectively. Further, in respect of Efficiency Conversion of Ingesta (ECI) of shell, the highest conversion of shell is shown by FC1xFC2 (13.56%) followed by MV1xS8 (10.14%), MV1 (9.73%) and PMxCSR2 (9.36%), having a percentage difference of 63.27%, 36.08%, 32.11% and 28.30% respectively over PM. The silk conversion efficiency of bivoltine double hybrid FC1xFC2 was found to be profoundly high, followed by multivoltine cross-breeds and pure line (PM) tested.

Keywords: *Bombyx mori,* Conversion traits, Feed conversion efficiency, Nutritional indices, Silkworms.

INTRODUCTION

The mulberry silkworm, *Bombyx mori* is a monophagous insect that feeds exclusively on the mulberry foliage for its nutrition and produces the natural proteinous fibre. Mulberry, the sole food plant of *Bombyx mori*, which contains all the nutrients for its growth and development (Nasreen *et al*., 1999); plays a pivotal role in the overall development of silkworms, resulting in better cocoon production. The ability of silk synthesis by the silkworm is inferred by the genetic constitution as well as the quality of mulberry feeding (Ramesha *et al*., 2012; Zhang *et al*., 2019) and varies with the mulberry varieties as well as silkworm breeds/hybrids (Kumar *et al.*, 2014). Mulberry nutritional values are augmented in the silkworms' growth through the energy cycles and stored as a reserve for subsequent metamorphic events. They are simultaneously used for the silk synthesis required for spinning the cocoon shell. Through breeding programmes in both mulberry and silkworm, researchers have focused on these aspects and developed molecular introversion for improving silk conversion ability. Thus, nutritional intake has a direct impact on larval, cocoon, silk output, pupation, and reproduction traits.

It is a known fact that in silkworms, quantitative and qualitative traits of cocoons improve with enhanced nutrition (Legay, 1958; Adolkar *et al.,* 2007; Gangwar, 2010; Andadari *et al.,* 2021). For the sustainability of a breed/hybrid in the field, it should be nutritionally efficient as reflected by shell conversion, besides being disease resistant and exhibiting superior survival, yield and quality

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(Ramesha *et al*., 2010). The leaf consumption efficiency of the silkworm depends on the genetic background and varies with the gene expression of physiological or nutritional unit in gene regulation (Giacobino *et al.*, 2003; Milner, 2004; Kang, 2008; Ogunbanwo and Okanlawon, 2009). The introduction of hybrids significantly improved the yield and quality of the silk and brought a new revolution in the sericulture industry. The silkworm breeds/hybrids with improved cocoon yield and superior quality silk made sericulture more sustainable in India (Ramesha *et al*., 2010). Hence, nutritional physiology plays an imperative role in the identification of genomic responses related to nutrient metabolism. Hence, the present study aims to assess the nutritional indices and conversional traits of the mulberry silkworm races and hybrids.

MATERIALS AND METHODS

Silkworm breeds and hybrids

The silkworm pure breed PM (Pure Mysore), MV1 (Improved multivoltine), PMxCSR2 (popular hybrid - Kolar gold) and MV1xS8 (Improved cross-breed Cauvery gold) from Multivoltine Breeding Laboratory, bivoltine double hybrid (FC1xFC2) from Silkworm Seed Production Centre, Mysuru, were used and studied in the Silkworm Physiology Laboratory of Central Sericultural Research and Training Institute, Mysuru.

Silkworm rearing

The rearing activities was carried out as per the standard rearing package (Rajan *et al.,* 2001) with leaves of the mulberry variety V1. All the test breeds/hybrids were reared in a single batch until the resumption of 4th moult in the same environmental condition. In the fifth instar, each batch was divided into two and one maintained as backup, while the second was subjected to standard gravimetric analysis.

Study of nutritional traits

The feed conversion study including nutritional indices and conversional traits were carried out during the fifth instar larvae as major *i.e.,* 80-85% of total leaf consumption takes place in this instar. On resumption from 4th moult (5th instar), 50 healthy silkworm larvae per breed/hybrid were separated with three replications and fed twice a day with known quantities of mulberry leaves. Sample of mulberry fed to the silkworm each day was weighed and dried for determination of ingesta. Simultaneously, additional batches (back up) of larvae for each breed/hybrid were maintained parallelly and the dry weight and the larval growth were recorded as suggested by Maynard and Loosli (1962). The healthy larvae were counted daily in each replication; the unequal, unhealthy and dead larvae were removed, and replaced with the same number of larvae from the backup batch. From day one of 5th instar until spinning the litter was collected on subsequent days at 10 am. The excreta and left-over leaves were separated manually and dried in a hot air oven at 100°C. The cocoons were harvested (on 6th day of spinning), dried and weighed to determine the conversion traits. The data on the dry weight of left-over leaves, excreta, larvae, cocoon and shell in each breed was recorded. The nutritional indices traits such as ingesta, digesta, excreta, approximate digestibility (AD), reference ratio (RR), consumption indices (CI), relative growth rate (RGR), respiration and metabolic rate (MR) and the nutritional conversion traits; efficiency conversion of ingesta (ECI) and digesta (ECD) for larva, cocoon, and shell, ingesta and digesta required for producing one gram of cocoon and shell (I/g and D/g) were calculated as described by standard gravimetric methods (Slansky and Scriber, 1985; Waldbauer, 1968). The data were analyzed using one-way ANOVA.

RESULTS AND DISCUSSIONS

In sericulture, the silkworm breeds and mulberry varieties play an important role in its sustainability. The nutritional efficiency of the mulberry, the digestive and the conversion efficiency of silkworm has collective impact on silk productivity. A large volume of literature is available on the development of breeds in respect of digestibility, disease tolerance, silk productivity, stress tolerance (Basavaraja, 1995; Sudhakar Rao *et al.*, 2001; Ramesh babu *et al*., 2002; Suresh kumar *et al.*, 2003; Ramesha *et al.*, 2012). Similarly, the mulberry varietal influences on the cocoon productivity have been reported by several investigators (Maribhashetty *et al.*, 1991; Raman *et al.*, 1995; Zhang *et al*., 2019). In the present study few multivoltine breeds/hybrids; MVI, PMxCSR2, MV1xS8 and FC1xFC2 were assessed over with PM for their nutrigenic and conversional traits.

Variability was recorded among the test breeds and hybrids in respect of nutritional indices such as ingesta, digesta, excreta, AD, RR, CI, RGR, respiration and MR (Table 1). The capacity to ingest mulberry leaves was lowest in PM (control) followed by MV1, MV1xS8, FC1xFC2 and PMxCSR2. Digesta was lowest in MV1 followed by PM, MV1xS8, FC1xFC2 and PMxCSR2. The lowest excreta found in PM (1.21g/ larvae) and highest in FC1xFC2 (2.35 g/larvae). The AD ranged from 34.08% to 49.00% among the breeds. The RR and CI were lowest in FC1xFC2 followed by MV1xS8, MV1, PMxCSR2 compared to PM. The RGR ranged from 0.13 to 0.17. The highest growth rate observed in the hybrids such as FC1xFC2 and MV1xS8. Respiration was lowest in FC1xFC2 (0.43 g) and highest in PMxCSR2 (0.64 g). Similarly, the MR was lowest in MV1xS8, MV1, FC1xFC2 compared to control (PM).

Analysis of nutritional indices can lead to understanding the physiological basis of insect response to host plants (Lazarevic and Peric-Mataruga, 2003) and the selection of suitable silkworm breeds and mulberry varieties for sustainable silk production. In the present investigation, the ingestion, digestion and assimilation capacity of the silkworms were assessed by their CI and RGR. The duration of the feeding period was found to be an effective factor in the RGR. If the CI of the breed was low, the rate of passage of leaf through the digestive system increased the time for digestion and assimilation and the growth rate was higher (Figure 1). The lowest consumption index and highest growth rate was found in FC1xFC2 followed by MV1xS8 and MV1.

The control Pure Mysore showed lowest values for ingesta and digesta, when compared to the test breed/hybrids (Figure 1). Although many studies have reported higher digesta associated with low ingesta, the present study revealed an improvement in both parameters as indicated by percentage difference in the improved hybrids in respect to ingesta and conversion. The percentage of difference (PD) in respect of ingesta was 5.29% in MV1, 30.43% in MV1xS8, 32.71% in FC1xFC2 and 37.98% in PMxCSR2, over PM. With respect to digesta it was 0.26% in MV1xS8, 12.75% in FC1xFC2 and 18.07% in PMxCSR2. The variations among the silkworm breeds/hybrids (Table 1) were very prominent and the corresponding value of approximate digestibility (AD %) *i.e.*, the feeding frequency was low in FC1xFC2 followed by MV1xS8, MV1 and PMxCSR2.

Table 1. Nutritional indices traits of multivoltine, cross-breeds and bivoltine double hybrid.

Silkworm breed/ hybrid	Ingesta /larvae (g)	Digesta /larvae (g)	Excreta /larvae (g)	AD (%)	RR	CI	RGR	Respiration	MR
PM	2.33 ± 0.05*	1.14 ±0.03*	1.21 ±0.05*	49.00 ±0.34*	1.90 ±0.03*	0.21 ±0.01*	0.13 ±0.01*	0.63 ±0.82 NS	0.04 ±0.01 NS
MV1	2.46 ± 0.02*	1.12 ±0.03*	1.53 ±0.00*	37.49 ±0.87*	1.59 ±0.01*	0.38 ±0.01*	0.15 ±0.01*	0.50 ±0.65 NS	0.02 ±0.01 NS
PMxCSR2	3.43 ±0.03*	1.37 ±0.01*	2.06 ±0.03*	39.96 ±0.56*	1.65 ±0.04*	0.21 ±0.01*	0.13 ±0.01*	0.64 ±0.50 NS	0.03 ±0.01 NS
MV1xS8	3.17 ±0.04*	1.14 ±0.05*	2.02 ±0.02*	35.49 ±0.27*	1.56 ±0.03*	0.17 ±0.003*	0.16 ±0.01*	0.44 ±0.78 NS	0.02 ±0.01 NS
FC1xFC2	3.25 ± 0.20*	1.30 ±0.01*	2.35 ±0.05*	34.08 ±0.51*	1.34 ±0.01*	0.16 ±0.01*	0.17 ±0.10*	0.43 ±0.05 NS	0.02 ±0.01 NS

Values represent the mean, ± standard deviation (SD±) of three separate observations.
*Statistically significant ($P < 0.005$), NS-Statistically not Significant. AD-Approximate digestibility; RR-Reference ratio; CI-Consumption index; RGR-Relative growth rate; MR-Metabolic rate

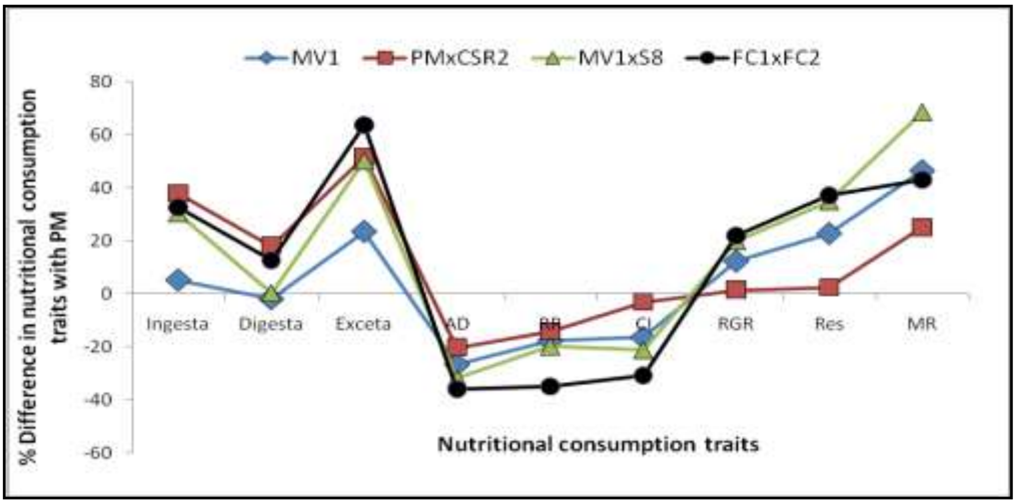

Figure 1. Percentage difference in the nutrition consumption traits of silkworm groups with control (PM).

The lower feeding frequency is positively correlated with the CI and RGR. The lowest the CI, the decline in the PD value 30.68% in FC1xFC2 and 21.31% in MV1xS8 and the improvement in the RGR with the PD value of 22.22% and 19.86% respectively with the PM (Table 1). The results clearly indicate that, the minimum the feeding frequency ultimately reduces the consumption index, improves the absorption of nutrients and growth rate of the silkworm. This is well demarked among the breeds and found better improvement in FC1xFC2 followed by MV1xS8. The parameters such as excreta, RR, respiration and MR are studied as nutrigenic traits and found significant difference with PM in all test groups. The FC1xFC2 has shown comparatively high ingesta and excreta due to its high food intake. The highest RR was found in PMxCSR2 and lowest in FC1xFC2. The MR ranged from 0.02 to 0.04 per larvae and found higher in PM and lowers in MV1xS8.

Silkworm breeding is aimed at improving the genetic entity of silkworms in relation to their economic utility, physiological aspects contribute a lot in the selection of breeds/hybrids (Trivedy and Nair, 1999). Among the different physiological aspects, feed conversion traits that reflect on the silkworm biomass and silk associated with active consumption and conversion of mulberry leaf to proteinaceous silk gland, are considered most important. Significant variations were recorded in respect of nutritional conversion traits such as efficiency conversion of ingesta, and digesta for larva, cocoon, shell and ingesta and digesta required for producing one gram of cocoon and shell (Table 2). The efficiency of mulberry ingested digested and converted to body weight varied prominently among the breed/hybrids. Highest conversion efficiency of ingesta found in FC1xFC2 (18.18%) followed by MV1xS8 (16.63%), MV1 (12.74%), PMxCSR2 (8.92%) compared to control PM (8.65%). Similar trend was observed in ECD to body mass and the range was observed from 18.15% (PM) to 52.65% (FC1xFC2). The conversion efficiency of ingesta and digesta to cocoon and shell was recorded highest in FC1xFC2 followed by MV1x S8, MV1, and PMxCSR2. The highest ingesta to cocoon and shell of 26.75% and 13.56%, respectively, were recorded in FC1xFC2. The ECD to cocoon ranged from 34.52% (PM) to 74.56% (FC1xFC2). Similarly, ECD to shell ranged from 14.77% (PM) to 39.27% (FC1xFC2). The ingesta required to produce 1g of cocoon and shell was also recorded; highest ingesta to cocoon, shell was recorded in PM *i.e.*, 6.07g and 14.18g; lowest ingesta to cocoon, shell of 3.92g and 7.45g was found in FC1xFC2. Similar trend was observed in digesta per g cocoon and shell. The digesta per g cocoon ranged from 1.35g (FC1xFC2) to 2.89g (PM). The digesta per g shell ranged from 2.54g (FC1xFC2) to 6.76g (PM).

Table 2. Nutritional conversion traits of multivoltine, cross-breeds and bivoltine double hybrid.

Silkworm breed/ hybrid	ECI larva (%)	ECD larva (%)	ECI cocoon (%)	ECD cocoon (%)	ECI Shell (%)	ECD shell (%)	I/g cocoon (g)	D/g cocoon (g)	I /g shell (g)	D/g shell (g)
PM	8.65 ±0.24*	18.15 ±0.26*	16.45 ±0.20*	34.52 ±0.76*	7.04 ±0.13*	14.77 ±0.06*	6.07 ±0.07*	2.89 ±0.06*	14.18 ±0.26*	6.76 ±0.03*
MV1	12.74 ±0.17*	35.25 ±0.69*	21.06 ±0.68*	56.12 ±0.43*	9.73 ±0.63*	25.99 ±0.23*	4.75 ±0.15*	1.78 ±0.08*	10.29 ±0.67*	3.86 ±0.33*
PMxCSR2	8.92 ±0.08*	20.39 ±0.15*	21.71 ±1.71*	54.49 ±0.36*	9.36 ±0.06*	24.37 ±1.27*	4.62 ±0.37*	1.84 ±0.15*	10.67 ±0.07*	4.10 ±0.21*
MV1xS8	16.63 ±0.26*	44.36 ±0.59*	21.59 ±0.58*	59.88 ±0.43*	10.14 ±0.69*	28.09 ±0.39*	4.64 ±0.33*	1.68 ±0.17*	9.88 ±0.68*	3.57 ±0.29*
FC1xFC2	18.18 ±0.50*	52.65 ±0.12*	26.75 ±0.19*	74.56 ±1.02*	13.56 ±0.47*	39.27 ±0.81*	3.92 ±0.12*	1.35 ±0.03*	7.45 ±0.25*	2.54 ±0.28*

Values represent the mean, ± standard deviation (SD±) of three separate observations. *Statistically significant ($P < 0.005$). ECI-Efficiency of conversion of ingesta; ECD-Efficiency of conversion of digesta; I/g cocoon-Ingesta per gram cocoon; I/g shell-Ingesta per gram shell; D/g cocoon-Digesta per gram cocoon; D/g shell-Digesta per gram shell.

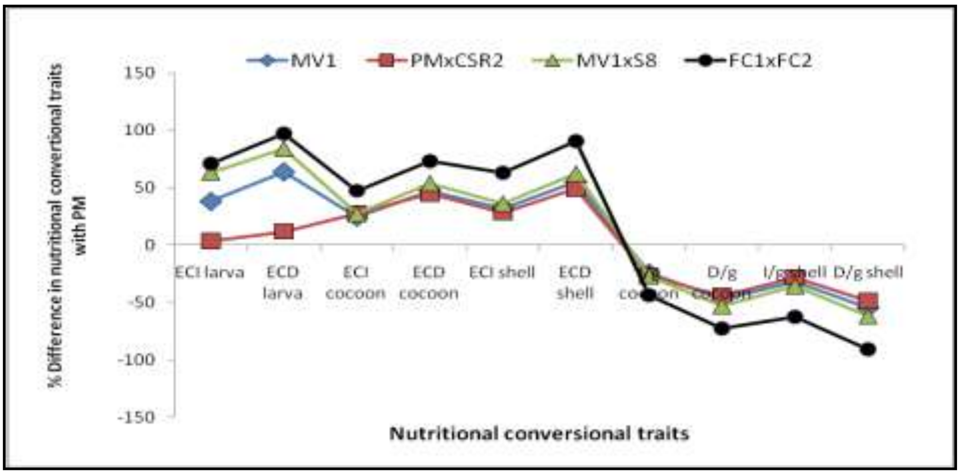

Figure 2. Percentage difference in the nutrition conversional traits of silkworm groups with control (PM).

The silkworms require sufficient nutrients for their growth, development and silk production, which is matched by the food consumed and its conversion during cocooning (Ramesha *et al.,* 2012). The efficiency with which the food substance was ingested and converted to larval body mass was observed with higher percentage difference (PD) of 71.03% in FC1xFC2 followed by MV1xS8 (63.12%), MV1 (38.23%) and PMxCSR2 (3.11%) compared to PM (control). Similarly, in respect of ECD to body, highest PD over PM was recorded in FC1xFC2 (94.46%), followed by 83.85% in MV1xS8, 64.06% in MV1 and 11.65% in PMxCSR2. The ECI cocoon and ECD cocoon also showed large differences among the test breeds/hybrids (Table 2). The highest ECI to cocoon found in FC1xFC2 (47.68%) and lowest by MV1 (24.57%) compared with PM (control). Similar results were obtained in the ECD cocoon and FC1xFC2 found to be more efficient in the conversion traits as shown by PD value of 73.40% followed by MV1xS8 (53.72%).

In silkworm breeding programmes, emphasis is given to the shell ratio, and the efficiency of a breed is always correlated with the higher rate of silk synthesis. The ECI is calculated as the ingesta of the silkworm and its conversion to the shell produced, otherwise known as the leaf shell ratio. This is an ultimate indices in the evaluation of production ability of the silkworm breeds/hybrids and the mulberry varieties for the commercial purposes (Machi and Katagiri, 1991; Maribhashetty et al., 1999; Prabhakar *et al.,* 2000; Ramesha *et al.,* 2010). In the present study, ECI shell ranged from 7.04% (PM) to 13.56% (FC1xFC2); the highest conversion of ingesta to shell was found in FC1xFC2 (PD of 63.27% over that of PM) followed by MV1xS8 (36.08%), MV1 (32.11%) and PMxCSR2 (28.30%) and similar trend was noticed in the ECD of shell.

Further, ingesta and digesta per gram cocoon and shell were inversely proportional to the conversion efficiency of ingesta and digesta (Table 2 and Figure 2). The popular bivoltine double hybrid, FC1xFC2 exhibited higher ingesta and digesta conversion to cocoon and shell with low ingest and digesta per cocoon and shell. The PD value of I/g cocoon was - 43.13% in FC1xFC2 followed by MV1xS8 (-26.67%), PMxCSR2 (-27.19%) and MV1 (-24.37%). Noteworthy is the fact that D/g cocoon also showed the same trend. In PM, the nutrients are being utilized for the growth and development and partially diverted to the silk synthesis as reflected by its nutritional indices and contribution to conversion, which is significantly inferior to evolved breeds and hybrids. Further, the study showed, the ingesta required by PM per shell was 14.18g, which significantly reduced by 62.20% in FC1xFC2 (7.45g), reflecting on the genetic control of the hybrid potential for commercial silk production. Similarly, the cross breed MV1xS8 showed moderately superior traits with 9.88 I/g shell (PD - 35.73%) and PMxCSR2 required 10.69 I/g shell with reduced PD value of 28.22%.

CONCLUSION

The quality and the quantity of silk output are directly related to the nutritional intake in silkworm. The selection of silkworms on their efficient conversion traits *i.e.*, the ones with relatively low consumption of mulberry and higher conversion ratio is highly preferred. The consumption and conversion traits are genetically controlled and physiological studies support to assess the superiority over the existing ones. The present study has revealed the superior conversion traits in the bivoltine double hybrid FC1xFC2 followed by the improved cross breed MV1xS8.

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