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Influence of host plants on biochemical composition in different strains of eri silkworm (*Samia cynthia ricini* Boisduval)

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Abstract
Eri silkworm (*Samia cynthia ricini* Boisduval) is a vital non-mulberry species in Indian sericulture, known for its adaptability to diverse climatic conditions and polyphagous feeding behaviour. The present investigation aimed to evaluate the impact of different host plants (green castor, Pink castor and Tapioca) on the biochemical composition of three strains of eri silkworm *viz*., Yellow Plain, Green Plain and Hybrid (Green × Yellow). Biochemical analysis revealed significant variation in carbohydrate and protein content in the fat body and haemolymph across different treatments. The highest carbohydrate and protein levels were recorded in larvae reared on Local Green + Local Pink and Local Green + Local Green combinations. However, correlation analysis indicated that these biochemical parameters had weak and mostly non-significant relationships with economic traits. These findings suggest that while host plant combinations can alter the biochemical profile of eri silkworms, their direct influence on commercial traits remains limited.

Keywords: Biochemical composition, castor, carbohydrate, ERI silkworm, fatbody, host plant haemolymph, protein, *Samia cynthia ricini*, tapioca

Introduction
Eri silkworm (*Samia cynthia ricini* Boisduval) is a robust, domesticated non-mulberry silkworm species extensively reared in the northeastern states of India, particularly Assam, Nagaland and Manipur. India stands as the second-largest silk producer globally, with a total raw silk production of 36,582 MT with 7,349 MT eri raw silk (Anonymous, 2023) [1]. Renowned for its adaptability to diverse climatic conditions and resistance to many diseases, eri silkworm plays a vital role in sustainable sericulture practices. The primary host plant of eri silkworm is castor (*Ricinus communis* L.), owing to its superior foliar nutritional profile and ease of cultivation. However, other plants such as tapioca (*Manihot esculenta*), papaya (*Carica papaya*) and champa (*Plumeria acutifolia*) also serve as alternate hosts (Singh and Das, 2006) [11]. The growth, development and economic traits of eri silkworms are significantly influenced by the type and quality of host plants consumed during larval stages. The nutritional quality of the host plant not only affects growth and productivity but also plays a vital role in shaping the internal physiology of silkworms, especially their biochemical composition (Kedir Shifa *et al.*, 2014) [4].
Biochemical constituents such as carbohydrates and proteins in the fat body and haemolymph are essential indicators of the silkworm's physiological and metabolic status. These biomolecules play crucial roles in energy storage, tissue development, silk protein synthesis and immune functions. The quality of larval nutrition directly affects these biochemical profiles, which in turn can impact not only growth but also disease resistance and silk productivity (Manjunath and Sannappa, 2014) [7]. Understanding these biochemical changes provides deeper insights into how eri silkworm strains physiologically respond to different host plant combinations (Longvah *et al.*, 2011) [5]. Such information is important for optimizing feeding strategies and selecting suitable host-strain pairings that could enhance rearing success and economic returns. In this context, the present investigation was undertaken to evaluate the impact of selected host plant combinations on the biochemical composition of eri silkworm strains.

Materials and Methods

The present investigation was carried out at the Department of Studies in Sericulture Science, University of Mysore, Manasagangotri, Mysuru, to study the influence of host plants on the biochemical composition of eri silkworm strains. The experiment involved three eri silkworm strains *viz*., Green Plain, Yellow Plain and their hybrid (Green × Yellow) which were reared on three different host plant combinations. The host plants included Local Pink castor (*Ricinus communis* L.), Local Green castor (*Ricinus communis* L.) and Tapioca (*Manihot esculenta*). Freshly harvested leaves were used for feeding the larvae throughout the experiment.

Eri silkworm cocoons were procured from the College of Sericulture, University of Agricultural Sciences, Chintamani and incubated under controlled conditions of 25-28 °C and 60-80% relative humidity (RH). Prior to rearing, the rearing house and appliances were thoroughly cleaned and disinfected with a 5% bleaching powder solution as per standard disinfection process (Dandin and Giridhar, 2014) [2]. Rearing was carried out following the standard procedure (Dayashankar, 1982) [3]. During the chawki stage (up to the 3rd instar), all silkworm strains were fed with Local pink castor leaves. For the late-age worms (4th and 5th instars), larvae were fed with three host plants. Treatment details were given in table 1. Each treatment included 50 larvae per replication with three replications.

Table 1: Treatment details

Late age silkworms (4th and 5th Instar)			
Strains	**Host plants**		
	T_1	**T_2**	**T_3**
Green	Green castor variety	Pink castor variety	Tapioca
Yellow			
Hybrid (Green X Yellow)			

Biochemical analysis was carried out on the fifth day of the fifth instar. Haemolymph and fat body samples were collected and preserved at-20 °C for the estimation of carbohydrate and protein content. Carbohydrate content in both tissues was determined using the Anthrone method (Sadasivam and Manickam, 2008) [8]. Fat body samples (0.1 g) were homogenized in 10 ml of trichloroacetic acid (TCA) and centrifuged at 3000 rpm for 20 minutes. One milliliter each of haemolymph and fat body extract was mixed with 4 ml of Anthrone reagent and boiled for 8 minutes. Distilled water was used as a blank in place of the supernatant. The pale green colour intensity was measured using a spectrophotometer at 630 nm and the results were interpreted using a standard graph and expressed as mg/ml for haemolymph and mg/g of wet tissue for fat body.

Protein content was estimated using the Lowry's method (Lowry *et al.*, 1951) [6], with absorbance recorded at 660 nm. at body samples (0.1 g) were similarly homogenized in 10 ml of TCA and centrifuged at 3000 rpm for 20 minutes. One millilitre of each haemolymph and fat body sample was added with 5 ml of protein reagent and 0.5 ml of Folin's reagent (1:1) and incubated for 30 minutes at room temperature. Distilled water served as the blank, and Bovine Serum Albumin (BSA) at 0.75 mg/ml was used as the standard. The resulting blue colour intensity was read at 660 nm using a spectrophotometer, and the results were expressed as mg/ml for haemolymph and mg/g of wet tissue for fat body. The data were statistically analysed using a two-way factorial completely randomized design (CRD) at a 5% level of significance. All statistical analyses were carried out using the OPSTAT software (Sharon, 1999).

Results and Discussion

The biochemical composition of eri silkworms was assessed by estimating carbohydrate and protein content in the fat body and haemolymph across different host plant combinations and silkworm strains.

Carbohydrate Content in Fat Body and Haemolymph

The total carbohydrate content in both fat body and haemolymph of eri silkworms was significantly influenced by the host plants, while the effect of silkworm strains and their interaction was statistically non-significant (table 2, fig.1). In the fat body, the highest carbohydrate content was recorded in silkworms reared on Local Green + Local Green castor (1.282 mg/g), whereas the lowest was observed in those fed with Local Green castor + Tapioca (1.074 mg/g). Among the eri silkworm strains, the Green strain exhibited the highest carbohydrate content (1.221 mg/g) and the hybrid strain showed the lowest (1.193 mg/g). In the interaction effects, the maximum carbohydrate content was noted when the Green strain was reared on Local Green + Local Green castor (1.290 mg/g), while the minimum was found in the Hybrid strain reared on Local Green castor + Tapioca (1.067 mg/g).

In the haemolymph, carbohydrate content also varied significantly among the host plants. The highest value was recorded in Local Green + Local Pink castor (1.652 mg/ml) and the lowest in Local Green castor + Tapioca (1.359 mg/ml). Among the strains, the Yellow strain had a slightly higher carbohydrate content (1.524 mg/ml), while the Green strain had the lowest (1.506 mg/ml). With respect to interaction, the highest carbohydrate content was observed in the Hybrid strain reared on Local Green + Local Pink castor (1.670 mg/ml), whereas the lowest was again found in the Hybrid strain reared on Local Green castor + Tapioca (1.343 mg/ml).These findings suggest that Local Green + Local Green and Local Green + Local Pink castor combinations support higher carbohydrate accumulation in eri silkworm tissues, possibly due to their superior foliar nutritional composition.

Protein Content in Fat Body and Haemolymph

The total protein content in eri silkworms showed significant variation among host plants, while the effects of strains and their interaction were statistically non-significant in most cases (table 3, fig. 2). In the fat body, protein content varied significantly with host plant combinations. The highest protein content was recorded in silkworms reared on Local Green + Local Pink castor (1.418 mg/g), and the lowest was observed in Local Green castor + Tapioca (1.021 mg/g). Among eri silkworm strains, the Yellow strain showed slightly higher protein content (1.278 mg/g), followed closely by the Green strain (1.270 mg/g). In terms of interaction effects, the maximum value was found in the Yellow strain reared on Local Green + Local Pink castor (1.433 mg/g), whereas the minimum was in the Hybrid strain reared on Local Green castor + Tapioca (1.007 mg/g).

In the haemolymph, total protein content also differed significantly among host plants, while the effect of strains and interaction was relatively minimal. The highest protein content was observed in silkworms fed on Local Green + Local Green castor (1.614 mg/ml), and the lowest in Local Green castor + Tapioca (1.251 mg/ml). Among the strains, the Hybrid strain recorded the highest protein content (1.502 mg/ml), while the Yellow strain showed the lowest (1.480 mg/ml). In the interaction effects, the highest value was found in the Hybrid strain reared on Local Green + Local Pink castor (1.647 mg/ml), and the lowest in the Yellow strain reared on Local Green castor + Tapioca (1.243 mg/ml).

Carbohydrates serve as a major energy reserve and play an essential role in metabolic activities, while proteins are crucial for tissue development, silk gland function and enzyme activity. Total carbohydrate content in fat body and haemolymph did not vary significantly with host plants, eri silkworm strains or their interaction. However, Yellow strain larvae reared on Local Pink + Tapioca recorded higher carbohydrate content, while the Hybrid strain reared on Local Pink + Local Green stood best in fat body. In haemolymph, higher carbohydrate content was observed in Hybrid strain reared on Local Pink + Tapioca, followed by Local Pink + Local Green castor. Total protein content in the fat body differed significantly among host plants, though strain and interaction effects were non-significant. Highest protein content was found in Yellow strain reared on Local Pink + Local Pink and Hybrid strain on Local Pink + Local Green. In haemolymph, no significant differences were observed, yet higher protein levels were noted in Hybrid strain reared on Local Pink + Tapioca and Local Pink + Local Pink. The biochemical composition of eri silkworms varied notably with different host plant combinations. These results align with Manjunath and Sannappa (2014) [7], who reported higher protein, carbohydrate and amino acid contents in Yellow-Plain strain compared to others. Pallavi and Sannappa (2019) [8] also highlighted the role of biochemical composition of castor leaves, especially from DCH-177 and Local Pink, in enhancing bioenergetic indices and economic traits in eri silkworms.

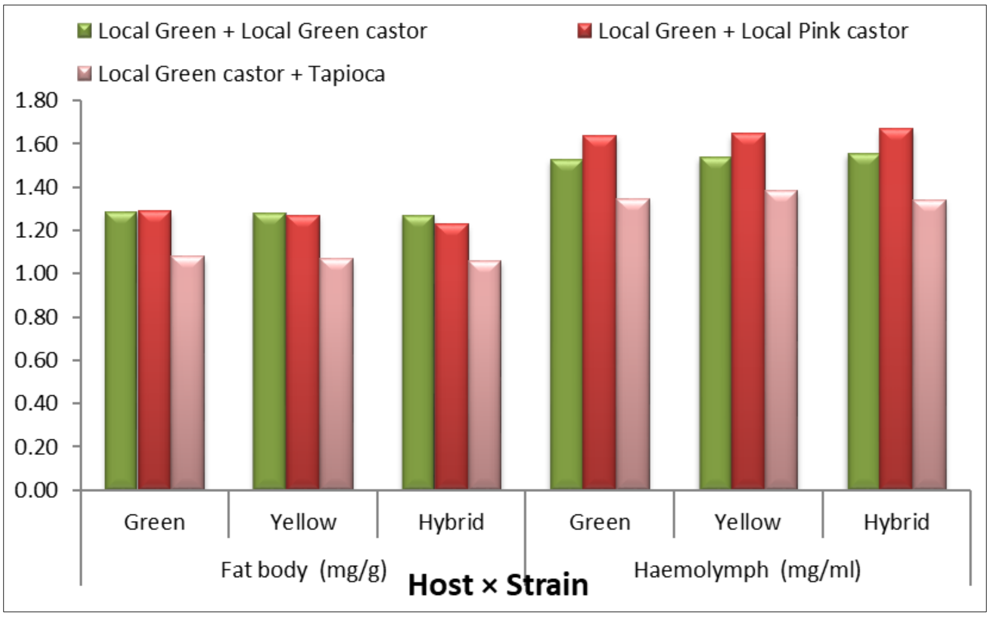

Fig 1: Carbohydrate content in fat body and haemolymph among three strains of eri silkworm reared on different host plants

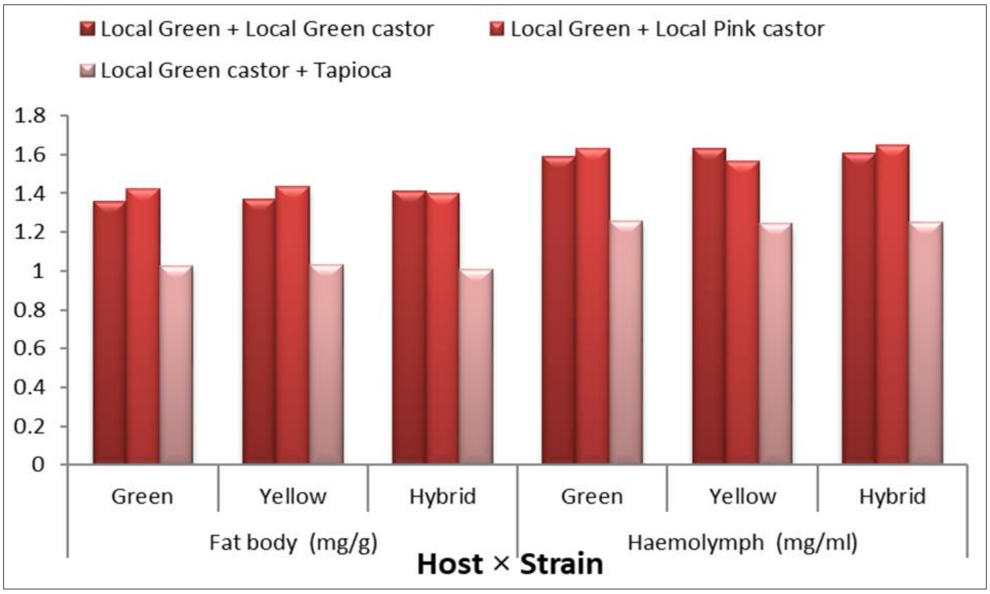

Fig 2: Protein content in fat body and haemolymph among three strains of eri silkworm reared on different host plants

Table 2: Carbohydrate content in fat body and haemolymph among different strains of eri silkworm reared on different hosts

Host (Young-age + Late-age)	Fat body (mg/g)				Haemolymph (mg/ml)			
	Strain				Strain			
	Green	Yellow	Hybrid	Mean	Green	Yellow	Hybrid	Mean
T_1. Local Green + Local Green castor	1.290	1.280	1.277	1.282	1.530	1.540	1.560	1.543
T_2. Local Green + Local Pink castor	1.290	1.273	1.237	1.267	1.640	1.647	1.670	1.652
T_3. Local Green castor + Tapioca	1.083	1.073	1.067	1.074	1.347	1.387	1.343	1.359
Mean	1.221	1.209	1.193		1.506	1.524	1.524	
Two-Way ANOVA	SE(m)±	SE(d)±	C.D. at 5%		SE(m)±	SE(d)±	C.D. at 5%	
Host	0.029	0.041	0.086		0.023	0.032	0.067	
Strain	0.029	0.041	--		0.023	0.032	--	
Host X Strain	0.050	0.070	--		0.039	0.055	--	

Table 3: Protein content in fat body and haemolymph among three strains of eri silkworm reared on different host plants

Host (Young-age + Late-age)	Fat body (mg/g)				Hemolymph (mg/ml)			
	Strain				Strain			
	Green	Yellow	Hybrid	Mean	Green	Yellow	Hybrid	Mean
T_1. Local Green + Local Green castor	1.360	1.370	1.413	1.381	1.587	1.633	1.607	1.609
T_2. Local Green + Local Pink castor	1.423	1.433	1..397	1.418	1.633	1.563	1.647	1.614
T_3. Local Green castor + Tapioca	1.027	1.030	1.007	1.021	1.257	1.243	1.253	1.251
Mean	1.270	1.278	1.272		1.492	1.480	1.502	1.270
Two-Way ANOVA	SE(m)±	SE(d)±	C.D. at 5%		SE(m)±	SE(d)±	C.D. at 5%	
Host	0.016	0.022	0.047		0.023	0.033	0.069	
Strain	0.016	0.022	--		0.023	0.033	--	
Host X Strain	0.027	0.039	--		0.040	0.057	--	

Conclusion

The present study highlights that host plant combinations significantly affect the biochemical composition of eri silkworms. Among the tested combinations, Local Green + Local Pink and Local Green + Local Green castor leaves promoted higher accumulation of carbohydrates and proteins in both the fat body and haemolymph. These biochemical variations, although crucial for larval physiology, exhibited weak correlations with economic traits, suggesting that internal nutrient buildup alone may not guarantee improved silk productivity. The results reinforce the importance of host plant selection for optimal silkworm health and hint at the need for integrated approaches combining nutritional, genetic, and environmental factors to enhance overall performance in eri silkworms.

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Authors Contribution

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Disclaimer (artificial intelligence)

Author(s) hereby declares that no generative AI technologies such as large language models (ChatGPT, Copilot, etc) and text-to-image generators have been used during writing or editing of manuscripts.

Declaration

The authors declare that they have no conflict of interest.

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