



Enhancing Silk Production: Impact of Milk Fortified Mulberry Feed on Silkworm (*Bombyx mori* L.) Growth and Cocoon Quality

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Authors' contributions

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

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ABSTRACT

Enhanced silk production measures directly contributes to the economic upliftment of farmers engaged in Sericulture. Fortification of mulberry leaves to enhance silk production in silkworms is a promising approach to boost the quality and quantity of silk. By enriching mulberry leaves with additional nutrients such as proteins, vitamins, and minerals, the overall health and development of silkworms can be significantly improved. Therefore, this study investigates the efficacy of fortified mulberry feed on the growth and cocoon quality of *Bombyx mori* L. by using commercially available milk samples. The research aims to enhance the nutritional status of mulberry leaves through milk

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fortification to improve the biological characteristics and silk production of silkworms. Various milk samples, including Aavin, Arokya, and Bovine, were used to fortify the mulberry leaves fed to the monophagous silkworm larvae. The results of the present study demonstrated a significant increase between the control and the experimental setups—MST 1, MST 2, and MST 3. Notable improvements were observed in cocoon weight, pupal weight, and shell weight across the experimental categories compared to the control, with values recorded as follows: cocoon weight (1.15 ± 0.10 to 1.25 ± 0.14), pupal weight (0.86 ± 0.13 to 0.96 ± 0.11), and shell weight (0.21 ± 0.07 to 0.27 ± 0.23). These findings highlight the efficacy of milk fortification in significantly enhancing cocoon production, highlighting its potential to improve the efficiency and productivity of the sericulture industry. Further, these findings suggest that milk fortification of mulberry leaves could be a practical and effective method to boost silk production and improve the livelihoods of farmers engaged in sericulture.

Keywords: Mulberry; *Bombyx mori*; silk production; milk fortification; sericulture.

1. INTRODUCTION

Mulberry belongs to the genus of flowering plants in the family Moraceae, which consists of diverse species of deciduous trees commonly known as mulberries, originating from Asia. This plant has been cultivated for over 4,500 years in the temperate regions (Ruiz-Eraza et al., 2023). The *Bombyx mori* L is a phytophagous feeder on *Morus alba* L. (mulberry leaves). Scientists have tried to identify a suitable and an alternative host for the rearing of silkworm, but they were not cost effective. Based on the quality, mulberry has a great influence on the silkworm growth (Ravikumar, 1988). The overall development of silkworm larvae depends on getting the required food which can be easily digested and assimilated in the body tissues properly for the silk synthesis. Keeping in view, the above facts, an attempt was made to increase the growth of young ages of silkworm and cocoon yield using suitable mulberry variety and food additives (Salimath et al., 2007). Because, nutrition in the silkworm, *B. mori* L directly influences the quality and quantity of its silk production (Benjamin and Jolly, 1986). The supplementation or fortification of mulberry leaves is a recent technique in sericulture research to try achieving the said target of maximizing the yield (Murugan et al., 1998). Many attempts have been made by many workers to increase the nutritional quality of mulberry leaves by fortification of mulberry leaves, using plant extract, beneficial effects of chemicals (Bhattacharayya and Medda, 1981, Bhattacharayya and Medda, 1983, Mujamdar, 1982, Gomma et al., 1976), hormones (Akapanthu, 1987, Garel, 1983, Singh, 1991) and plant growth regulators (Agarwal, 1984, Kamda and Ito, 1984). The soybean flour proved to be a good supplement for getting quality cocoons. Konala et al., (2013) had suggested that *B. mori*

larvae can be fed with mulberry leaves treated with bovine milk for better growth rate and increased silk production. Enriching mulberry leaves by nutrient supplementation is one of the ways to improve growth rate in *B. mori* (Konala et al., 2013). The effect of mulberry leaves enriched with amino acids on the growth of *B. mori* has been studied by Radjabi (2010). The mulberry silkworm, *B. mori* is reared successfully as the main source of natural silk. Recently, considerable attention has been given to improve rearing techniques of silkworms to increase the production of raw silk in Egypt to meet with the higher demands for industrial purpose. Furthermore, developing and improving the practical and applicable techniques for increasing the productivity of silkworm especially for silk and eggs production is necessary. So, the nutritional studies on the silkworm, *B. mori* L. are of much importance pertaining to its productivity and the nutritional value of local indigenous plants, wild herbs and edible seeds rich in protein are of great significance. Therefore, it has been reported recently that better production of cocoon crops and eggs is possible when mulberry leaves are supplemented with certain nutritional materials (Rouhollah, 2010, Singh et al., 1993, Zannoon, 1994, Ashour, 1997, El-Sayed et al., 1998). The larval growth and production of silk by silkworm depends greatly on the nutritional composition and quality of mulberry foliage offered to the silkworms (Mesbah et al., 2000, Seidavi et al., 2005, Kantwa et al., 2006). and the Mulberry plants play a major role in the progress of sericulture industry and in turn cocoon and silk production (Manjula et al., 2011). Honey contains bioactive components in the form of proteins, carbohydrates, free amino acids, trace quantity of vitamins, and metals (Nagaraju, 2002, Falco et al., 2003, Garcia et al., 2005, Ball, 2007). It enhances silk productivity and quality and

reduces floss output which is treated as the sericultural wastage (Moniruzzaman et al., 2014). Hence the research was planned with a defined objectives to understand the significant role of feed fortification especially in *Morus alba*.

2. MATERIALS AND METHODS

2.1 Preparation of Rearing Room

The rearing room and the equipment were thoroughly cleaned, including washing the floor with 5% bleaching powder solution and disinfecting the entire room with 2.5% Sanitec solution in 0.5% slacked lime solution before initiating the rearing processes (Thulasi and Sivaprasad, 2015). The rearing environment was maintained at an optimal temperature of 25±1°C and a relative humidity of 75±5%. The rearing trays were lined with paraffin paper at the bottom and wet foam rubber strips at the edges for optimal conditions (Dandin et al., 2003).

2.2 Silkworm Feeding Frequency

The silkworms were fed with fresh mulberry leaves (MR2) during the first to fourth instar stage without any treatment. The leaves were harvested in the early morning and late evening hours; washed and were wiped to remove the excess water. Freshly harvested mulberry leaves were only given as feed till the entire course of the study to observe the growth (Benjamin and Jolly, 1986).

2.3 Rearing Modality

The study involved in rearing the mulberry silkworm *Bombyx mori* L. (FC1XFC2), under controlled conditions. The rearing room and the equipment were thoroughly cleaned, including washing the floor with 5% bleaching powder solution and disinfecting the entire room with 2.5% Sanitec solution in 0.5% slacked lime solution before initiating the rearing processes (Dandin et al., 2003) The rearing trays were lined with paraffin paper at the bottom and wet foam rubber strips at the edges for optimal conditions (Benjamin & Jolly 1986).

2.4 The Experimental Setup

Silkworms of the desired age class was taken for the present study under three different experimental categories as experimental set up 1 with Milk Sample Test – MST1, experimental set

up 2 with Milk Sample Test – MST2 and experimental set up 3 with Milk Sample Test – MST3 along with the control set up to compare the significant differences. MST1 is a combination of the commercially available Aavin milk taken for the fortification study, while MST2 and MST3 are the combination of the commercially available milk samples, Arokya and Bovine milk respectively.

List 1. Experimental set up

	Exp. Setup 1	Exp. Setup 2	Exp. Setup 3	Control Set up
Categories	MST1	MST2	MST3	CST
	Aavin fortified mulberry leaves	Arokya fortified mulberry leaves	Bovine milk fortified mulberry leaves	Plain mulberry Leaves Without feed fortification

2.5 Statistical Analysis

The values from all the experiments of the present investigation were obtained in triplicates and were expressed as mean with standard error in tables and figures. All the analyses were carried out using MS excel and Statistical Package for Social Sciences (SPSS) software package for windows (version 22.0).

3. RESULTS AND DISCUSSION

This research study is attempted to test the Efficacy of Fortified Mulberry Feed on the Growth and Cocoon Quality of *Bombyx mori* L. Three experimental setups with three different composition of milk samples namely MST1, MST2, and MST3 were run to monitor and compare with the control group. Silkworm larvae were fed with mulberry leaves fortified with three different milk samples that are commercially available. Weight of the larva under treatment was monitored at a regular interval and it was recorded to be gradually increasing. The study further discusses about feeding *Bombyx mori* larvae with mulberry leaves fortified with milk sample to enhance their biological characteristics due to the rich milk nutrients, such as proteins and lipids, which can boost the larval growth and development. It was also studied by various researchers and observed that casein, found in milk, contains essential nutrients like fatty acids, cholesterol, sugars, vitamins, and minerals, which can improve the feeding efficiency of *Bombyx mori* larvae. This research study too

revealed a significant result in terms of differences in cocoon weight, pupal weight and shell weight when compared to the control ($1.15 \pm 0.10 \pm 1.25 \pm 0.14$), ($0.86 \pm 0.13 \pm 0.96 \pm 0.11$) and ($0.21 \pm 0.07 \pm 0.27 \pm 0.23$).

3.1 Effect of Different Milk Samples on Cocoon Quality

The primary food source for silkworms is mulberry leaves, and the nutritional content of these leaves can impact considerably on the cocoon quality. Fortification of mulberry leaves with milk had significantly enhanced the cocoon quality with constructive effect. The cocoons harvested from the experimental units were found to be bright white and sturdy in nature. The results in terms of the weight of the cocoons produced show variations for the three different experimental set ups from the control as (control - 1.5 ± 0.10 ; MST 1 - 1.17 ± 0.18 ; MST 2 - 1.20 ± 0.13 and MST 3 - 1.25 ± 0.14). Further it is inferred from the findings that fortifying bovine

milk with mulberry leaves can improve the cocoon weight, which in turn can increase the quality cocoon production.

3.2 Effect of Different Milk Samples on Pupal Weight

The study reveals that the larva is gradually transformed into a pupa during the transformative developmental process and it occurs with a change in considerable weight gain of the pupa inside the cocoon during spinning. Therefore, pupal weight plays a crucial role in determining the weight of the cocoon in the transformative weight change from larva to pupa and cocoon. It is recorded that in the control unit, it was nearly 58% of the total body weight of the larva which is converted into pupa and whereas in the MST 3 only 53% of the body weight is converted into pupa. This is a positive result which could maximally increase the cocoon and the shell weight respectively.

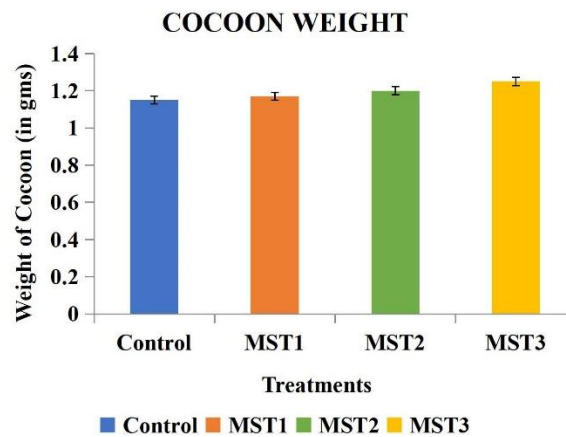


Fig. 1. Comparative growth chart of larval weight under different experimental setup (Control, MST1, MST2 and MST3)

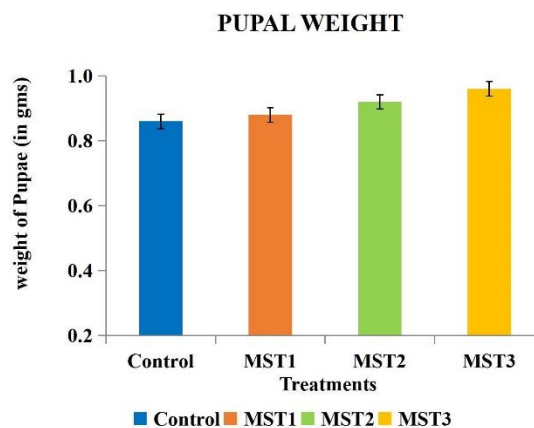


Fig. 2. Comparative pupal weight under different experimental setup (Control, MST1, MST2 and MST3)

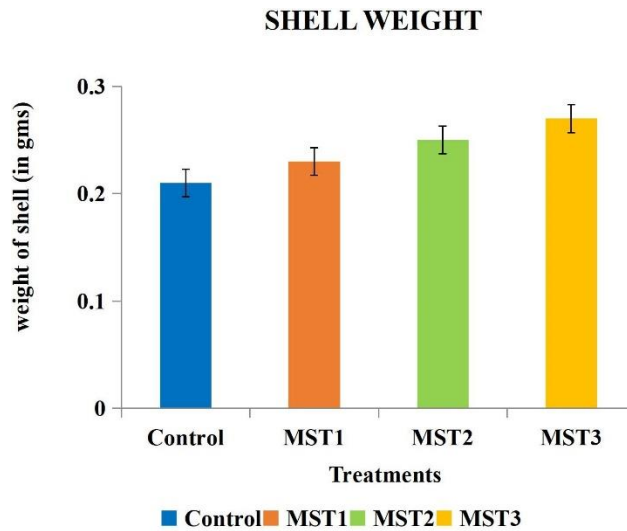


Fig. 3. Comparative shell weight under different experimental setup (Control, MST1, MST2 and MST3)

3.3 Effect of Different Milk Samples on Shell Weight

Shell weight is one of the vital indexes which indicates the quantity of raw silk that can be reeled from the fresh cocoons. The shell weight normally ranges from 0.45g to 0.5g as specified by the Directorate of Sericulture, Assam. The results explain clearly that the milk sample treatment MST3 meets the requirements of the same discussed above (MST 3 - 0.27 ± 0.23) over the control set up.

3.4 Effect of Different Milk Samples on the Growth of Larva

The larval growth and the significant differences in the weight of the larva under different milk samples were found to be showing a distinctive

result for the three experimental setups as against the control. It is observed that the fifth instar larvae fed with mulberry leaves fortified with bovine milk sample (MST3) had significantly gained weight over the period of growth when compared to those fed with fresh mulberry leaves (control) as well as the leaves fortified with other milk samples; MST1 and MST 2 respectively. It indicates that the bovine milk can accelerate the growth rate and thus the silk production of *Bombyx mori* larvae considerably. It is further observed that during the Vth stage of larval growth, there is a gradual improvement in weight of the larva in all the three samples of milk (MST1, MST2 and MST3) from day 1 to day 9 when compared to the control. Among the three samples taken for the study, MST3 (Bovine Milk) showed a significant variation as 0.56 ± 0.14 gms (Day 1) to 3.24 ± 0.35 grams. (Day 9).

Table 1. Comparative growth chart of larval weight (Vth Stage) under different experimental setup (Control, MST1, MST2 and MST3)

Days	LARVAL WEIGHT			
	Control	MST1	MST2	MST3
Day 1	0.44±0.90	0.52±0.10	0.45±0.90	0.56±0.14
Day 2	0.59±0.99	0.89±1.44	0.89±0.79	0.93±0.11
Day 3	0.63±0.07	0.68±0.15	0.70±0.08	0.78±0.11
Day 4	0.88±0.19	1.14±0.18	1.25±0.39	1.32±0.30
Day 5	1.30±0.25	1.36±0.24	1.40±0.32	1.47±0.29
Day 6	1.80±0.26	1.91±0.38	1.99±0.41	2.15±0.31
Day 7	2.44±0.31	2.51±0.46	2.61±0.26	2.65±0.21
Day 8	2.60±0.47	2.63±0.37	2.83±0.27	2.86±0.44
Day 9	2.63±0.42	2.77±0.35	2.82±0.30	3.24±0.35

A successful cocoon production in sericulture depends mostly on healthy larval growth. It is observed that nickel chloride can be used at low concentrations for enhancing the economic traits of *Bombyx mori* larvae. According to Bentea et al. (2011) in the last few years, greater importance has been given to the enhancement of nutrition to *Bombyx mori* larvae (Bentea et al., 2011).

The results of this study agree with the earlier studies of bovine milk as a growth promoting supplementation (Loh and Maznah, 1999, Black et al., 2002). The larvae may have gained weight because of nutrient-rich milk, but the exact cause of weight-gain needs to be investigated in further experiments. The previous study conducted to test the Impact of yeast extract on the economic traits of silkworm, *Bombyx mori* L. The mulberry leaves tested with yeast extract (YES1, YES 2) were fed to silkworm larvae. The yeast extract was found to be giving a significant result in the weight of the larvae, cocoon and shell weight when compare to the control group. Therefore, the conclusion of in this study yeast extract is positively and significantly yielding the result (Nivetha et al., 2024). The result of this study fortification of mulberry leaves by using supplementary nutrient and feeding to the silkworms is a useful modern technique to increase economic value of cocoon (Etebari et al., 2004). The previous study investigation concluded that two probiotics namely spirulina and yeast significantly promote the cocoon characters and silk quality, when compared to *Lactobacillus acidophilus* and *Lactobacillus sporogens* (Masthan et al., 2017). The result of this study can be a desirable intervention for the sericulturists and can play an important role to get high return from silkworm-rearing activities (Bhatti et al., 2019). Rahmathulla V. K. et al., 2007 in his research suggested that oral administration of folic acid during 5th instar silkworm significantly influences the growth rate pattern of silkworm larva and silk gland. This higher growth and development also influence the economic characters like cocoon weight, shell weight and shell ratio and subsequently quality of silk. The study indicated that the plant extracts exhibit the presence of certain growth stimulants and can be used to increase the silk yield in commercial silkworm rearing (Rahmathulla et al., 2007). The *A. longifolia* Ner at 1:50 concentration fed

daily recorded higher food consumption and utilization as well as the cocoon parameters from all the treatments so as the *A. longifolia* Ner leaf extract at 1:50 concentration daily. One feeding may be recommended to the sericulturists to promote the economic characters like cocoon weight, shell weight, shell ratio, filament length, filament weight and denier (Jadhav et al., 2016).

4. CONCLUSION

The present study conducted to test the efficacy of fortified Mulberry feed on the growth and Cocoon quality of *Bombyx mori* L. Mulberry leaves fortified with three different milk samples (MST1, MST2 & MST3) were fed to silkworm larvae. The experimental study could highlight the fact that among the three test run samples, the bovine milk was found to be giving a significant result in the weight of the larva, cocoon quality, pupal weight and shell weight, when compared to the control unit. Therefore, it is concluded that the bovine milk is positively and significantly yielding the result.

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 the writing or editing of this manuscript.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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