



## The variation in economic traits and biomolecules in silkworm as influenced by mulberry leaf enriched with *Aloe vera*

Bhavani M\*, Thippeswamy L, Shiva Kumar B, Sanjana M

Department of Studies in Sericulture Science, Laboratory of Silkworm Physiology and Biochemistry, University of Mysore, Manasagangotri, Mysuru, Karnataka, India

Corresponding author: Bhavani M

### Abstract

Mulberry is the only sole food of silkworm *Bombyx mori*, which influences on the physio-biochemical properties of the larva. In this present investigation, the effect of mulberry leaf enriched with *Aloe vera* at different concentrations viz., 0.1, 0.3 and 0.5% on biomolecules and economic parameters of silkworm hybrid FC1 × FC2 were recorded. The results of the study disclosed that the larvae supplemented with *Aloe vera* at 0.5% concentration registered maximum protein and carbohydrate content in fat body and mid gut over respective control batches. Similarly, economic parameters viz., larval weight, cocoon weight, shell weight, shell ratio, filament length, filament weight and raw silk percentage were recorded highest at the same concentration compared with rest of the treated groups as well as control.

**Keywords:** Silkworm, *Aloe vera*, Supplementation, Biomolecules, Economic parameters

### Introduction

The silkworm, *Bombyx mori* L., is a strictly monophagous insect that depends exclusively on mulberry leaves (*Morus* spp.) for its nourishment. As a result, the entire nutritional requirement for its growth, development, and silk synthesis must be derived from mulberry foliage. Any fluctuation in the nutritional composition of mulberry leaves can influence the physiological performance of the silkworm and ultimately affect cocoon and silk production. Although mulberry leaves generally provide most of the nutrients required for larval development, there are occasions when certain essential nutrients may be present in inadequate amounts, leading to sub-optimal larval growth and reduced cocoon yield. Consequently, successful cocoon production is largely dependent on the availability of nutritionally rich mulberry foliage (Ravikumar, 1988) [19]. Previous studies have also demonstrated that appropriate dietary management significantly influences both the quantitative and qualitative aspects of silk production in *B. mori* (Murugan *et al.*, 1998) [16]. The nutritional composition of mulberry leaves, however, is not constant, it varies according to several factors including the genotype of the plant, soil fertility, climatic conditions, rainfall or irrigation patterns, agronomic practices, and plant protection measures. When leaf quality is poor or nutritionally imbalanced, supplementation through fortified nutrients such as proteins, amino acids, vitamins, carbohydrates and minerals becomes essential (Sengupta *et al.*, 1972) [23]. Such supplementation can help bridge nutritional deficiencies in the leaves, enhance larval appetite, and improve disease resistance, ultimately resulting in higher survival rates and improved cocoon productivity (Akhtar and Asghar, 1972) [1].

Several plant-derived substances have been reported to possess insect growth regulatory properties, including effects on juvenile hormone activity and metabolic regulation. Supplementation of silkworm diets with botanicals extracts such as *Tribulus terrestris*, *Psoralea*

*corylifolia*, *Phyllanthus niruri*, *Polypodium species*, *Parthenium hysterophorus*, and *Tridax procumbens* has been reported to enhance larval growth and improve cocoon parameters (Rajashkargouda, 1991; Murugan *et al.*, 1998; Shiva Kumar *et al.*, 1995; Muruges, 2002) [16, 17, 24]. Similarly, application of *Lantana camara* and *Clerodendrum inerme* extracts at 5% concentration to fifth instar larvae significantly improved important economic traits (Mamadapur, 1994; Santosh Kumar, 1997) [13]. Furthermore, supplementation of mulberry leaves with ascorbic acid enhanced larval growth and resulted in heavier cocoons (Karaksy, 1990; Madhu Babu, 1992) [8, 12]. Although several studies have documented the positive effects of botanical extracts and nutritional supplements on the economic parameters of silkworm rearing, limited information is available regarding their influence on the biochemical changes occurring within the silkworm. Therefore, a comprehensive investigation into the biochemical responses of silkworm larvae to botanical supplementation is necessary to better understand their physiological mechanisms and potential benefits. In this context, the present study was undertaken.

### Materials and Methods

#### Rearing of silkworm and supplementation of *Aloe vera*:

The present experimental study was conducted in Silkworm Physiology and Biochemistry Laboratory, Department of Sericulture science, University of Mysore, Mysuru. The disease free layings of popular double hybrid FC1xFC2 are procured from National Silkworm Seed Organization, Mysuru and reared with popular V1 mulberry variety as per the methods of Krishnaswami (1986). The *Aloe vera* formulation was procured from kamicka organics and stock solution was prepared by dissolving appropriate quantity of *Aloe vera* and further diluted at the rate of 0.1, 0.3 and 0.5%. The prepared solutions were preserved at 4°C until further use. For the experimentation, larvae were divided into four batches each with 150 worms after third moult.

The batch I, II and III larvae were reared on mulberry leaves treated with *Aloe vera* of respective concentrations. The treated leaves were dried under shade at room temperature and fed to silkworm daily once as first feed during 4<sup>th</sup> and 5<sup>th</sup> instars and remaining two feeding with normal leaves. In contrast, control batch (IV) worms were fed with mulberry leaves soaked with distilled water. Ten larvae were used from each treatment in three replications to analyse biomolecules and economic parameters. The parameters such as shell ratio, filament length, denier, renditta and raw silk percentage, were calculated by using following formulae:

$$\text{Shell ratio (\%)} = \frac{\text{shell weight}}{\text{cocoon weight}} \times 100$$

Filament length (m):  $L = R \times 1.125$

R = Number of revolutions recorded by an epprouvette.

1.125 = Circumference of epprouvette in meter.

$$\text{Denier} = \frac{\text{filament weight}}{\text{filament length}} \times 9000$$

It indicates thickness of the silk filament.

$$\text{Renditta} = \frac{\text{weight of cocoons reeled}}{\text{weight of raw silk obtained}}$$

Unit quantity of cocoons required to produce one unit of raw silk.

$$\text{Raw silk percentage (\%)} = \frac{\text{reeled silk weight}}{\text{green cocoon weight}} \times 100$$

The obtained data was statistically analysed by standard deviation method.

#### Estimation of protein content in fat body and mid gut tissues

The protein content in fat body and midgut tissue were estimated by adopting the method of Lowry *et al.* (1951)<sup>[11]</sup>. 100mg of tissue was homogenized with 10ml of distilled water and centrifuged at 3000rpm for 10 minutes. From the reaction mixture, 1ml supernatant was taken, for this 5ml of protein reagent was added and incubated at room temperature for 10 minutes. Then 0.5ml of folin's reagent was added. The contents were mixed thoroughly and kept for 30 minutes until color development at dark condition. 1ml of bovine serum albumin (BSA) was used as a standard. A blank was prepared using 1 ml of distilled water. OD was measured at 660nm against blank.

$$\text{Amount of protein (mg/g)} = \frac{\text{OD of the sample} \times \text{mg of BSA in standar}}{\text{OD of the standard} \times \text{mg of tissue taken}} \times 1000$$

#### Estimation of carbohydrate in fat body and mid gut tissues

The carbohydrate content in fat body and midgut tissues was estimated by adopting the anthrone method as described by Dubios *et al.* (1956)<sup>[4]</sup>. 100mg of tissue was homogenized with 10ml of distilled water and centrifuged at 3000rpm for 10 minutes. To the 1 ml of supernatant, 4 ml of anthrone

reagent was added and the content was boiled for 8 minutes in a water bath, then cooled with running water. 1ml of glucose solution was used as a standard. OD was measured at 630 nm against blank and the carbohydrate content was expressed in mg/g of tissue. The obtained data were statistically analysed by adopting the standard deviation method and mean values were expressed.

$$\text{Amount of carbohydrate (mg/g)} = \frac{\text{OD of the sample} \times \text{mg of glucose in standard}}{\text{OD of the standard} \times \text{mg of tissue taken}} \times 1000$$

## Results and discussion

### Impact of fortified mulberry leaves with *Aloe vera* on protein content:

Proteins are fundamental biomolecules that plays an indispensable role in the biological functions of the silkworm, *Bombyx mori* L. They are directly involved in regulating growth, development, metamorphosis, and silk formation, which are critical processes determining the productivity of this economically valuable insect in sericulture. The fat body acts as the principal metabolic and storage organ where most protein synthesis and accumulation occur. These stored proteins provide the necessary resources for larval growth and are particularly important during the transition from the larval to the pupal stage. During the fifth instar, a large proportion of the synthesized proteins is transported to the silk glands, where they are utilized in the synthesis of silk proteins required for cocoon formation. The results of the present study indicate that silkworm larvae fed with *Aloe vera* exhibited the highest protein content in the fat body at a 0.5% concentration (120.05 mg protein/g tissue) compared with the midgut (85.64 mg protein/g tissue) and their respective control groups (109.09 and 74.31 mg protein/g tissue) (Fig. 1). The observed enhancement in protein levels in both the fat body and midgut tissues may be attributed to the improved utilization of nutrients present in *Aloe vera*, which possibly enhanced metabolic activity and protein synthesis in the larvae. The results are parallel with the earlier study of Damodhara *et al.* (2024)<sup>[2,3]</sup> who have opined that larvae supplemented with *Aloe vera* at 0.6% concentration enhance the protein content in fat body (129.74 mg/g) and mid gut (113.3 mg/g) when compared to their control batches (102.47 and 99.01 mg/g, respectively). Similar trend was also observed with ayurvedic formulations namely Hingwastaka and Kapikacchuat (Thanvi Rehaman *et al.*, 2025).

### Impact of fortified mulberry leaves with *Aloe vera* on carbohydrate content

Carbohydrates are essential nutritional constituents that significantly affect the growth, development, and physiological efficiency of the silkworm *Bombyx mori* L. As the primary source of metabolic energy, they play a vital role in supporting the normal functioning of various biological processes in the insect. The success of silkworm rearing is greatly influenced by the availability and effective utilization of carbohydrate resources, which are mainly derived from mulberry leaves. These energy-rich compounds supply the fuel required for cellular metabolism and contribute to the formation of important biomolecules necessary for growth and development. During the

advanced stages of larval life, especially in the fifth instar, a large portion of the energy obtained from carbohydrates is utilized by the silk glands to sustain the intense process of silk protein synthesis. Therefore, adequate carbohydrate metabolism is crucial for maintaining physiological activities and promoting proper development and silk production in the silkworm. In the current study larvae reared on mulberry leaves fortified with *Aloe vera* at different concentration showed significant impact on carbohydrate content in fat body and mid gut tissues. When larvae nourished with 0.5% *Aloe vera* exerted maximum carbohydrate content in fat body (36.21 mg carbohydrate/g tissue) over mid gut (32.84 mg carbohydrate/g tissue) against respective control batches (30.10 and 27.05 mg carbohydrate/g tissue). It is due to effective utilization of nutrients present in *Aloe vera*. The results were also supported by Dyana Selin *et al.* (2018) [5] who have observed that silkworm larvae fed on mulberry leaves enriched with spirulina exerted maximum carbohydrate content at 3% concentration in fat body (16.34 mg/g) against control group (12.07 mg/g). According to Kumar and Balasubramaniyan (2014) [10] the worms fed with spirulina at 1% concentration scored maximum total carbohydrate content of 16.35 mg/ml compared to the control (15.90 mg/ml).

#### Influence of fortification of *Aloe vera* on larval and cocoon Parameters

The silkworms reared on mulberry leaves fortified with *Aloe vera* at different concentration showed markable variations in larval and cocoon parameters. When larvae treated with *Aloe vera* at 0.5% concentration recorded higher larval weight, cocoon weight, shell weight and shell percentage of 4.60g, 2.07g, 0.46g and 22.28%, respectively. In contrast, the control group recorded comparatively lower values of 4.31g, 1.85g, 0.39g and 20.69%. Among the tested concentrations, the 0.1% *Aloe vera* treatment showed the lowest values for larval weight, cocoon weight, shell weight and shell percentage (4.39g, 1.92g, 0.41g and 21.32%, respectively) compared with the other treatments. (Table. 1). These corroborate the earlier findings of Roja *et al.* (2025) [20] when larvae treated with mulberry leaves extra foliated with *Aloe vera* at different concentrations registered increase in larval weight, cocoon weight, shell weight, shell ratio. Similar results were also observed when larvae enriched with mulberry leaf treated with spirulina at 500 ppm during 4<sup>th</sup> and 5<sup>th</sup> instar (Sameena Maqbool *et al.*, 2023) [21]. The same trend was also observed with some other plant extracts such as *Curcuma longa*, *Ocimum sanctum* and *Withania somnifera* (Sudeepthi *et al.*, 2025; Hajam *et al.*, 2024) [7, 25].

#### Influence of fortification of *Aloe vera* on reeling parameters

Silkworms fed on mulberry leaves extra foliated with *Aloe vera* at different concentrations exhibited enhanced reeling parameters *viz.*, filament length, filament weight, denier, renditta and raw silk percentage. Longest filament length (1137m) and highest filament weight (0.38g) was recorded when larvae nourished with 0.5% *Aloe vera* as against control batch (998m and 0.32g respectively). Among the treatments it was shortest (1055m) and lowest (0.34g) in 0.1% *Aloe vera* treated group when compared to remaining concentrations (0.3 and 0.5 %). The worms treated with 0.5% *Aloe vera* scored lowest denier and renditta of 2.96 and 4.82 kg. In contrast, control batch showed highest denier and renditta (2.75 and 5.3kg). However, maximum raw silk percentage of 19.7% was noticed at 0.5% as against to control batch (17.87%). Among the treated groups it was minimum in 0.1% *Aloe vera* treated group (Table 1). The results are inconformity with the findings of Damodhara *et al.* (2024) [2, 3] who have noticed that larvae supplemented with 0.6% *Aloe vera* exhibited longer filament length, higher filament weight and raw silk percentage. More over the parameters like denier and renditta was lesser in *Aloe vera* treated group compared to un-supplemented batches. The similar results were also recorded with fortification of *E. prostrate*, *C. asiatica*, *W. somnifera* and *C. sativus* (Gayathri, 2005; Maribashetty *et al.*, 2010 and Murari *et al.*, 2008) [6, 14, 15].

#### Conclusion

The silkworm larvae reared on mulberry leaf fortified with *Aloe vera* at different concentration showed notables variations growth, development and biochemicals constituent of the larvae. The worms nourished with *Aloe vera* at 0.5% concentration enhance larval, cocoon and reeling parameters with the elevation of protein and carbohydrate content in the fat body and midgut of silkworm. However, protein and carbohydrate contents were relatively higher in the fat body when compared to midgut tissue in all the treatments. Hence, *Aloe vera* could be used as fortifying agent for silkworm rearing after conducting more field trails.

#### Conflict of Interest

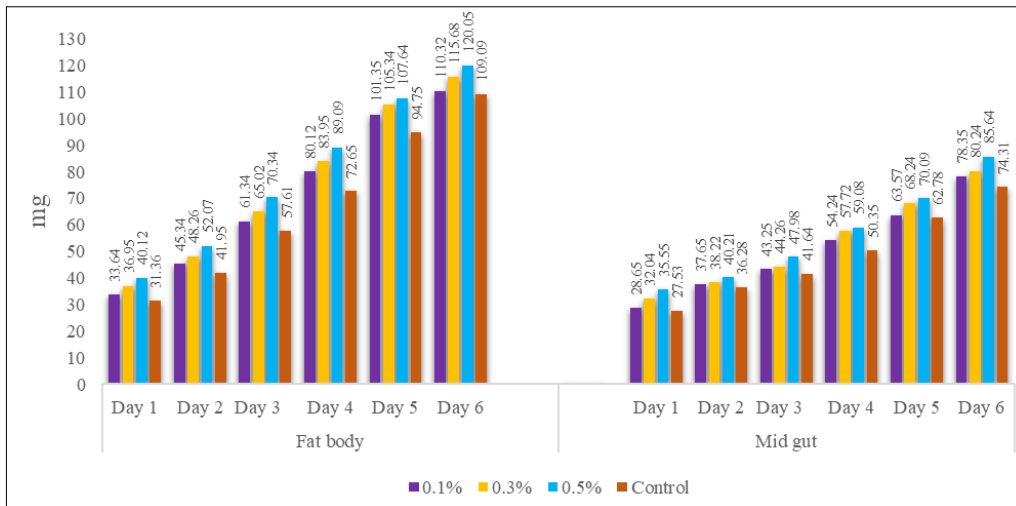
Authors have declared that no competing interests exist.

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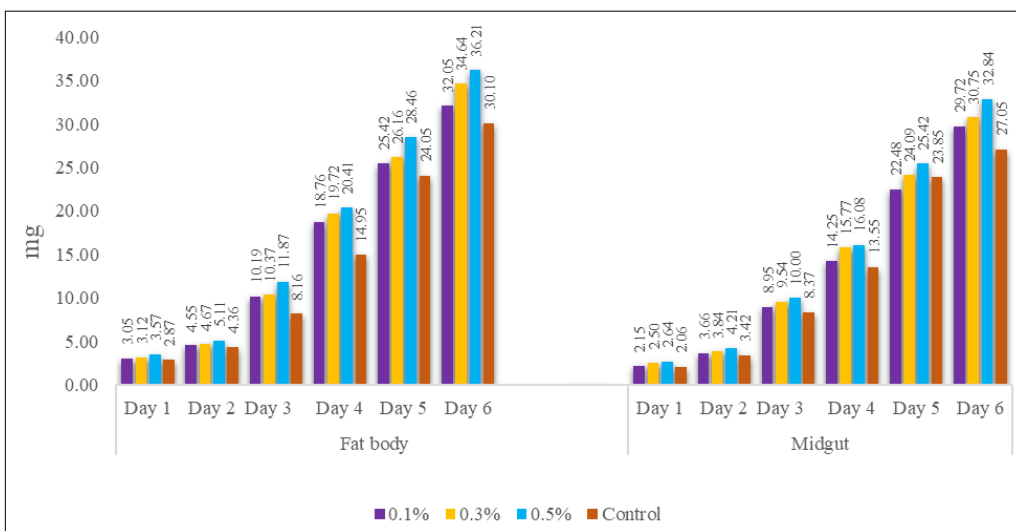
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**Table 1:** Effect of mulberry leaves fortified with *Aloe vera* at varied concentrations on economic traits of F1 x FC2 silkworm hybrid

Concentration	Larval weight (g)	Cocoon weight (g)	Shell weight (g)	Shell ratio (%)	Filament length (g)	Filament weight (g)	Denier	Renditta (kg)	Raw silk percentage (%)
0.1%	4.39±0.06	1.92±0.04	0.41±0.01	21.32±0.10	1055±4.58	0.34±0.05	2.92±0.08	5.03±0.09	18.53±0.41
0.3%	4.43±0.05	1.96±0.20	0.42±0.02	21.76±0.35	1108±7.00	0.37±0.05	2.94±0.07	4.93±0.10	19.37±0.16
0.5%	4.60±0.08	2.07±0.12	0.46±0.01	22.28±0.32	1137±6.81	0.38±0.07	2.96±0.10	4.82±0.06	19.7±0.31
Control	4.31±0.04	1.85±0.07	0.39±0.06	20.69±0.41	998±3.61	0.32±0.04	2.75±.10	5.30±0.08	17.87±0.11



**Fig 1:** Fortification of mulberry leaf with *Aloe vera* on protein content in fat body and mid gut of fifth instar FC1 x FC2 silkworm hybrid



**Fig 2:** Fortification of mulberry leaf with *Aloe vera* on carbohydrate content in fat body and mid gut of fifth instar FC1 x FC2 silkworm hybrid.

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