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# INFLUENCE OF NEUTRAL RED ON THE ECONOMIC, REPRODUCTIVE AND BIOCHEMICAL PARAMETERS OF SILKWORM BOMBYX MORI. L

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## Abstract

Nutritional requirement in food consumption have direct impact on the overall growth of silkworm and also increase the cocoon weight, cocoon length, shell weight, pupal weight, pupal length, silk length, silk weight, denier, moth efflorescence, fecundity, fertilized egg, unfertilized egg, hatching percentage and brushing percentage, carbohydrate, amino acid and protein in haemolymph and silk Gland of *B.mori*. The use of natural dyes for textile fibres has become an alternative to solve issues as environmental pollution and health risks by the larvae fed with neutral red treated mulberry leaves. In the present study the silkworm *B. mori* were fed with the concentration of neutral red (0.006 and 0.003ppm) treated mulberry leaves from the $5^{th}$ instar larvae. The findings elucidates that more vital dyes may use for the production of colour silk without causing any harm to the silkworm physiology. Dye plays a major role in the cocoon and silk production, it is assumed that it may not affect the economical reproductive parameters and biochemical parameters. This study was to find out the economical parameters of cocoon, pupa and silk, the reproductive parameters such as moth efflorescence, fecundity, fertilized egg, unfertilized egg, hatching percentage, brushing percentage and biochemical parameters such as the carbohydrate, amino acid and protein in haemolymph and silk Gland of *B.mori*. The observed results were statistically analysed and discussed.

**Key words:** Neutral red, *Bombyx mori*, Nutrition, Fecundity

## Introduction

Sericulture, a significant agro-based industry, offers work at several levels, like raising silkworms, growing host plants, reeling, spinning, and weaving yarn. Its quick development and labour-intensive nature greatly aids in fostering growth and reducing poverty in rural areas. The final result of this enterprise is silk fibre. Due to its modest initial investment and relatively high returns, this activity is highly suited for rural farmers, business owners, and craftspeople (Sreenivasa and Hiriyanna, 2014). During the last larval period the insect feeds voraciously, grows rapidly and assumes great economic significance to the sericulture industry because of the synthesis of silk protein in this stage. Silkworms are reared for their cocoons, from which pure silk is extracted through reeling. The silk so extracted

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forms a composite thread fit for weaving. In essence, silkworm rearing and reeling are inter-dependent on each other for economic stability and prosperity (Tokano and Arai, 1978).

Quality silkworm seed is vital for viable sericulture industry. Quality silkworm seed refers to richness of laying, egg viability, hatching uniformly and more importantly good rearing performance of the progeny (Ullal and Narashimhanna, 1981). Several factors influence the fecundity and fertility of silkworm races including aberrations in sex organ, faculty handling of moth during mating and egg laying (Biram *et al*., 2009).

Biochemical studies on the silkworm have been initiated with the aim of understanding the biochemical correlates of silkworm metamorphosis and silk production. The biochemical parameter which attracted considerable attention in this regard is the silk protein fibroin. The metamorphosis of silkworm involves rapid growth of the silk gland and increased protein and fibroin synthesis. Silkworm proteins and their developmental changes have been examined (Sarangi 1985). Pursuits in this direction have led to the studies on amino acids and on the enzymes involved in protein and amino acid metabolism (Parenti *et al.*, 1985).

Silkworm larva obtains different amino acids from the mulberry leaves and uses to synthesize silk proteins secreted during spinning. Silkworm has almost become an important tool for several biochemical studies. Protein synthesis in insects for better productivity of major biomolecules such as carbohydrates, lipids and proteins, play an important role in biochemical process underlying growth and development of insects (Wyatt, 1967).

## MATERIALS AND METHODS

### Experimental Organism -*Bombyx mori*

The silkworm, *B. mori* (Lepidoptera) CSR2 strain was used in this study. Eggs of CSR2 obtained from the Grainage centre, Sericulture Department in the Regional Deputy Director's office, Tenkasi, Tamil Nadu.

### Silkworm feed

*Morus alba* is the common mulberry plant and the sole preference of the silkworm larvae. This perennial plant is well suited for the Indian climate. All the essential elements such as protein, carbohydrate, inorganic salts, essential vitamins and minerals, required for the physiological functions of the silkworm are present in it. The MR2 mulberry leaves to feed the control animal were collected from the mulberry garden. The control animals fed regularly with mulberry leaves throughout the period. The experimental group fed with the prepared modified diet.

### Preparation of dye added diet

The dye selected in this study is rhodamine, a vital dye. After some trials, the concentration (0.003and 0.006ppm) which has no harmful effect on silkworm life cycle was selected. The dye solution sprayed uniformly on the mulberry leaves and fed the silkworms from the fifth instar. Three groups of fifth instar larvae were made, which include one untreated control group and two neutral red treated group. Each group was reared 50 larvae.

### Economic parameters of *B. mori*

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Following parameters were analysed such as cocoon weight, cocoon length, shell weight, pupal weight, pupal length, silk length, silk weight and denier.

**Cocoon Weight (gm)**

Ten randomly selected cocoons were taken from all groups and weighted using an electronic balance. The weight of each cocoon from each group was recorded separately and expressed in gram per cocoon.

**Cocoon Length (cm)**

Ten randomly selected cocoons were taken from all groups. Length of each cocoon from each group was recorded separately and expressed in cm per cocoon.

**Shell Weight (gm)**

Randomly selected 10 cocoons were cut open with the help of a blade, and the shell weight was taken accurately. The mean weight of the shell was calculated and expressed in gram per shell.

**Pupa Weight (gm)**

Ten randomly selected pupa were taken from all groups and weighted using an electronic balance. The weight of each pupa from each group was recorded separately and expressed in gram per pupa.

**Pupa Length (cm)**

Ten randomly selected pupa were taken from all groups. The length of each pupa from each group was recorded separately and expressed in cm per pupa.

**Silk Length (m)**

Ten randomly selected silks were taken from all groups. The length of each silk from each group was recorded separately and expressed in cm per silk.

**Silk Weight (gm)**

Ten randomly selected silks were taken from all groups and weighted using an electronic balance. The weight of each silk from each group was recorded separately and expressed in gram per cocoon.

**Denier (gm)**

Denier is the unit used to denote the thickness of silk filament. It is the weight of a 9,000-metre length of silk expressed in grams. The value of denier varies from 1.7 to 2.8. Filament denier was measured using an epprouvette and a denier scale. The denier was calculated by the following formula: It is calculated by using the formula

$$\text{Denier (D) (gm)} = \frac{\text{Weight of the filament}}{\text{Length of the filament}} \times 9000$$

**Reproductive parameters of *B. mori***

**Moth Efflorescence (%)**

It is the number of pierced cocoons divided by the total number of cocoons and was recorded as an average of three laying's in each replicate. It was calculated by the following formula:

$$\text{Moth Efflorescence (\%)} = \frac{\text{Number of pierced cocoon}}{\text{Total number of cocoons}} \text{ x } 100$$

**Fecundity (numbers)**

It was obtained by counting all of the eggs that the female moth laid, and it represents the total number of eggs laid by a single mother moth.

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**Rate of Fertilized Egg (%)**

It is the number of pigmented eggs from the total eggs laid by a mother moth and was recorded as an average of three laying's in each replicate. It was calculated by the following formula:

$$\text{Rate of fertilized egg (\%)} = \frac{\text{Number of pigmented eggs}}{\text{Total number of eggs}} \times 100$$

**Rate of unfertilized egg (%)**

It is the number of dead eggs from the total eggs laid by a mother moth and was recorded as an average of three laying's in each replicate and expressed in percentage. It was calculated by the following formula:

$$\text{Rate of unfertilized egg (\%)} = \frac{\text{Number of dead eggs}}{\text{Total number of eggs}} \times 100$$

**Hatching percentage**

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

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

**Brushing percentage**

It is the number of larvae brushed out from the total eggs laid by a mother moth, which was recorded as an average of three laying's in each replicate and expressed in percentage. It was calculated by the following formula:

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

**Biochemical parameters of *B.mori***

In the biochemical parameters,of *B.mori* the carbohydrate, amino acid, and protein contents of the haemolymph and silk gland were analysed.

**Sample preparation**

**Haemolymph Preparation**

Ten random samples from fifth-instar larvae in their third day were selected from the control and treatment groups and kept in the refrigerator for 4–5 minutes to facilitate the free running of haemolymph. Prolegs were cut, and the haemolymph was collected in cleaned, sterilised, and pre-cooled Eppendorf tubes. A pinch of n-phenyl thiourea was added to prevent the oxidation of haemolymph. The samples were centrifuged at 10,000 rpm for 10 minutes. The supernatant was removed and kept at -20 °C for analysis.

**Silk Gland**

Body tissue and silk glands were dissected out. The unwanted fat bodies, blood vessels, and nervous system were removed from the body tissue with the help of forceps and homogenised in 2 ml of Tris buffer (pH 6.8) or 10% TCA for glucose, 20% TCA for protein, and a 2:1 chloroform-ethanol (cholesterol) mixture.

**ESTIMATION OF CARBOHYDRATE, PROTEIN AND AMINO ACID**

The collected and prepared samples were used for the estimation of various biochemical constituents like carbohydrates, amino acids, and proteins**.**

**Carbohydrate Estimation**

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The anthrone method of Schiefter *et al.* (1950) was used to determine the silk gland and haemolymph carbohydrate. The carbohydrate of the silk gland was expressed as mg/g and haemolymph as mg/ml.

**Total Amino Acid Estimation**

The method of Chinard (1952) was used for the estimation of total amino acids in the silk gland and haemolymph. The total amino acids of the silk gland were expressed as mg/g and haemolymph as mg/ml.

**Protein Estimation**

The method of Lowry *et al.* (1951) was used for the estimation of total proteins. The total protein content of the silk gland was expressed as mg/g and haemolymph as mg/ml.

**Statistical analysis**

All the data were presented as mean ± standard deviation and Two – way analysis of variance (ANOVA) was used to test the significance of differences between mean values of independent observations of all the selected parameters of silkworm *B.mori*. The significance was calculated at 5% level (values are significant when $P < 0.05$).

**RESULTS**

The experimental group silkworm fed with neutral red sprayed mulberry leaves were observed for feeding and morphological changes. Table 1 shows the effects of different concentrations of dyes on cocoon weight, cocoon length, shell weight, pupal weight, pupal length, silk length, silk weight, and denier of the $V^{th}$ instar of the control and the experimental groups, were studied.

**Table 1:** Economical Parameters of control and different concentrations of neutral red fed group of *B.mori*.

| Concentration (ppm) | Cocoon weight (mg) | Cocoon length (cm) | Shell weight (mg) | Pupa weight (mg) | Pupa length (cm) | Silk length (cm) | Silk weight (gm) | Denier (gm) |
|---|---|---|---|---|---|---|---|---|
| Control | 840±5.71 | 3.38±0.77 | 171±2.65 | 669±5.15 | 3.4±0.43 | 596.4±8.3 | 0.122±0.002 | 1.84±0.002 |
| 0.006 | 872±4.19 (3.81) | 3.47±0.26 (2.66) | 174±3.7 (1.75) | 698±4.63 (4.33) | 3.5±0.001 (2.94) | 623.4±7.2 (4.53) | 0.135±0.003 (10.65) | 1.95±0.001 (5.98 ) |
| 0.003 | 880±4.39 (4.76) | 3.48±0.92 (2.96) | 178±2.3 (4.09) | 702±4.81 (4.93) | 3.6±0.001 (5.88) | 652.6±6.9 (9.42) | 0.138±0.001 (13.11) | 1.90±0.003 (3.26 ) |

- Each value represents the mean ± SD of five replicates.
- Significant level indicated by ($p<0.05$)

**Table 2**: Reproductive Parameters of control and different concentrations of neutral red fed group of *B.mori*.

| Concentration (ppm) | Moth efflorescence (%) | Fecundity (number) | Rate of fertilized egg (%) | Rate of unfertilized egg (%) | Hatching percentage | Brushing Percentage |
|---|---|---|---|---|---|---|
| Control | 71.43±6.48 | 244±20.81 | 92.62±8.69 | 7.38±0.04 | 88.11±7.63 | 83.20±5.91 |
| 0.006 | 75±5.24 (5.00) | 257±19.06 (5.33) | 93.38±0.23 ( 0.82 ) | 6.61±0.03 ( -10.43 ) | 90.27±8.47 ( 2.45 ) | 83.27±8.06 ( 0.08 ) |
| 0.003 | 75±4.32 (5.00) | 262±17.63 (7.38) | 94.66±8.61 ( 2.20 ) | 5.34±0.01 ( -27.64 ) | 91.22±8.4 ( 3.53 ) | 87.02±7.38 ( 4.59 ) |

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- Each value represents the mean ± SD of five replicates.
- Significant level indicated by ($p<0.05$)

**Table 3**: Biochemical Parameters of control and different concentrations of neutral red fed group of *B.mori*.

| | Hemolymph | | | Silk gland | | |
|---|---|---|---|---|---|---|
| Concentration (ppm) | Carbohydrate (mg/ml) | Amino acid (mg/ml) | Protein (mg/ml) | Carbohydrate (mg/g) | Aminoacid (mg/g) | Protein (mg/g) |
| Control | 120.34±1.91 | 5.97±0.05 | 122.90±2.12 | 174.87±3.29 | 9.46±0.05 | 120.95±1.49 |
| 0.006 | 162.83±1.22 (35.31) | 6.50±0.02 (8.88) | 166.47±3.74 (35.45) | 175.19±3.69 (0.18) | 14.51±0.23 (53.38) | 137.42±1.06 (13.62) |
| 0.003 | 181.12±2.93 (50.51) | 17.14±0.98 (187.10) | 185.60±2.81 (51.02) | 176.74±3.62 (1.07) | 14.60±0.07 (54.33) | 142.76±2.23 (18.03) |

- Each value represents the mean ± SD of five replicates.
- Significant level indicated by ($p<0.05$)

**Economic parameters of *B. mori***

**a) Cocoon Weight (mg):**

In control group of *B.mori*, the cocoon weight 840 ± 5.71 mg was recorded. In experimental group, the cocoon weight (872±4.19 and 880±4.39 mg) were recorded on the exposure of 0.006 and 0.003 ppm of neutral red respectively. All these three groups 0.003 ppm treated groups were more effective than the other groups.

**b) Cocoon length (cm):**

In control group of *B.mori*, the cocoon length 3.38±0.77 cm was recorded. In experimental group, the cocoon length (3.47±0.26 and 3.48±0.92 cm) were recorded on the exposure of 0.006 and 0.003 ppm of neutral red respectively. All these three groups 0.003 ppm treated groups were more effective- than the other groups.

**c) Shell Weight (mg):**

In control group of *B.mori*, the shell weight 171±2.65 mg was recorded. In experimental group, the shell weight (174±3.7 and 178±2.3 mg) were recorded on the exposure of 0.006 and 0.003 ppm of neutral red respectively. All these three groups 0.003 ppm treated groups were more effective than the other groups.

**d) Pupal Weight (mg):**

In control group of *B.mori*, the pupal weight 669±5.15 mg was recorded. In experimental group, the pupal weight (698±4.63 and 702±4.81 mg) were observed on the exposure of 0.006 and 0.003 ppm of neutral red respectively. All these three groups 0.003 ppm treated groups were more effective than the other groups.

**e) Pupal length (mg):**

In control group, the pupal length 3.4 ± 0.43 mg was recorded. In experimental group, the pupal length (3.5 ± 0.001 and 3.6 ± 0.001 mg) were obtained on the exposure of 0.006 and 0.003 ppm of neutral red respectively. All these three groups 0.003 ppm treated groups were more effective than the other groups.

**f) Silk length (cm):**

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. In control group, the silk length 596.4±8.3 cm was recorded. In experimental group, the silk length (623.4±7.2 and 652.6±6.9cm) were recorded on the exposure of 0.006 and 0.003 ppm of neutral red respectively. All these three groups 0.003 ppm treated groups were more effective than the other groups.

**g) Silk Weight (gm):**

In control group, the silk weight (0.122±0.002) gm was recorded. In experimental group, the silk weight (0.135±0.003 and 0.138±0.001 gm) were recorded on the exposure of 0.006 and 0.003 ppm of neutral red respectively. All these three groups 0.003 ppm treated groups were more effective than the other groups.

**h) Denier (gm):**

In control group, the denier (1.84±0.002) gm was recorded. In experimental group, the silk weight (1.95±0.001 and 1.90±0.003 gm) were recorded on the exposure of 0.006 and 0.003 ppm of neutral red respectively. All these three groups 0.003 ppm treated groups were more effective than the other groups.

**Reproductive parameters of *B. mori***

**a) Moth efflorcsence (%):**

In control group, the moth efflorcsence 71.43±6.48 gm was recorded. In experimental group, the moth efflorcsence (75±5.24 and 75±4.32gm) were recorded on the exposure of 0.006 and 0.003 ppm of neutral red. All these three groups 0.003 ppm treated groups were more effective than the other groups.

**b) Fecundity (number):**

In control group, the fecundity 244 ± 20.81 (number) was recorded. In experimental group, the cocoon width (257 ± 19.06 and 262 ± 17.63 number) were recorded on the exposure of 0.006 and 0.003 ppm of neutral red respectively. All these three groups 0.003 ppm treated groups were more effective than the other groups.

**c) Rate of fertilized egg (%):**

In control group, the rate of fertilized egg (92.62±8.69 %) was recorded. In experimental group, the rate of fertilized egg (93.38±0.23 and 94.66±8.61 %) were recorded on the exposure of 0.006 and 0.003 ppm of neutral red. All these three groups 0.003 ppm treated groups were more effective than the other groups.

**d) Rate of unfertilized egg (%):**

In control group, the rate of unfertilized egg (7.38±0.04 %) was recorded. In experimental group, the rate of unfertilized egg (6.61±0.03 and 5.34±0.01 %) were recorded on the exposure of 0.006 and 0.003 ppm of neutral red. All these three groups 0.003 ppm treated groups were more effective than the other groups.

**e) Hatching percentage:**

In control group, the rate of hatching percentage (88.11±7.63 %) was recorded. In experimental group, the rate of hatching percentage (90.27±8.47 and 91.22±8.4 %) were recorded on the exposure of 0.006 and 0.003 ppm of neutral red. All these three groups 0.003 ppm treated groups were more effective than the other groups.

**f) Brushing Percentage:**

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In control group, the rate of hatching percentage (83.20±5.91 %) was recorded. In experimental group, the rate of hatching percentage (83.27±8.06 and 87.02±7.38 %) were recorded on the exposure of 0.006 and 0.003 ppm of neutral red. All these three groups 0.003 ppm treated groups were more effective than the other groups.

**Biochemical Parameters of Silkworm *Bombyx Mori* L.**

**Carbohydrate content on haemolymph and silk gland of Silkworm *Bombyx mori* L.**

In control group, the carbohydrate on haemolymph (120.34±1.91 (mg/ml) was recorded. In experimental group, the carbohydrate on haemolymph (162.83±1.22 and 181.12±2.9 mg/ml) were recorded on the exposure of 0.006 and 0.003 ppm of neutral red. All these three groups 0.003 ppm treated groups were more effective than the other groups. While in control group, the carbohydrate on silk gland (174.87±3.29 mg/g) was recorded. In experimental group, the carbohydrate on silk gland (175.19±3.69 and 176.74±3.62 mg/g) were recorded on the exposure of 0.006 and 0.003 ppm of neutral red. All these three groups 0.003 ppm treated groups were more effective than the other groups.

**Amino acid content on haemolymph and silk gland of Silkworm *Bombyx mori* L.**

In control group, the amino acid on haemolymph (5.97±0.05 mg/ml) was recorded. In experimental group, the amino acid on haemolymph (6.50±0.02and 17.14±0.98 mg/ml) were recorded on the exposure of 0.006 and 0.003 ppm of neutral red. All these three groups 0.003 ppm treated groups were more effective than the other groups. Whereas in control group, the amino acid on silk gland (9.46±0.05 mg/g) was recorded. In experimental group, the amino acid on silk gland (14.51±0.23 and 14.60±0.07 mg/g) were recorded on the exposure of 0.006 and 0.003 ppm of neutral red. All these three groups 0.003 ppm treated groups were more effective than the other groups

**Protein content on haemolymph and silk gland of Silkworm *Bombyx mori* L.**

In control group, the protein on haemolymph (122.90±2.12 mg/ml) was recorded. In experimental group, the amino acid on haemolymph (166.47±3.74 and 185.60±2.81 mg/ml) were recorded on the exposure of 0.006 and 0.003 ppm of neutral red. All these three groups 0.003 ppm treated groups were more effective than the other groups. While in control group, the protein on silk gland (120.95±1.49 mg/g) was recorded. In experimental group, the protein on silk gland (137.42±1.06 and 142.76±2.23 mg/g) were recorded on the exposure of 0.006 and 0.003 ppm of neutral red. All these three groups 0.003 ppm treated groups were more effective than the other groups.

**Discussion**

The economic parameters of the silkworm *B. mori* are significantly influenced by the nutritional elements. The number and quality of dye-treated leaves offered to the silkworms can thus readily affect their economic development. According to Chinnaswamy Ramesh (2011), silkworm breeding has a significant impact on economic features. According to Dahi *et al.* (2016), the evaluation of nutritionally effective silkworm breeds must take into account the economic aspects of silkworms. According to Suresh (2012), the genetic make-up of silkworm larvae influences whether or not they can produce eggs. The results of the investigation on the performance of neutral red treated leaves for the six reproductive metrics, including moth, and fecundity rate of fertilized egg, rate of unfertilized egg, hatching percentage and brushing percentage are intriguing.

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Supplemental feeding has a significant impact on life cycle features like development, reproduction, and lifespan in insects. The experimental group of larvae had a considerably higher brushing percentage (8.88%) than the control group. Twinkle *et al.* (2019) finding that the silkworms treated just once per day had an increase in brushing percentage (96.78%) supports these findings. In the study conducted by Singh *et al.* (2017), *B. mori* silkworm larvae brushed more frequently in all directions.

Biochemical aspects of the silkworm larvae treated with mulberry leaves were assessed. The four biochemical parameters were provided by the study's results on the effectiveness of neutral red and rhodamine-b, which are presented here. Interesting findings can be obtained by looking at the carbohydrate, amino acid and protein contents of haemolymph, and silk glands. Our research indicated that the experimental groups of *B. mori* had considerably higher carbohydrate concentrations in their haemolymph and silk glands.

Further research by Kavitha *et al.* (2014) revealed that consumption of a ZnCl2-enriched mulberry meal considerably raised the quantities of carbohydrates in haemolymph and silk gland. In the fifth instar larvae of *A. mylitta*. Lakshmi (2012) found that the amount of carbohydrates in the fat body has increased. Smitha and Vijaya (2010) hypothesized that selenium-treated silkworm larvae of *B. mori* considerably increased the carbohydrate content of haemolymph.

**Conclusion**

In the present study to be concluded that economic, reproductive and biochemical parameters of *B. mori.* L was comparatively more when the silkworm fed with 0.003 neutral red treated mulberry leaves than the control and 0.006 ppm neutral red treated groups.

**Reference**

Biram, SNM, Tribhuwan, S & Beera, S 2009, 'Occurrence of unfertilized eggs in the mulberry silkworm, *Bombyx mori* L. (Lepidoptera: bombycidae)', International Journal of Industrial Entomology, vol. 18, pp. 1-7.

Chinnaswamy Ramesha, Hothur Lakshmi, Savarapu Kumari, S, Chebba M, Anuradha & Chitta Kumar, S 2011, 'Nutrigenetic Screening strains of the mulberry silkworm, *Bombyx mori,* for nutritional efficiency', Journal of Insect Science, vol. 12, no. 3, pp. 1 – 18.

Dahi HF, Rohab, H, Taha & Walaa, G, Ibrahim 2016, 'Nutritional efficiency and its relation to *Bombyx mori L.* productivity under different constant temperatures', Journal of Plant Protection and Pathology, Mansoura University, vol. 7,no. 1, pp. 21 – 26.

Kavitha sampath, sivaprasad Srinivasamurthy, Bano Saidullah & Yellamma Kuna 2014, 'Total Carbohydrates and economic parameters of *Bombyx mori*. under the influence of Zinc Chloride enriched mulberry diet', International Journal of Pure and Applied Bioscience, vol. 2, no. 5, pp. 196-202.

Lakshmi velide 2012, 'Studies on biochemical components of the larval haemolymph, fatbody and silkgland of tropical Tasar silkworm, *Anthereae mylitta* Dury (Daba T.V) under cold stress condition', European Journal of Experimental Biology, vol. 2, no. 6, pp. 2238-2242.

ISSN PRINT 2319 1775 Online 2320 7876
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Parenti, P, Giordana, B, Saochi, VF, Hanozct, GM & Guerritore, A 1985, 'Metabolic activity related to the potassium pump in the midgut of Bombyx mori larvae,' Journal of Experimental BioIogy,' pp. 11669-78.

Robert, BG & Rutt, MF 1982, 'Protein synthesis during early, adult and 2nd pupal development in silk moth *A. Polyphemus*,' Insect Bio-chemistry, vol.12, pp. 687-698.

Sarangi 1985, 'Alkaline protease in the midgut of the silkworm *Bombyx mori* L: changes during metamorphosis', Animal Sciences, vol. 94, pp. 567–572.

Singh, J, Kumar, A, Mukherjee, S, Singh, GP, Ray, S, Rawat, KS & Sinha, AK 2017, 'Assessment of Brushing date and direction on Tasar silkworm Antheraea mylitta D production and protection', International Journal of Current Microbiology and Applied Sciences, Vo. 6, no. 10, pp. 4853 – 4859.

Smitha, S & Vijaya Bhaskara Rao, A 2010, 'Alterations in the haemolymph Biochemical parameters of silkworm *Bombyx mori*. L. treated with selenium', America - Eurasian Journal of Toxicological Sciences, vol. 2, no. 1, pp. 51-53.

Sreenivasa & Hiriyanna, 2014, 'Study On the Factors Influencing Adoption Of New Technologies In Non Traditional Sericultural Area Of Chitradurga District, Karnataka. Global journal of biology', Agriculture and health science. Vol. 3(1), pp. 239-243.

Suresh Kumar, N & Harjeet singh 2012,' Evaluation of the reproductive potential of bivoltine silkworm hybrids of *Bombyx mori*. L under high temperature and high humidity and high temperature and low humidity conditions of the tropics', Universal Journal of Environmental Research and Technology, vol. 2, no. 5, pp. 443-449.

Takano K & Arai N 1978, 'Studies on the food value on the basis of feeding and cocoon productivity in the silkworm, *Bombyx mori* L,'The Journal of Sericultural Science of Japan. Vol. 57, pp. 134–142.

Twinkle Raghuvanshi, Bali, RK & Rubia Bukhari 2019, 'Effect of different feeding frequencies on the commercial characters of silkworm (*Bombyx mori* L.),' International Journal of Current Microbiology and Applied Science, vol. 8, no. 6, pp. 3193 – 3203.

Ullal, SR & Narashimhanna, MN 1981, 'Handbook of practical sericulture', central silk Board, Bangalore, India, pp. 61-82.

Wyatt, GR 1967, 'The biochemistry of sugars and polysaccharides in insects, Advanced Insect Physiology, vol. 4, pp. 287-360.