

## Effect of alternative plants on physiological and biological characteristics of silkworm *Bombyx mori* L.

El-Shewy A.M. and Elgizawy K. KH

Plant Protection Dept., Fac. of Agric. Moshtohor, Benha Uni., Egypt.

Received: 11 Sept. 2017 / Accepted: 04 Dec. 2017 / Publication date: 20 Dec. 2017

### ABSTRACT

Nutrition has an important role in improving growth and development of the silkworm *Bombyx mori* L. Although *B. mori* L. larvae are normally reared on mulberry leaves (*Morus alba*), those can be successfully reared on leaves of alternate plants. The aim of this study was to evaluate the effect of alternate plants, namely grapes leaves, *Bougainvillea glabra*, *Lettuce sativa*, *Lantana camara* and *Ficus retusa* with different concentrations of mulberry leaves powder aiming to increase the silk and egg production of *Bombyx mori* L., and also to improve low nutritional value of autumn rearing season. The results showed that treatment of mulberry leaves powder with grape leaves at 1 and 2% concentrations resulted high consumption of food. That was followed by lettuce leaves and Bougainvillea leaves. While, *lantana camara* and *Ficus retusa* leaves caused the least consumption of food. It was also found that, grapes and lettuce leaves caused significant increases in all the tested biological and physiological parameters during autumn season. Also, the new alternate food produced healthy cocoons and increased of eggs production. Meanwhile, the alternate plants *lantana comara* and *Ficus retusa* were the least efficient treatments.

**Key words:** *Bombyx mori* L., alternate plants, mulberry leaves, nutrition, biological and physiological parameters

### Introduction

The mulberry silkworm *Bombyx mori* L. (Lepidoptera:Bombycidae) is one of the most important commercial insects, producing finest natural silk, that called (Queen of textiles). *Bombyx mori* is monophagous and survives solely on mulberry leaves which play an important role in nutrition of the silkworm and in turn the resultant cocoons and silk production (Nagaraju, 2002). There had been always been a search for alternate host plant which can raise silkworm larvae to produce better silk in quality and quantity (Gopal, 1910) silk worm (*Bombyx mori* L.) the silkworm larvae can be reared successfully on leaves of peepal (*Ficus religiosa*) and produce healthy cocoons as compared to those raised on mulberry and other alternate host plants (Nasreen *et al.*, 1999). The present investigation aimed to study the effect of feeding on leaves alternate plants on silk worm *B. mori* productivity and a trial to fortify the autumn mulberry leaves by using alternate plants to improve silk and egg production in autumn rearing season. Another target was to study the effects of tested plants on growth of silkworm larvae, some biological parameters and to estimate the relationship between proteins and silkworm development.

### Materials and Methods

#### *Silkworm Resources and rearing:-*

The fourth instar larvae of mulberry silkworm *Bombyx mori* L. (hybrid Giza) were chosen for the present study. The eggs were obtained from the Sericulture Research Department, Plant Protection Research Institute (A.R.C) at Giza, Egypt. The larvae were reared on trays and provided with suitable amounts of fresh leaves of mulberry *Morus alba* (Balady variety) up to the last instar laboratory hydrothermic conditions of  $18 \pm 2$  C° and  $70 \pm 5\%$  R. H in the autumn season.

**Corresponding Author:** Elgizawy K. KH, Plant Protection Dept., Fac. of Agric. Moshtohor, Benha Uni., Egypt. E-mail: karam.elgizawy@fagr.bu.edu.eg  
1268

*Tested plants and treatments:-*

Five plant species were chosen as supplementary nutrients for the silkworm *B. mori*. These species were as follows:

*Bougainvillea glabra*, grapes leaves, *lettuce sativa*, *Ficus retusa* and *lantana camara*. Three different concentrations (1, 2 and 3%) of mulberry leaves powder were prepared. The tested plant leaves were soaked in each concentration of mulberry leaves powder for 30 seconds, and then dried in air. The tested plants were fed to the freshly molted fourth instar larvae. Control insects were reared on fresh mulberry leaves. Three replicates per each concentration (treatment) and control containing 100 larvae/treatment.

*Biological parameters:*

These studies were conducted for evaluating the effect of tested plants on the larval mortality, durations of the fourth larval instar and pupal stage. Also fresh weights of mature larvae, dry silk glands and cocoons were weights. After the formation of cocoons, ten randomly chosen cocoons were harvested from each replicate. Each cocoon was cut carefully and weight pupae individually and cocoon shell on an electronic balance (Nirwani and Kaliwal, 1996). Five couples of moths from each treatment and control were recorded. To determine the fecundity of emerged female.

*Physiological parameters:*

The food consumption parameters such as the efficiency of conversion of both digested food (E. C. D.) and growth rate (G. R.) were estimated according to Waldbauer (1968). The total haemolymph protein (T. H. P) was estimated according to Lowery *et al.*, (1951) by drawing the haemolymph of six instar random larvae from each treatment and control. The six<sup>th</sup> instar mature larvae from each treatment and control were dissected and the silk glands were drawn and dried in an oven at 50°C. A suitable weight was placed in flask of protein digestion to measure the total proteins of silk glands by the method of Vogel (1961).

*Statistical analysis*

Statistical analysis was carried out using ANOVA with two factors under significance level of 0.05 for the whole results using SPSS (ver.19) and Data were treated as complete randomization design according to Steel *et al.* (1997). Multiple comparisons were carried out applying LSD.

## **Results and Discussion**

### **1-Effect of alternative plants on the physiological characteristics of *Bombyx mori* L.**

The effects of feeding *B. mori* larvae on tested plants on their physiological parameters are shown in Table (1). The obtained data showed substances was high by feeding on grape leaves 1% (E. C.D 46. 36%) with the growth rate (G.R.) 0. 17, among the other tested plants, but they were still higher than that of the control (E.C.D 28. 25%) and growth rate (G.R.) 0. 11. The lowest parameters were recorded for treatments of *Ficus* 2% which recorded (E.C.D 22.16%) and growth rate (G. R.)0. 10.

These results agree with Mahesha *et al.* (1999) who reported that among the various plant products and tested concentrations, *Tridax Procumbens* L. (30%) and *Parthenium hysterophorous* (20%) exhibited Phagostimulant property and increased food consumption parameters. Also Nasreen *et al.*, (1999) indicated that larvae of *B. mori* could be reared successfully on leaves of peepal (*Ficus religiosa*) and consumed and utilized the new food. Results of the present experiments support those of Muthunagai (2016) showing that worms of *B. mori* fed with *Ricinus communis* consumed almost 2 to 3 kgs of plant leaves and the feeding rate increased tremendously to enormous volume of 12 to 40 kgs during the fourth instars.

The haemolymph proteins levels of 4<sup>th</sup> instar larvae were relatively higher in grape leaves 1% and lettuce 1% and showed a significant increases which recorded 603.27 mg/ml and 586.90 mg/ml, respectively, and silk gland total proteins were 6.40 and 2.62%, respectively, in comparison with the control which produced (347.77 mg/ml) in total haemolymph proteins and (3.03%) with silk glands proteins. While Ficus 3% gave the lowest records of total haemolymph proteins (184.51 mg/ml) and (2.62%) of silk glands total proteins. A similar trend of data was demonstrated by Manoharan (1997) who found that the supplementation of hydrolyzed soybean caused increase in total haemolymph proteins of the *B. mori* fifth instar larvae. Moreover, the importance of vitamins in silkworm nutrition such as nicotinic acid, thiamin, riboflavin and niacin, was reported by Govindan *et al.* (1998). The protein content of the silk gland, fat body and muscles was found to increase significantly when larvae were fed with 1 and 2% ascorbic acid (Quraiza *et al.*, 2008).

**Table 1:** Effect of alternative plants on physiological parameters of silkworm, *B. mori* L. during autumn season

Plants	Conc. (%)	E.C.D.	G.R.	Total haemolymph proteins (mg/ml)	Silk glands proteins (%)
A	1	46.36±0.66	0.15±0.005	603.27±3.23	6.40±1.25
	2	45.13±0.95	0.16±0.002	561.32±2.32	5.47±0.25
	3	41.63±2.64	0.17±0.003	521.21±2.35	5.46±1.45
B	1	39.40±1.49	0.14±0.01	442.62±1.25	4.34±2.52
	2	32.58±4.98	0.13±0.006	357.84±4.25	4.66±1.52
	3	24.30±4.23	0.13±0.003	430.68±2.25	4.43±2.45
C	1	37.10±1.72	0.14±0.004	586.90±1.52	5.91±1.25
	2	35.46±1.05	0.15±0.005	470.55±1.23	4.48±0.85
	3	38.20±2.64	0.14±0.004	453.53±2.25	4.47±1.25
D	1	36.03±3.01	0.13±0.002	422.62±4.21	3.61±2.25
	2	31.86±3.003	0.13±0.007	423.80±3.45	3.91±1.25
	3	27.43±3.01	0.12±0.002	393.59±2.25	2.96±1.52
E	1	23.06±3.003	0.11±0.007	232.00±3.45	2.66±2.45
	2	22.16±1.47	0.10±0.004	206.77±1.25	3.21±1.45
	3	24.23±1.16	0.10±0.003	184.51±2.52	2.62±0.25
Control		28.26±	0.11±	347.77±1.35	3.03±2.25

A= Grape leaves    B= *Bougainvillea glabra*    C= *lettuce sativa*    D= *lantana camara*    E= *Ficus retusa*

### Effect of feeding on tested plants on the biological parameters of silkworm *Bombyx mori* L:-

#### Larval characteristics

The statistical analysis of data (Table 2) showed that the percentage of larval mortality did not exceed (6.20%) among all treatments, in comparison with the control which recorded (7.07%). In this present study, the results indicated that all tested plants caused significant reduction in larval duration especially with grapes leaves 2% (4.99 days) and lettuce 3% (5.01 days), but still significantly lower than the control (9.98 days).

#### Cocoon characteristics

The demonstrated results in Table (2) indicated that the heaviest larval, pupal and silk gland weights were gained for the treatments of (1, 2% and 3% with grape leaves which recorded for larvae (3.37, 3.06 and 3.18g, respectively), pupae (1.68, 1.56 and 1.72g), and for silk glands (1.64, 1.61 and 0.87g, respectively). All these weights were significantly heavier than those of the control. Among the other treatments, the lowest weights (1.02, 1.02 and 0.40g), respectively, were recorded for treatment of Ficus 1%. As a general trend, the average weight of fresh cocoon and cocoon shells from all used concentrations were significantly higher than that of control, especially in case of grape leaves treatment. Grape leaves at 3% (1.60 and 0.26 g), while those of control recorded (1.09 and 0.14 g). On contrary, the treatments of Ficus and *lantana camara* were less effective and gave lower averages of weight, although still significantly heavier than control. The treatment of grape leaves at 1%, significantly, increased the number of deposited eggs/female up to 375.53 eggs/female followed by

treatment of lettuce 3% and lantana 3% (366.30 and 353.83 eggs/female, respectively), in comparison with the control which produced (310.00 eggs/female. All tested plants induced significant increases in hatchability especially grape leaves recording (88.43 %) followed by lettuce plant (87.77%) and *lantana camara* (86.66%), while Ficus leaves caused lowest mean values (74.29) compared to the Control (75.20). All the tested plants induced significant increase over the control except Ficus plant. These results agree with Subburathginam *et al.* (1990) who showed that the enrichment of mulberry leaves with calcium chloride increase the cocoon characters as cocoon weight, shell weight cocoon, shell ratio and silk proteins (Fibroin and sericin) . Ganga and Gowri (1990) noticed that the dietary supplementation of wheat and rice flours resulted an increase in fibroin content of the silkworm and increased some growth and cocoon parameters. On the other hand Etebari and Matindoost (2004) recorded prolongation in the larval stage to 31 days and larvae turned into pupae as affected by hyper vitaminosis with vitamin B3. Also Quader *et al.*, (1992) found that nutritional value of mulberry leaves was directly reflected on cocoon characters of *B. mori*, that may explain the difference in cocoon characteristic among treatments. Moreover, Bentea *et al.*, (2011) reported that the use of substances (mineral and vitamin additives) improve the quality parameters of cocoon. Also Lokesh and Anantha Narayana (2011) found that when larvae treated with different concentrations of vitamins C and B that increased the food consumption and conversion leading to increased growth of silkworms. Also, Rajesh Kumar and Gangwar (2010) found that the food plant of different species influenced the larval growth, larval duration and cocoon and pupal weight, shell weight and fecundity. Also Muthumagai (2016) found that when larvae of *B. mori* were fed on alternative plant *Ricinus communis* increased food consumption in all the larval stages, the weight of larva, cocoon and shell. Also, Masthan *et al.*, (2017) found that when larvae of *B. mori* were fed on different food probiotic organisms, that gave better cocoon quantitative parameters like cocoon weight, shell weight, pupal weight, shell percentage and silk filament length of silkworm *Bombyx mori* .

**Table 2:** Effect of alternative plants on biological parameters of silkworm, *B. mori* L. during autumn season

Plants	Conc. %	Larval mortality (%)	Larval duration (day)	Wt. of larvae (g)	Wt. of pupae (g)	Wt. of silk glands (g)	Wt. of fresh cocoon (g)	Wt. of cocoon shells (g)	No. deposited eggs/female	Hatchability (%)
A	1%	6.01±0.88	5.02±0.13	3.37±0.54	1.68±0.02	1.04±0.02	1.46±0.01	0.26±0.01	373.53±2.80	88.43±1.46
	2%	6.01±0.01	4.99±0.09	3.06±0.02	1.56±0.02	1.01±0.01	1.35±0.01	0.21±0.01	341.33±1.15	82.57±1.60
	3%	5.01±0.96	5.70±0.60	3.18±0.06	1.72±0.08	0.79±0.10	1.60±0.08	0.26±0.02	329.83±5.90	78.39±2.45
B	1%	5.00±0.82	7.98±1.22	2.06±0.54	1.35±0.16	0.87±0.19	1.22±0.03	0.18±0.03	244.26±6.05	84.36±2.90
	2%	5.75±0.55	6.72±0.52	2.34±0.01	1.27±0.08	0.75±0.07	1.31±0.04	0.24±0.02	322.80±9.37	86.66±2.04
	3%	5.72±0.43	6.02±0.43	2.54±0.10	1.29±0.02	0.72±0.15	1.14±0.07	0.23±0.01	328.36±1.05	85.05±1.41
C	1%	5.71±0.61	5.96±0.49	2.87±0.05	1.42±0.03	0.78±0.05	1.44±0.07	0.24±0.02	320.53±17.99	76.96±1.86
	2%	5.43±0.20	6.00±1.88	2.74±0.52	1.34±0.10	0.80±0.17	1.36±0.02	0.18±0.02	333.00±14.58	81.98±2.51
	3%	6.20±0.47	5.01±0.97	2.84±0.18	1.31±0.03	0.70±0.04	1.21±.19	0.24±0.01	366.30±2.73	87.77±2.96
D	1%	5.96±0.62	9.02±0.29	1.78±2.09	1.12±0.03	0.50±0.12	1.22±0.15	0.18±0.02	346.00±14.19	79.38±
	2%	5.41±0.49	10.02±0.56	1.78±0.47	1.13±0.02	0.65±0.12	1.24±0.05	0.20±0.01	344.40±21.98	82.15±1.12
	3%	5.43±0.62	8.98±0.29	2.02±0.09	1.09±0.03	0.52±0.12	1.20±0.15	0.24±0.02	353.83±14.19	86.66±3.89
E	1%	5.33±0.49	8.00±0.56	1.02±0.47	1.02±0.11	0.40±0.12	1.11±0.08	0.23±0.01	322.86±21.98	75.77±1.12
	2%	4.97±0.25	10.01±0.86	1.25±0.03	1.07±0.02	0.58±0.12	1.33±0.08	0.15±0.04	327.30±3.42	74.29±6.15
	3%	6.04±0.48	9.03±0.27	1.03±0.11	1.11±0.02	0.57±0.03	1.04±0.18	0.23±0.13	333.43±0.03	82.07±2.41
Control		7.07±0.25	9.98±1.25	2.49±2.52	1.90±0.25	0.28±0.01	1.09±0.52	0.14±0.02	310.00±5.23	75.20±3.25

A= Grape leaves

B= *Bougainvillea glabra*

C= *lettuce sativa*

D= *lantana camara*

E= *Ficus retusa*

## References

- Bentea, M., L.A. Marghitas, and A.Sara, 2011. The effect of some additives on bioproductive performances of silk worm *Bombyx mori* L., *Animal Science and Biotechnologies*, 44(1): 9-12.
- Etebari, K. R. Ebad and L. Matindoost, 2004. Effect of feeding mulberry's enriched leaves with ascorbic acid on some biological, biochemical and economical characteristics of silk worm *Bombyx mori* L. , *Int. J. Indust. Entomol.*, 8 : 81-87.
- Ganga, G. and G. Gowri, 1990. Flour diet to the silkworm to optimize cocoon characters. *Indian silk*, 13: 13-14.
- Gopal, N. M., 1910. *Hand Book of sericulture Bengal Secretariat Book Depot Calcutta, USA*, Pages: 298.

- Govindan, R., S.B. Magadam and T.K. Narayanaswami, 1998. Effect of supplementing vitamins B2, B6 and C to Eri-silkworm, *Samia Cynthia ricini* Boisid on economic traits. Agric. Sci., (1): 80 – 82.
- Lokesh, G. and S. R. Anantha Narayena, 2011. Changes in the protein profile of silkworm *Bombyx mori* L. (Lepidoptera: bombycidae) in response to the chemical mutagen .1. J.S.N., 2 (3): 559-563.
- Lowery, O.H., N.J.Rosebrough, A.I. Farr and R.J. Randall, 1951. Protein measurement with foline phenol reagent. J. Biol. Chem. 193: 265–276.
- Mahesha, H.M., R. Rajashekhargouda and S.G Rayar, 1999. Effect of aqueous extracts of few botanicals with special reference to weeds on *Bombyx mori* L., The proceeding of the XVIII<sup>th</sup> International Sericultural Commission Congress. Cairo- Egypt, 114-118.
- Manoharan, T., 1997. A study of hydrolyzed soybean protein. Supplementation on the reproductive system of mulberry silkworm, *Bombyx mori* L. PhD. Thesis Jamal Mohamed College, Bharathidasan University, Tiruchirappalli, India.
- Masthan, K.T., C.V. Rajkumar and M.Narasimha, 2017. Studies on fortification of mulberry leaves with probiotics for improvement of silk quality-International Journal of Biotechnology and Biochemistry, 13 (1): 73-80.
- Muthunagai, M., 2016. Comparative study on the feeding activity of *Bombyx mori* with host plant *Morus nigra* (Mulberry) and Alternative plant *Ricinus communis* (castor plant). Journal of Agriculture and veterinary science: 9 (12): 29-33.
- Nagaraju, J., 2002. Application of genetic principles in improving silk production–Current Science, 83 (4): 409 – 414.
- Nasreen Abida, G.M., M.Cheema and Q.Ashfa, 1999. Rearing of silkworm *Bombyx mori* L. on alternate food plants Pakistan Journal of Biological science, 2 (3):843-845.
- Nirwani, R.B and B. Bkaliwal, 1996. Effect of folic acid on economic traits and the change of some metabolic substances of Biovoltine silkworm, *Bombyx mori* L, Korean J. Ser. Sci. 38:118-123.
- Quader, M.A., M.A. Qaiyyum, A. A.Sarker, M.A. Rab and S.U. Ahmed, 1992. Varietal response to NPK Fertilizers in combination with foliar spray of urea on leaf yield and leaf quality of Mulberry. Bulletin of sericulture Research, 3: 64 – 66.
- Quraiza, M.T.F., C.R.Rathic, P.T.Muthu, S.M. Das, and M.R.Bai, 2008. A study on the effect of ascorbic acid in modifying tissue protein content and economic traits of *Bombyx mori* L. Journal of Basic and applied Biology; 2(4): 32–35.
- Steel, R.G.D., J.H. Torrie and D.Dickey, 1997. Principles and Procedure of Statistics. A Biometrical Approach 3<sup>rd</sup> Ed. McGraw Hill Book Co. Inc., New York. pp. 352-358.
- Subburathinam, K. M., V.Kabila and J. Sulochanachetty, 1990. Mineral spray can increase cocoon quality. Indian silk, 28 (12):35-36.
- Vogal, A.L., 1961. A text book of quantitative organic analysis, pp. 12-16.
- Waldbauer, G.R., 1968. The consumption and utilization of food by insects. Advances in insect physiology). (Academic press, London and N. Y. S: 229-283.