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Effect of Giloy & Ashwagandha on the reproductive parameters of *Bombyx mori* (L.)

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Abstract- Sericulture plays important role in enhancing the economy of India & also provides employment. The present study was an attempt to access the different concentration i.e. 1%, 3%, 5% of Giloy & Ashwagandha. In this study ovarian length was observed at 1% Giloy it is 131.65mm & at 5% Giloy it is 140.6mm & at 1% Ashwagandha it is 132.9mm & at 5% Ashwagandha it is 141.1mm. Weight of gonads at 1% Giloy testis is of 5.98mg & at 5% Giloy 7.29mg, ovary at 1% Giloy 6.25 mg & at 5% Giloy 7.68mg. Testis at 1% Ashwagandha is 5.68 mg & at 5% Ashwagandha it is 6.32mg. Ovary at 1% Ashwagandha is 6.35 mg & at 5% Ashwagandha 8.68mg. The fecundity, egg per female at 1% Giloy is 435 & at 5% Giloy it is 498. At 1% Ashwagandha fecundity is 449 & at 5% Ashwagandha 525. Egg hatchability at 1% Giloy is 76.5% & at 5% Giloy is 76.5% & at 5% Giloy it is 83.8%. At 1% Ashwagandha it is 77.4% & at 5% Ashwagandha it is 85%. In this study both the botanicals i.e. Giloy & Ashwagandha are identified for the impact on the reproductive characters & also strengthen the reproductive system which can be further utilized for improving the seed crop production of *Bombyx mori* (L.).

Keywords: Giloy, Ashwagandha, *Bombyx mori* (L.), Fecundity, Ovary.

INTRODUCTION

Silkworms are reared to produce silk by the process called “Sericulture”. India is an agriculture country & where after village industries like handloom & khadi. Sericulture plays very important role as it provides full or partial employment to about 6.5 million people in India. Sericulture also plays important role in enhancing the economy of India & also proves employment to the farmers & attracts profile seeking entrepreneurs as this farming requires low investment with relatively higher returns & also provides production of higher quality textiles. As this not only marked with this field, but also play important

role in medicine biotechnology, food supplements & also in biomaterials etc.

India ranks 2nd largest after its homeland china.¹ It was recorded that in 2016-2017, raw silk produced in India was 30348 MT out of which 21.273 MT is being contributed by *Bombyx mori* (L.), which makes it the 2nd largest producer in the world after china. From current data collected from the Indian sericulture market, Report ID: SR112023A511 reported that Indian sericulture market size reached 451.6 billion in 2022. IMARC group in 2028 expects the market to reach INRI, 194.5 billion, exhibiting a growth rate (CAGR) of 17.7% during 2023-2028. India is not only 2nd largest producer of raw silk but also largest consumer of raw silk & also silk fabrics & products. As from the data collected from the www.thehindu.com, chief

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executive officer P. Sivakur not only expressed confidence of India sericulture industry surpassing raw silk producing country in world, but also emphasized the need for Indian sericulture industry to start focusing more on the byproducts. The sole food of *Bombyx mori* (L.) is mulberry leaves which contain nutrients like phytosteroids, sugar, ascorbic acid & others were found as phytoestimulative action for silkworm. To enhance the quality and quantity of silk, the sleek thread, the investigation on silkworm nutrition may form active area for research. To promote silk quality & quantity supplementation & fortification of mulberry leaves is a recent technique in sericulture research. From studies it has been confirmed that the effect of many plant extracts with various metabolic activities results in acceleration of spinning.² The increase in larval cocoon as well as shell weight was reported.³ The improvement of reeling parameters was also reported by various researchers.⁴⁻⁸ The increase in pupal weight, increase in silk content, silk filament length & weight was also reported.^{3,7} All these studies were emphasized to improve the rearing parameters of silkworm. Many botanicals have been used to improve the reproduction in vertebrates⁹⁻¹¹ & in lower invertebrate^{12,13}.

In successful egg deposition by silk moth (female), several factors including hormonal, physical, chemical, environmental, behavioural aspects etc. have been attributed significant role among all photoperiod, temperature, humidity duration, frequency of mating etc. have been significant in oviposition behavior.^{14,15} It was reported that the reproduction physiology such as mating, vitellogenesis, oviposition, ovulation, weight of pupae & emerged adult, inclination & surface texture also imported factors for viable egg deposition by adult female moth.¹⁶ It was also reported that in *Culex tarsalis* oviposition showed bimodal pattern, in which egg laying occurred in the 1st few hrs following the onset of photo-phase & again after the beginning of scotophase.^{17,18} Moths of genus *Crambus* have been showed to oviposit mainly during the early scotophase.^{19,20} In various breed of silkworm (*Bombyx mori*) egg laying capacity vary even under identical atmospheric mating, nutritional & laying conditions.²¹⁻²⁹ Hatching of silkworm is predominantly diurnal & it seems to be under the circadian control.³⁰ Multivoltine, Nistari was selected because it is robust indigenous & local breed.³¹ Hatching in mulberry silkworm was predominantly diurnal under natural day condition. It seems to be under circadian control.³² Rhythmic patterns in hatching of

B. mori influenced by photoperiod.^{32,33} In recent years attempts have made in sericulture with nutrient such as protein, carbohydrate, amino acids, vitamins, sterols, hormones & antibiotics etc. to enhance the performance & get higher yield, quality & quantity cocoons. Giloy (*Tinospora cordifolia*) also called as Guduchi, Heart-leaves moonseed, Amrita etc. It is herbaceous vine belonging from the family Menispermaceae, which is indigenous to tropical region of Indian subcontinent. It has been used in ayurvedas for treatment of various disorders. It contains alkanoid, carbohydrate, glycosides, protein, amino acid, tannins, saponins, phytosterols, terpinoids, flavonoids, phenols, phytoestrogen, glycosides etc. Ashwagandha (*Withania somnifera*) commonly called as winter cherry, is an evergreen shrub belonging from the "Solanaceae" or nightshade family grows in India, Middle East & part of Africa. The plants particularly root powder, has been used for centuries in traditional Indian medicine. Although it is also used as herbal medicines & sold as dietary supplements there is insufficient scientific evidence that Ashwagandha is safe effective for treating any health condition or disease. Ashwagandha contains phenolics, alkanoids, saponin, tannin, glycosides, carbohydrates, flavonoids, amino acids, terpinoids, starch etc. Therefore, the study intended to observe the influence of Giloy & Ashwagandha fortification with mulberry leaves as a feed on reproductive performances of *Bombyx mori* (L.).

MATERIALS & METHODS

Collection & Rearing of *Bombyx mori* (L.):

The disease free layings of *Bombyx mori* (L.) & its larvae were collected from the central silk board, NSSO, Purnea (Bihar). These were reared under the standard conditions at 26°C ± 2°C, following the rearing method of Krishnaswami (1978)³⁴ with some modifications. The mulberry leaves were used as food for the *B. mori*. Larvae were reared in plastic trays (60 larvae per tray) & were exclusively fed with mulberry leaves. Fresh mulberry leaves were collected & stored in wet gunny bags. They were chopped prior to feeding. The leaves were fed four times per day at regular interval i.e. 6:30h, 11:30h, 16:00h & 22:00h.

Preparation of Botanical Extracts:

Giloy leaves were collected from the botanical garden of Patna Science college, Patna (Bihar). Ashwagandha roots collected from the botanical garden, Patna (Bihar).

They were washed thoroughly with distilled water to remove the surface contaminants & were dried at room temperature. The dried parts of both the botanicals i.e. Giloy leaves & Ashwagandha roots were mixed separately in the motor grinder & powder was prepared. This prepared powder was shocked in 150 ml of distilled water separately overnight; then filtered through a muslin cloth & these filterates were centrifused at 300 rpm for 15 minutes & from this stock solution three different concentrations were prepared i.e. 1%, 3%, & 5% of each filtrate.

Experimental Procedure:

The prepared extracts of Giloy & Ashwagandha, were taken at three different concentrations in this experiment i.e. 1%, 3% & 5% and were supplemented to *B. mori* (L.). Three different concentrations of botanical extracts were taken in which mulberry leaves were dipped differently in each different concentration of prepared extracts and were allowed for few minutes for water evaporation & fed in experimental larvae as the first feed. Larvae fed with

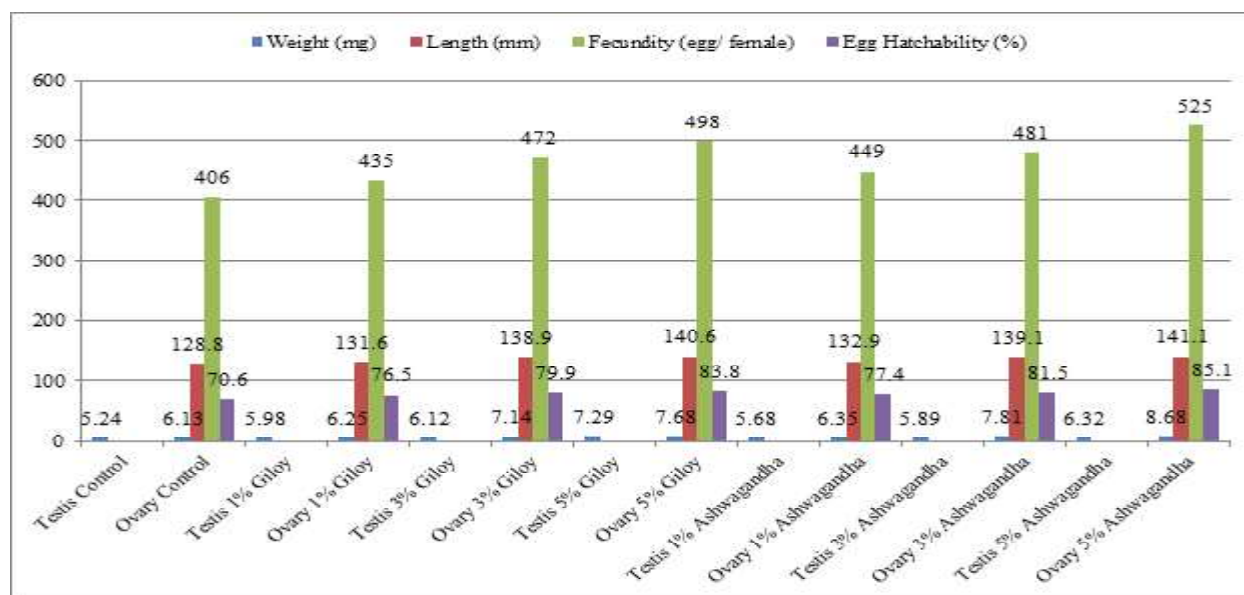
normal mulberry carried out according to the standard method suggested by Krishnaswami S. (1978)³⁴ with some modifications, while rearing silkworms were grouped into seven batches of 60 larvae for each treatment to examine the reproductive character & related parameters.

Reproductive Characters:

The female moth of *Bombyx mori* (L.) emerged from experimental and control group, were allowed to lay eggs after mating and their fecundity was counted separately. The hatching percentage was calculated in both Ashwagandha & Giloy supplemented groups & control groups. The freshly emerged female of control & supplemented groups was anesthetized to determine the length of the ovariole. The abdomens of each were dissected out & length of each ovariole was measured. The testes were isolated from male *Bombyx mori* (L.) & were weighted. The testes & ovaries were homogenized with 0.1M Tris HCL buffer of pH 7.4 for ovarian length, testis weight, fecundity & egg hatchability.

Table 1- Effect of Giloy & Ashwagandha supplemented with mulberry leaves on testis weight, length & ovaries weight, length, fecundity & egg hatchability of silkworm, *Bombyx mori* (L.).

| Parameter | Control | | 1% Giloy | | 3% Giloy | | 5% Giloy | | 1% Ashwagandha | | 3% Ashwagandha | | 5% Ashwagandha | |
|------------------------|-------------|--------------|-------------|--------------|-------------|--------------|-------------|-------------|----------------|--------------|----------------|--------------|----------------|-------------|
| | Testis | Ovary | Testis | Ovary | Testis | Ovary | Testis | Ovary | Testis | Ovary | Testis | Ovary | Testis | Ovary |
| Weight (mg) | 5.24 ± 0.49 | 6.13 ± 0.53 | 5.98 ± 0.46 | 6.25 ± 0.56 | 6.12 ± 0.49 | 7.14 ± 0.74 | 7.29 ± 0.52 | 7.68 ± 0.65 | 5.68 ± 0.47 | 6.35 ± 0.58 | 5.89 ± 0.47 | 7.81 ± 0.76 | 6.32 ± 0.54 | 8.68 ± 0.67 |
| Length (mm) | - | 128.8 ± 1.01 | - | 131.6 ± 1.12 | - | 138.9 ± 1.47 | - | 140.6 ± 1.7 | - | 132.9 ± 1.16 | - | 139.1 ± 1.46 | - | 141.1 ± 1.8 |
| Fecundity (egg/female) | - | 406 | - | 435 | - | 472 | - | 498 | - | 449 | - | 481 | - | 525 |
| Egg Hatchability (%) | - | 70.6 | - | 76.5 | - | 79.9 | - | 83.8 | - | 77.4 | - | 81.5 | - | 85.1 |



Graph 1- Effect of Giloy & Ashwagandha supplemented with mulberry leaves on testis weight, length & ovaries weight, length, fecundity & egg hatchability of silkworm, *Bombyx mori* (L.).

RESULTS & DISCUSSION

Ovariole Length:

The ovary length in the freshly emerged female adult of *B. mori* was found to increase in the supplemented groups compared with the control. The freshly emerged adult of female *B. mori* of control the ovariole length was 128.86mm. The female *B. mori* (L.), obtained for larvae fed with 1% Giloy & Ashwagandha, the ovariole length increased to 131.6mm at 1% Giloy & 132.9mm at 1% as given in table 01. The ovariole length reached the maximum of 141.1mm at 5% Ashwagandha which is in comparison with 5% Giloy is higher i.e. 140.61 mm. A positive co relation was noticed between the ovariole lengths with different concentrations of Giloy and Ashwagandha.

Weight of Gonads (Testis & Ovary of *B.mori*):

The ovarian weight of freshly emerged female was found to increase in Ashwagandha as compared to Giloy & the weight of testis of freshly emerged male was found to increase in Giloy as compared to Ashwagandha with respect to control. The freshly emerged adult male of control has 5.24mm of weight. In male, obtained from larvae fed with 1% Giloy the testis weight was 5.98mm & 1% Ashwagandha was 5.68mm. The maximum weight of testis emerged at 5% Giloy i.e. 7.29mm which is highest as compared to 5% Ashwagandha & control. The freshly emerged adult female of control has 6.13mm weight of ovary. In females, larvae obtained were fed with 1% Giloy the ovary weight was 6.25mm & at 1% Ashwagandha it was 6.35mm. The maximum weight of ovary inserted at 5% Ashwagandha i.e. 8.68mm which is highest as compared to 5% i.e. 7.68mm & control i.e. 6.13mm.

Fecundity (Egg/Female)

In the control group of *B. mori* (L.) female moth laid 406 egg/female. At 1% of Giloy supplemented female moth showed fecundity increased to 435 egg/female. The fecundity at 1% Ashwagandha supplemented female moth showed fecundity increased to 449 egg/female. The fecundity was maximum of 525 egg/female at 5% Ashwagandha as compared to 5% Giloy i.e. 498 egg/female & control i.e. 406 egg/female. A positive linear regression was observed between different concentration of Giloy & Ashwagandha and fecundity of eggs per female. The computation of data on one-way ANOVA revealed that fecundity length was significantly influenced by different concentration of Giloy & Ashwagandha.

Egg Hatchability:

In control group the egg hatchability was 70.6%. At 1% Giloy it was 76.5% hatchability at 3% Giloy it was 79.9% hatchability & at 5% Giloy supplement the hatchability was 83.8%. With Ashwagandha supplement the hatchability was also observed at 1% Ashwagandha the hatchability was 77.4% at 3% Ashwagandha the hatchability was 81.5% & at 5% Ashwagandha the hatchability was 85.1%. The maximum hatchability was observed at 5% Ashwagandha supplement which was also highest as compared with the 5% Giloy supplement i.e. 83.8%.

CONCLUSION

The present study was an attempt to access the influence of Giloy & Ashwagandha extract at different concentration i.e. 1%, 3% & 5%; Giloy contains phytochemicals having phytosterols, alkanoids, carbohydrates, saponin, glycoside, carbohydrates, amino acids etc. which possess the activity to influence the qualitative and quantitative characters of silkworm, *B.mori* (L.). Screening of Giloy, which are abundantly found in nature & are in reach of sericulture, which proves to be useful in augmenting reproductive performance & also strengthening the reproductive system of silkworm, *B.mori* (L.). In this study both the botanicals identified for the impact on the reproductive characters & also strengthen the reproductive system which can be further utilized for improving the seed crop production of silkworm.

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