## EFFECT OF FOLIC ACID ON THE HAEMOGRAM and SILK PRODUCTIVITY OF MULBERRY SILKWORM *Bombyx mori* L: (LEPIDOPTERA- BOMBYCIDAE)

# Gad, Abir A.; Magda H. A. Salem and Hanan M. Ramadan Depart. of Economic Entomology, Fac. of Agric., Alex. Univ.

## ABSTRACT

The present investigation reports the effect of folic acid as supplemented feed on haemogram and some biological parameters of *Bombyx mori* L. All the tested concentrations of folic acid especially 10mg/100ml significantly increased the weight of larvae ,pupae, fresh cocoon and cocoon shells. Also, treatment of folic acid increased total haemolymph protein, total haemocyte count, differential haemocyte count and corpora allata activity.

Keywords: vitamin, haemocytes , silk production , Bombyx mori L.

#### INTRODUCTION

Recently, many researches have been done on the diet supplementation of mulberry leaves fed to silk worms. These supplementations includes vitamins such as ascorbic acid, thiamin, miacin, folic acid and multi-vitamins (Nirwan and Kaliwat, 1996; Saha and Khan, 1996; Etebari, 2002 and Etebari and Matindoost, 2004).

Nutration is an important growth regulating factor in silk worm .lt has been reported that the vitamins of B- complex group and certain essential sugars, proteins ,amino acids, minerals are responsible for the proper growth and development of the silk worm ,*B .mori* .(Horie and Ito, 1963; Horie *et. al.*1966; Sengupta *et.al.*1972 ; Khan and Saha,1997 ;Faruki,1998).

Folic acid (Folinic acid, Folacin, Pteroy -glutamic) is essential for the synthesis of adenine and thiamine that make up genes, DNA and chromosomes. It stimulates production of red and white blood cells. Folic acid, also known as (vitamin B9), is a part of the B group vitamins and is water soluble (Anonymous, 1998).

The present work is aimed to determine the effect of folic acid on several biological and physiological parameters in the mulberry silk worm *B. mori.* 

## MATERIALS AND METHODS

All experiments were performed on the last larval instar of *Bombyx mori* L. under the normal laboratory conditions of  $25 \pm 2$  °C and 60 - 65% RH. For this study the source of folic acid was (Folic acid) supplied by the Nile Co.

for this study the source of folic acid was (Folic acid) supplied by the Nile Co. for pharmaceuticals and chemical Industries.

The concentrations of folic acid used in the present work were, 2.5, 5, 7.5, 10 and 12.5 mg/ml. prepared by dissolving folic acid tablets in tape water by (0.5, 1, 1.5, 2 and 2.5 tablet/ 100 ml water). Mulberry leaves were treated by dipping in these solutions then air dried and supplied to the 5<sup>th</sup> larval instar. Each treatment contain 3 replicate each have 50 larvae. The fresh weights of the mature larvae, silk gland, pupae, cocoons and cocoon

shells were determined. The number of deposited eggs/ female was considered.

Also, haemolymph protein was estimated using Biuret method described by Jones (1962). Total haemocyte counts (THC) were carried out using the Haemocytometer as reported according to Essawy (1985). Differential haemocyte counts (DHC) were calculated using the method described by Akai and Sato (1979),100 cells were identified to their typical haemocyte types after staining a smear of haemolymph with Wright's stain.

The corpora allata (CA) surface area used as an indicator of the glandular secretory activity of juvenile hormones Wirtz,(1973).Ten 5<sup>th</sup> instar larvae were immediately taken before spinning from each treatment and the control. The larvae were fixed in alcohol Bouin 's solution till measurements were taken . Head capsules were dissected and the CA were removed. The CA glands were drawn using Camera Lucida and CA surface areas were calculated using a planimeter. Data were statistically analyzed to check the significance of differences between treatments by using F test and L.S.D.

## **RESULTS AND DISCUSSION**

#### \* Effect of folic acid on the biology and productivity of *Bombyx mori* L.:

Data in Table (1) showed that folic acid at all different tested concentrations, showed significant increase in the mean weight of mature larvae, pupae and cocoon shells. The maximum improvement in larval weight was reached at concentration of (10 mg/100ml) whereas the rate of increase was about 44 % more than control. The same trend was observed in pupae ,cocoon and cocoon shells weight. Since the weights were 1.035 ,1.289 and 0.266 g, respectively at the treatment of this concentration. The minimum increase was obtained when larvae fed on mulberry leaves treated with (12.5mg/ml) as the weight of larvae ,pupae, cocoons and cocoon shells were 3.19, 0.904 ,1.16 and 0.228 g, respectively.

The statistical analysis showed that no significant difference were observed between different concentrations on the weight of larvae ,pupae, cocoon and cocoon shells while there are significant difference between different concentrations and control.

Treatment	Weight (g)				
	Larva	Pupa	Fresh	shell	Silk gland
		-	Cocoon		_
Control	2.22± 0.01 <sup>b</sup>	0.695±0.01 <sup>b</sup>	0.759±0.02 <sup>b</sup>	0.147±0.01 <sup>b</sup>	0.585±0.01 <sup>b</sup>
2.5mg/100ml	$3.20 \pm 0.02^{a}$	0.922±0.01 <sup>a</sup>	1.19±0.01 <sup>a</sup>	0.234±0.01 <sup>a</sup>	0.890±0.02 <sup>ab</sup>
5mg/100ml	$3.21 \pm 0.01^{a}$	0.946±0.03 <sup>a</sup>	1.19± 0.02 <sup>a</sup>	0.244± 0.01 <sup>a</sup>	0.920± 0.01 <sup>a</sup>
7.5mg/100ml	3.22± 0.02 <sup>a</sup>	0.983± 0.01 <sup>a</sup>	1.236± 0.01 <sup>a</sup>	0.252± 0.02 <sup>a</sup>	$0.980 \pm 0.03^{a}$
10mg/100ml	$3.25 \pm 0.03^{a}$	1.035± 0.03 <sup>a</sup>	1.289± 0.03 <sup>a</sup>	0.266± 0.02 <sup>a</sup>	1.03± 0.01 <sup>a</sup>
12.5mg/100ml	3.19± 0.03 <sup>a</sup>	0.904± 0.01 <sup>a</sup>	1.16± 0.01 <sup>a</sup>	0.228± 0.01 <sup>a</sup>	0.871±0.01 <sup>b</sup>

 
 Table (1):Effect of folic acid on some biological parameters of the mulberry silkworms Bombyx mori.

Each value presents the mean ±SE.

• Means at each column followed by the same letter are not significantly different at 0.01.

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Fig. (1) shows the effect of the folic acid on the mean number of deposited eggs per female moth which was treated during its larval stage with the tested concentrations .The vitamin at all concentrations, caused a significant increase in the mean number of deposited eggs/female .The highest average was 469 eggs/female at concentration 10 and 7.5mg/100ml while the lowest increment average was 414 eggs /female at concentration 12.5mg/100ml.

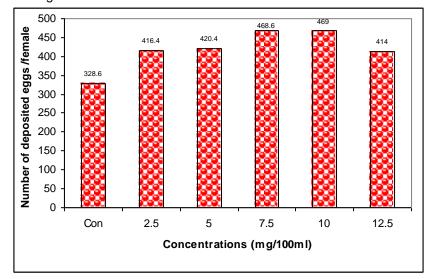


Fig. (1): Effect of folic acid on the mean number of deposited eggs/female of *Bombyx mori* L..

On the basis of the obtained results, the effect of folic acid on some physiological parameters such as haemolymph protein, silk gland and CA activity was studied to understand the mechanism by which the folic acid enhances the silk productivity.

\*Effect of folic acid on some physiological parameters of *Bombyx mori* L.

Table (1) indicated an increment in fresh silk gland weight treated with all the tested concentrations of folic acid The high weights were recorded at concentrations 10 followed by 7.5 and then 5 mg/100ml (1.03, 0.98 and 0.92 g), respectively. While, the light weights were observed at concentrations 12.5, 2.5 mg/100ml (0. 87 and 0.89 g), respectively, but still heaver than control (0.585).

The present data represented in Fig.(2) prove that folic acid as a food additives affected the CA surface area and the JH level. The maximum increase was observed at concentration 10 followed by 7.5 mg/100ml (0.991 mm<sup>2</sup>) and (0.882 mm<sup>2</sup>), respectively.

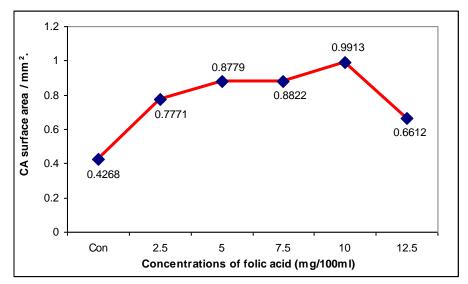


Fig.(2): Effect of folic acid on CA surface area of fifth instar larvae of Bombyx mori.

As shown in Fig. (3), Folic acid at concentration 10 mg/100ml increased the total haemolymph protein by 55.4% and at 7.5 mg/100ml by 47 % more than control. While the lowest effect was obtained at concentration 12.5 mg/100ml (30%).

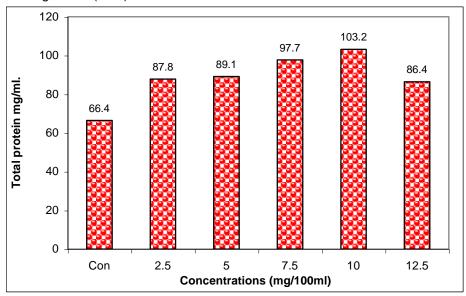


Fig.(3): Effect of folic acid on the total haemolymph protein of fifth instar larvae of *Bombyx mori*.

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The total haemocyte counts were higher than the control at all the tested concentrations .The maximum increase was observed when larvae fed on mulberry leaves treated with folic acid at 10 mg/100ml and 7.5 mg/100ml since the number of haemcytes /mm<sup>3</sup> were 28120 and 27100, respectively. The minimum increase was observed at concentration 12.5 mg/100ml (23500h/mm<sup>3</sup>) when compared with the control (Figure 4).

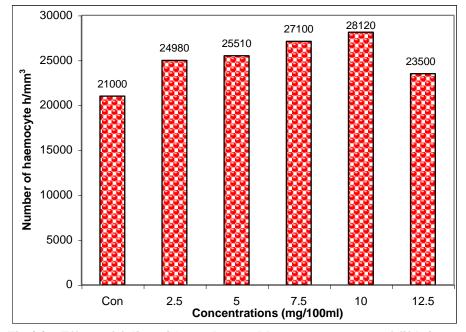


Fig.(4): Effect of folic acid on the total haemocyte count of fifth instar larvae of *Bombyx mori*.

Data in Fig. (5) showed that the treatments of folic acid increased the different haemocytes count of prohaemocytes, granulocytes, non-granulocytes, plasmatocytes and oenocytes. Treatment at concentration 10mg/100ml increase the number of prohaemocyte by about 87.5%, granulocyte 53.84%, non –granulocyte 141.6 %, plasmatocyte 72.7 % and Oenocyte 157% comparing with the control. While treatment at concentration 12.5mg/100ml showed slight increase in prohaemocyte (25%) ,Granulocyte (34.6%), non- granulocyte (100%), plasmatocyte (61.4%) and Oenocyte (114%) more than control.

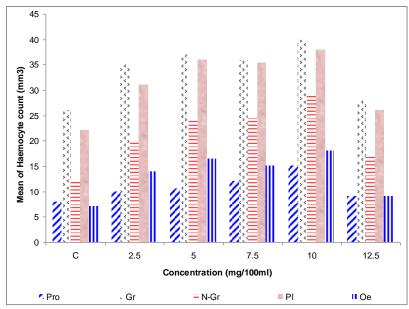


Fig. (5): Effect of folic acid on the different haemocyte count of of fifth instar larvae of *Bombyx mori*.

The present results are indicating the effect of folic acid that have been used at different concentrations on some biological and physiological parameters are in agreement with the finding of Khan & Saha, (1997) they found that the vitamins of B-complex group and certain essential sugars, proteins, amino acids are essential for the development of silkworm, *B. mori.* Govindan *et al.* (1989) noticed that the supplementing vitamins B, B complex and C to castor leaves at different concentrations increased the cocoon shell weight of the Eri –silk worm *Philosamia ricini*. SulaÇ and Emre (2000) recorded that use of folic acid in the diet of *Pimpla turionellae* showed positive effects on development and protein levels.

El-karaksy and Idriss (1990) stated that the amount and quality of silk production are strongly influenced by nutrition. Also, El karaksy *et al.* (1989) observed that increase of deposited eggs after the treatment with vitamins could be attributed to the direct effect of JH on the female reproductive system or its indirect effect on the blood protein.

Essawy and Idriss (1990) demonstrated that the JH affects the haemocyte poietic organ increasing the release of Oenocytes. The observed increase of all number of haemocyte types after treatment could be attributed to the indirect effects of folic acid on the haemapiotic organ.

In conclusion, the use of folic acid at concentration 10mg/100ml had a positive effect on the investigated biometrics of *B. mori* as well as the physiological characters and enhanced its silk and egg productivity.

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ت أثير حمض الفوليك على صورة المدم و إنتاجية الحرير فى دودة حرير القز**(Bombyx mori L. (LEPIDOPTERA- BOMBYCIDAE** عبير عبد المجيد جاد – ماجدة حسن على سالم – حنان محمد رمضان قسم الحشرات الاقتصادية - كلية الزراعة – جامعة الإسكندرية

تم في هذا البحث دراسة تأثير حمض الفوليك كمواد مكملة للغذاء على كلا من صورة الدم و إنتاجية الحرير لدودة القز. و قد وجد أن كل التركيزات المستخدمة (٧، ٥، ٥، ٧، و ١٢، ملجم/ ١٠٠مل لتر) أدت إلى زيادة في وزن كلا من اليرقات، العذاري، الشرنقة و كذلك قشرة الشرنقة و خاصة التركيز ١٠ ملجم / ١٠٠مل لتر.

و قد أدت المعاملة بحمض الفوليك أيضا إلى زيادة نسبة البروتين الكلى ، العد الكلى ، العد التمييزي للدم و كذلك زيادة نشاط غدة الجسم الكروي (CA).