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## Food Coloring Additives as a New Technology to Get Colorful Cocoons from Silkworm *bombyx mori*. L. and its Effect on the Economic parameters

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### ABSTRACT

The purpose of this study is to get cheap and new methods to color the silk of silkworm *bombyx mori* L. without affecting the economic parameters of the silkworm. Two colors were used to color the cocoons, Red and Blue. In each group, Meany parameters were studied. Both colors showed a positive significance compared to the control group. The percent from control was calculated and it had no negative effect on the parameters. Larval fresh weight at the end of the 5<sup>th</sup> instar was (2.857and 2.588gm) in red and blue, respectively. The maximum mean numbers were recorded in shell weight and shell ratio (0.284gm. and 20.87 %) in the blue color, respectively. Also, data cleared that using Food Coloring as a supplement in feeding detected a non-significant decrease in the parameters of silkworm compared with the control. However, the cost of the food colorants is cheaper than the industrial dyes. More studies need to assess how to get coloring silk at a low cost, is available, healthy, and enduring.

### INTRODUCTION

Silkworm, *bombyx mori* L. is one of the most beneficial insects. its plays an important role in the economic income in many countries. Many authors used nutrients as a food supplement to increase the silk yield (Sengupta *et al.*, 1992). To get a normal growth and good quality cocoons *Bombyx mori* L. requires sugars, amino acids, protein and vitamins. Palliva *et al.*, (2011), recommended that various factors are affected silkworm production such as environment (37%), mulberry leaves quality (38.2%), rearing technique (9.3%) and silkworm egg quality (3.1%) and others (8.2%). Silk is contents two natural proteins Fibroin and Sericin (Issa *et al.*, 2021). Fibroin is the main component that gives silk its characteristic. Sericin is a hydrophilic protein, that is responsible for absorption the of color (Kang *et al.*, 2011). Hybrids silkworm can produce greenish-yellow, yellow and golden yellow, but these colors were removed after the degumming stages. The colors of the cocoon depend on Carotenoids, Carotenes and Xanthophyll which transformed from mulberry leaves and appear in Sericin. Many stages take on silk to get colored silk in textile applications. The dyes processes during textile cause pollution and are more expensive (Baburaj and Das 2021) and (Nisal *et al.*, 2014). Trivedy Kanika *et al.*, (2016), reported that colored silk fiber is achieved by modifying genetically the silkworm. Other methods were taken to color silk fiber without modifying the silkworm was consisted of the natural physiology of silkworm (Kang *et al.*, 2011). Ritter, (2011) studied the effect of various

concentrations of Rhodamine to color the silkworm cocoons. the results showed that the cocoon was colored in the three concentrations and they suggested that, hydrophilic-hydrophobic interactions between the dye molecules and fibroin, make the colorful silks possible. Recently, researchers used a green technique to produce a colored silk-spinning from silkworm larvae at the end of the fifth instar after being fed with dyes as supplementary feeding depending on the permeability of the silk gland (Nisal, *et al.*, 2014). Synthetic dyes are used to color the silk (Trivedy, *et al.*, 2016). Nisal *et al.* (2014). They modified the feed of mulberry leaves by sprayed Azo dyes solution, the results revered to colored silkworm cocoons by these processes artificial degumming were reduced stages. The synthetic dyes were used as supplementary food to produce cocoon colures Dyes as Rhodamine B, Acridine Orange, Bismarck Brown, Acid Orange 142 and direct Red with different concentration (Issa *et al.*, 2021), the results showed that Rhodamine B get pink color and Acridine Orange, give orange color. The present work was initiated to produce a colored cocoon of silkworm by using different dyes safety, healthy, available and cheaper to use in food colors, especially sweets (Food Colorants) as a supplement food to the 5<sup>th</sup> instar larvae. More study needs to improve that these new dyes are the processes of making colored silk.

## MATERIALS AND METHODS

The present study was carried out in the laboratory of Plant Protection Institute, Shandawel, Agricultural Research Station, Agriculture Research Center, Sohag Governorate, during three successive rearing seasons of 2019, 2020 and 2021. The silkworm eggs (*Bombyx mori L.*) Imported by Sericulture Dep., Plant Protection Research Institute, Agri. Resch. Center, Ministry of Agriculture and Land, Reclamation, Giza, Egypt.

*Bombyx mori* were hatched and reared on the mulberry leaves (*Morus nigra*) under the laboratory condition as the standard methods of rearing, (Krishnaswami, 1978, Jolly, 1986 and Ghazy, 2014).

Larvae of the silkworm in the 5<sup>th</sup> instar were divided into three groups; each group was replicated three times. Group A: the larvae of silkworm fed on the mulberry leaves dipping on Food Red color. Group B: the larvae were fed with mulberry leaves after dipping with Food Blue color. Group C: the larvae silkworm fed without any additives (control). The total larvae in each group were 150 larvae. Larvae were fed 4/day.

### Treatments:

#### The Food Colorant Contents:

(Commercial bottle) Water – Glucose- Sugar – modified starch- Sodium Benzoate- Vanillin- Butyric Acid – Potassium Carbonate -Glycerin + water blue, or water red. Two bottles from each color were used without dilution (17gm / bottle) for 10 days of feeding 5<sup>th</sup> instar larvae.

### Measurements:

- 1- Larvae weight: two readings were detected in the 5<sup>th</sup> instar, and in the 3<sup>rd</sup> and 8<sup>th</sup> days. in each group.
- 2- Weight of silk gland: three larvae from each replicate were collected and the extract gland was then weighed and photographed.
- 3- Cocoon, pupal, and shell weight: Cocoons were collected randomized and weighted, then cutting the cocoons to get the pupal weight. shell weight was calculated.
- 4- Shell ratio %: Cocoon shell ratio % =  $\frac{\text{Wiegth of Cocoon shell}}{\text{weight of cocoon}} \times 100$

It was calculated according to Tanaka (1964).

- 5- Boiling and bleaching to remove colors: cocoons were put in the boiled water at (100°C) for 30 minutes. (Suna *et al.*, 2019).
- 6- Percent form control: were calculated and illustrated in figures.
- 7- Statistical analysis: Test using the statistical computer programmer Mstac. Duncan (1955). Correlation coefficients between some studies' parameters were calculated using the SPSS statistical program according to Wiley *et al.*, (1958) and Freund (1967).

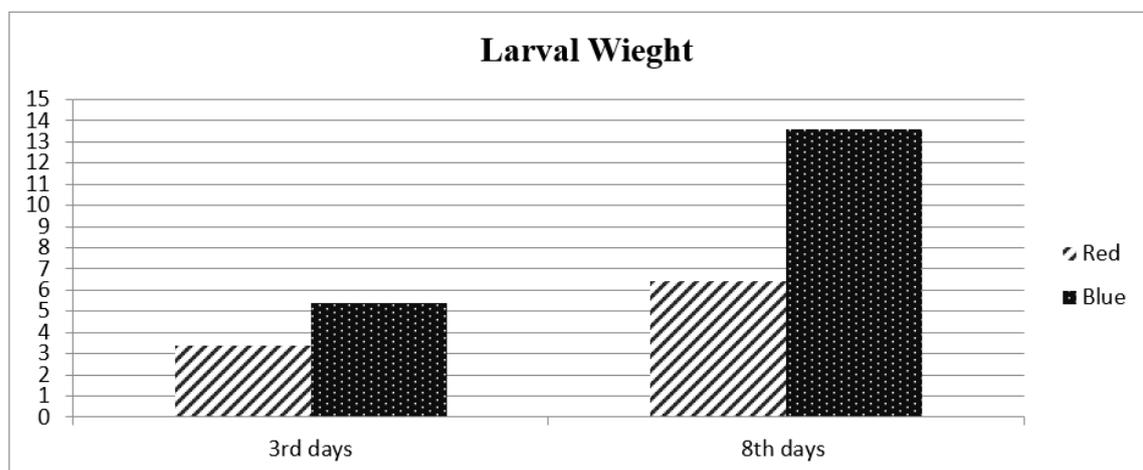
## RESULTS AND DISCUSSION

Data in Table (1) showed the affected of larval weight after feeding with food colorants on mulberry leaves during the three successive rearings. Revered that there was a significant difference between colors and control. The maximum mean weight on the 3<sup>rd</sup> day of the 5<sup>th</sup> instar was in blue color (2.137 gm), followed by control (2.103gm) then the red color (2.043 gm). While on the 8<sup>th</sup> day the end of the 5<sup>th</sup> instar, larval weight showed that the maximum mean was in the red color (2.857gm.) followed by the blue color (2.588gm) and then the control (2.559gm).

**Table 1:** The effect of food coloring on the larval weight of the 5<sup>th</sup> instar, during the three successive rearings.

Food colorants	Larval weight	
	3 <sup>rd</sup> day	8 <sup>th</sup> day
Red	2.043 C	2.857 A
Blue	2.137 A	2.588 B
Control	2.103 B	2.559 C

Means for each column followed by the same letters have not differed significantly at a 5% level of significance.



**Fig. 1:** The percent from control in the larval weight 3<sup>rd</sup> and 8<sup>th</sup> days fed with food colorants on mulberry leaves during the three successive rearings.

Figure (1), illustrated the percent of control which was calculated to present the effect of feeding silkworms with food colorants. Results summarized that both red and blue colors affected on the mean weight of larvae at the end of the 5<sup>th</sup> instar. The maximum increase was recorded in the blue color (13.60%). While Red color was (6.4%) at the end of the fifth instar. This result could be a concussed that using food colorants' color achieved a positive increase in the larval weight compared with the control. It could be revealed to the chemical structure of the food coloring its content of starch, sugar and glucose which

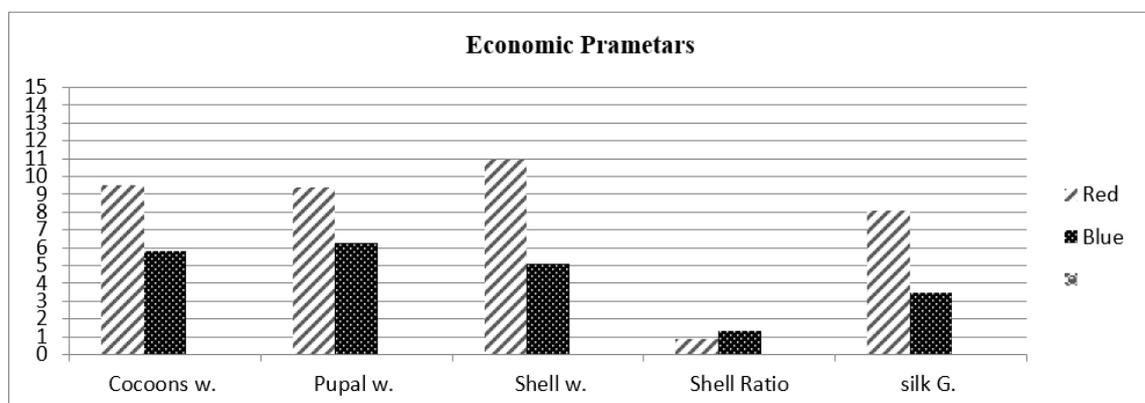
could be affected by the larval weight. This agreement with (Rashwan, 2010), reported that sugar treatment increases the larval weight in the local and imported hybrid *bombyx mori* L. Also using starch as supplementary feeding on mulberry leaves was revealed to increase the weight of the fifth instar larvae of the silkworm. (Shaki, 2015).

Data in Table (2) showed the effect of food colorants as a coloring substance on the economic parameters of silkworms during the three successive rearings. Data cleared that there were non-significant differences between color treatments and control. The maximum mean cocoon weight was (1.388 gm) followed by (1.364 and 1.337gm) in control, blue and red colors, respectively. The results in pupal weight were recent with control it was (1.115gm) in control followed by (1.081, 1.080gm) in red and blue color, respectively. The maximum mean of cocoon shell weight was recorded in Blue (0.284gm) while the red color was recorded (0.256gm). Also, results showed that there were non-significant differences between color treatments and control in the cocoon shell ratio. Blue color recorded a maximum mean number of the weight of silk gland (0.589gm) and shell ratio (20.87 %). Our results agree with (Issa *et al.*, 2021) conclusion that, adding dyes to mulberry leaves produced a cocoon colored without effect on the quality and weight of cocoons. Also, the percentage from control was calculated and illustrated in Figure (2). Data clear that feeding silkworms on mulberry leaves dropping in the color treatments showed a similar weight of control in all the economic parameters except the blue color treatment showed a simple decrease in the percent from control. The maximum increases were recorded in the red color (9.5, 9.4, 10.9, 0.9 and 8.1%) in the cocoon, pupa, shell weight, shell ratio and weight of the silk gland, respectively.

**Table 2:** Effect of food colorants on cocoon indices parameters of *bombyx mori* L during the three successive rearing.

Food coloring	Economic Parameters				
	Cocoons Weight (gm)	Pupal Weight (gm)	Silk gland (gm)	Shell Weight (gm)	Shell Ratio %
<b>Red</b>	1.337 A	1.081 A	0.573A	0.2560 A	19.08 A
<b>Blue</b>	1.364 A	1.080 A	0.58A	0.2843 A	20.87 A
<b>Control</b>	1.388 A	1.115 A	0.585 A	0.2707 A	19.83 A

Means for each column followed by the same letters have not differed significantly at a 5% level of significance



**Fig. 2:** The percent from control in the Economic parameters, of silkworm fed with food colorants on mulberry leaves during the three successive rearings.

### Food Coloring Additives as a New Technology to Get Colorful Cocoons from Silkworm

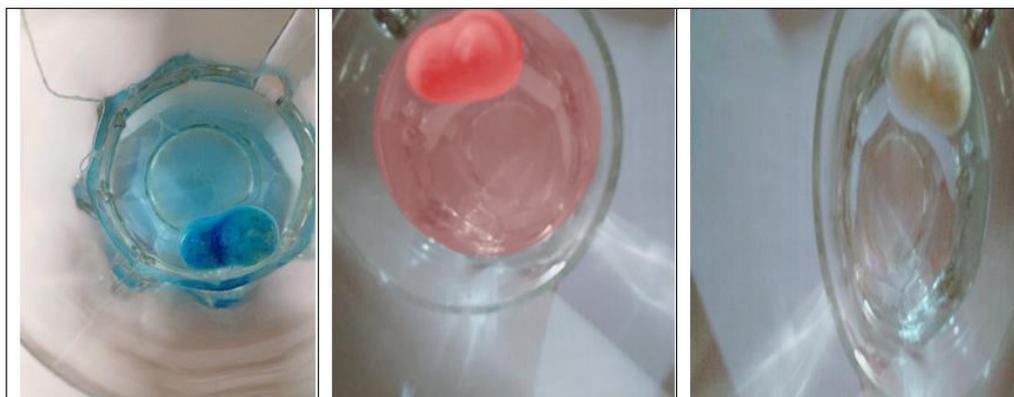
Data in Table (3) presented the statistical simple correlation between the weight of the silk gland and both of cocoon shell and shell ratio. Data cleared that there was a positive correlation between the parameters. The red color presented a highly significant correlation between silk gland weight both the cocoon shell weight and shell ratio (0.98 and 0.99 \*\*), respectively. Also, the blue color showed a highly significant correlation between cocoon shell weight and shell ratio with silk gland weight (0.82 \* and 0.89\*\*), respectively. This agreement with Mahesha *et al.* (2013), studied the level of correlation coefficient and regression on six pure silkworm breeds between larval weight, larval duration, cocoon weight, shell weight, shell ratio, filament length, denier and renditta. Data cleared that, there was a positive and significant correlation between silk gland and both shell weight and shell ratio. Also, Shaki (2015) and Taha and Shaki (2021), reported that there were an appositive and a highly significant correlation between the weight of silk gland and the economic parameters of silkworm after using different nutrition on mulberry leaves.

**Table 3:** The simple correlation "r" between silk gland & cocoon weight, shell weight and cocoon shell ratio during the three successive rearings.

Food colorants	The effect of	Cocoon Weight (mg)	Cocoon shell weight(mg)	Shell Ratio %
<b>Red</b>	<b>Silk gland</b>	0.16	0.98**	0.99 **
<b>Blue</b>		0.82*	0.89 **	0.40

\*\* Correlation is significant at the 0.01 level.

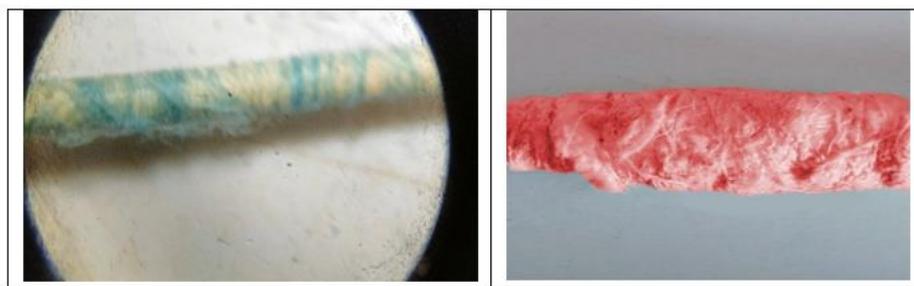
\* Correlation is significant at the 0.05 level.



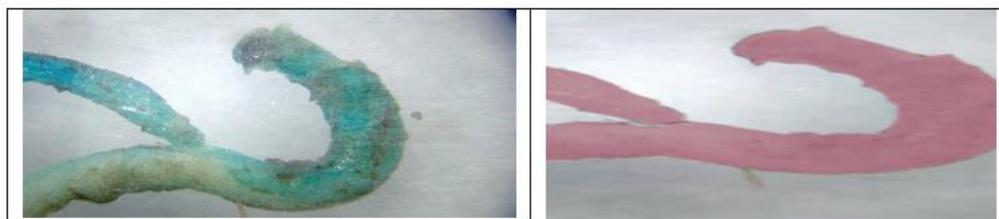
**Fig. 3:** The test of boiling and bleaching to remove colors. After 30 minuet's in boiling water (100<sup>0</sup>c) cocoon washed and treated according to the method of ( Suna *et al.*, 2019). The color didn't remove.



**Fig. 4:** The cocoon is colored blue and red after feeding with food colorants compared with the control.



**Fig. 5:** Cleared the silk colored red and blue after reeling compared with the control.



**Fig. 6:** Showed a part of the silk gland colored with the two colors of food colorants.

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## ARABIC SUMMARY

مكسبات اللون الغذائية كتقنية حديثة للحصول على شرانق ملونة من دودة القز (بومبيكس موري) وتأثيرها على الصفات الاقتصادية

شيماء يوسف عيسى شقل

قسم وقاية النبات – كلية الزراعة والموارد الطبيعية – جامعة أسوان .

تهدف الدراسة الى الحصول على تقنية حديثة ورخيصة الثمن لتلوين خيط الحرير الناتج من دودة القز بدون التأثير على الصفات الاقتصادية للخيط خاصة ان تكلفة استخدام المكسبات اللون الغذائية رخيصة بالمقارنة باستخدام الصبغات الصناعية

ولذلك تم استخدام نوعين من الالوان المستخدمة فى تلوين الحلويات (مكسبات اللون الغذائية) لتلوين شرانق الحرير وهما اللونان الاحمر والأزرق.

وأظهرت النتائج وجود تأثير موجب ومعنوي للونين كليهما على الصفات الاقتصادية مقارنة بالكنترول وعدم وجود اى فروق سلبية على الصفات الاقتصادية بعد استخدام هذه الملونات

كما سجل اللون الأزرق أعلى متوسط فى وزن غلاف الشرنقة (0.284جم) وكذلك معامل غلاف الشرنقة (20.87%) .

للحصول على شرانق ملونة بطريقة صحية ورخيصة وأمنة نحتاج الى المزيد من الدراسات فى هذا النحو.