

# Optimized Rearing Conditions for Silkworm *Bombyx mori* L. Egg Production Under Egyptian Conditions

H. Greiss\*, N. Petkov\*\*, and Miao Yungen\*\*\*

\*Agromier Co, 9 Malek El Afdal Str. 11211 Zamalek, Cairo, Egypt

\*\*Sericultural Experiment Station, 24 Mito Orozov Street, Vratza 3000 Bulgaria

\*\*\*Department of Sericulture, Zhejiang University, Hangzhou, 310029, China

**Optimized package of rearing conditions including adding secondary macro (Ca<sub>230</sub>, S<sub>50</sub>, & Mg<sub>55</sub>) and micronutrients (Mn<sub>3</sub>, Fe<sub>4.5</sub>, Zn<sub>3.5</sub>, Cu<sub>0.4</sub>, B<sub>0.5</sub>, Mo<sub>0.54</sub> & Cl<sub>200</sub>) to the mulberry garden beside the 1<sup>ry</sup> macronutrients (N<sub>300</sub>, P<sub>150</sub>, K<sub>120</sub>) per hectare in split doses after every leaves harvest. Disease free rearing management by double house disinfections, eggs, bed and larval body disinfection. Optimum physiological needs that include low temperature rearing for young instars and natural mounting for silkworms improved the egg production. This package for seed silkworm rearing led to the increase in fecundity by 67-121 eggs (15.12-26.22%) and yield of standard boxes per 1 parent egg boxes by 57-78 egg boxes (48.33-51.66%) respectively in comparison to the traditional cocoon production technology under Egyptian conditions. This technology is highly ecological with the elimination of formol. Also, the farmyard manure with its bulkiness and the possibility to transmit harmful nematodes and unwanted weeds to the mulberry garden was eliminated without reducing the nutritive value of the mulberry leaves.**

**Keywords:** Macro-nutrients, micro-nutrients, *Morus alba*, *Bombyx mori*, acetic acid, fecundity, egg box.

The adoption of the high silk-yielding P<sub>1</sub> pure races in the field would greatly improve the quality of hybrid F<sub>1</sub> seeds, however it is believed that the high yielding pure races have very poor pupation, and low fecundity. Improving viability and fecundity will accordingly increase silkworm egg production and the profits of both the farmers and the grainages, as well as improve the overall silk production of the hybrid F<sub>1</sub> eggs. For this purpose, parent silkworm growers must provide optimum rearing conditions, highly nourishing food and strong prophylaxis against different pathogens. From the economic point of view, rearing risks should be minimized assuring stable cocoon production throughout the year.

Due to continuous depletion of the micronutrients

from the soil by the mulberry, the soils become deficient in trace elements. (Singhvi et al. 1998). However information regarding micronutrients needed for plant growth in general and mulberry growth in particular is very little. (Lokanth & Shivashankar 1986).

Ikegami (1995) studied the effect of Chlorotic chemicals as a substitute to bleaching powder for disinfections of the silkworm rearing houses and equipment. Bleaching powder is the most suitable disinfectant for open type rearing houses. (Baig & Pradip Kumar 1987).

The higher value of silkworm pupation rate after egg surface disinfections is due to the effectiveness of used disinfectants against incorporation of pathogens on the egg surface and subsequently infecting the newly hatched larvae (Troitskaya 1991, Golovko 1995).

\*Corresponding author: miaoyg@zju.edu.cn

Disinfecting the eggs by acetic is a high ecological method (Kirichenko et al. 1995). Its germicidal effect has long been proven.

Manshev (1978) tested the efficiency of a mixture of active lime and bleaching powder 9:1 and reported its positive effect as a bed disinfectant. Barman (1991) studied the effect of some formulated chemical composites as larval body disinfectants for controlling viral and bacterial diseases of silkworm *B.mori* and reported the decrease of overall mortality over the control.

Tankov P (1909), Bastiani (1991) recommended rearing during the 1<sup>st</sup> and 2<sup>nd</sup> instars at 23-25°C. The body temperature of the silkworms influences the physiological activities, food intake and economic parameters. (Tzenov & Maldenov 1996, Muniraju et al. 1999). However we found that constant temperature rearing at 23°C saved heating costs and improved cocoon yield (Greiss et al. 2000).

Even if the silkworm crop is healthy, incorrect mounting methods, spinning conditions, mounting density, pre or over mature larval mounting and bad type of mountages can result in spinning of inferior quality cocoons. (Ullal & Narisimhanna 1987). The self-mounting method is the natural urge of the matured silkworm to climb up to start spinning cocoons. (Tzenov & Petkov 1999, Greiss & Petkov 2000a). It follows the natural physiology of the insect and reduces stress.

In the recent years new silkworm *B.mori* L. races and hybrids, having higher productivity have been selected in different countries. (Petkov & Nacheva 1996, Datta et al. 1999). However these highly productive races and hybrids require excellent feeding and rearing conditions in order to manifest full extent of their potential. (Datta et al. 1999). We combined all the positive elements described previously and created a package of practices we called improved technology.

## Materials and methods

Practical experiments were held in the Agromier Co. SAE mulberry plantation and rearing houses located in the Asyut province of Egypt 320 km south of Cairo in a desert area. The plantation is irrigated by water from the Nile river. Plants are watered regularly. Total salinity of irrigating water is 300 PPM.

Mulberry plantation is mainly of Kokuso 27 & Canva 2 varieties, shaped in bush type and pruned in fist form after 3 step up branch harvests every year. Soil is sandy loam, consisting of 51.4% silt, 8.8% clay and 39.8% sand. Soil pH is 8.1. The underground water table is below 3 meters. Moisture content at field capacity is 24%.

Rearing was performed using plastic trays 70cm x 40cm x 30cm (L x B x H) accommodating 100 larval

counted just after 2<sup>nd</sup> moult. During each experiment all the larvae were fed by equal amounts of leaves of the same variety. Bed spacing was performed everyday before morning feed, and bed cleaning was performed once after each moult.

First instar larvae were fed by leaves number 1&2, chopped at 5mm strips. (Largest Glossy Leaf [LGL] counted as leaf number 1). Second instar larvae were fed by leaves number 3&4, chopped at 10mm strips. Beds were covered by nylon sheet to preserve humidity in the beds during the first two instars. Third instar larvae were fed by shootlets with the upper seven leaves. Fourth and fifth instar larvae were fed by whole branches, 50 - 70 days old.

Bottlebrush type plastic mountages were used for cocoon spinning. Oviposition was done on cardboard sheets divided into 5 rows and 4 columns to give 20 equal squares 5cm x 5cm. Fertilized moths are engaged into iron funnels and allowed to oviposit. Iron funnels have a lower diameter of 5cm and upper diameter of 4cm. Mother moths were put in paper bags to lay eggs when loose eggs were needed.

The control and each experiment were maintained in 4 replicates of 100 larvae each of eleven breeds of silkworms (Table 1) during spring rearing of years 1999 and 2000.

### Control (Traditional Technology)

As described in literature. (Ullal & Narisimhanna 1987).

1. Fertilization of the mulberry plantation by 1<sup>st</sup> macro-nutrients (N<sub>300</sub>P<sub>150</sub>K<sub>120</sub>) and FYM<sub>15ton</sub>.
2. Disinfecting the rearing house by spraying Formol 3% solution plus 1 liter of active lime milk Ca(OH)<sub>2</sub>/18 lit solution, full wetting of the walls and appliances in an air tight chamber and temperature maintained above 20°C
3. Rearing houses were equipped with automatic temperature and humidity control devices and the rearing was done at standard regimes of temperature and humidity (Table 2.)
4. Rearing was performed using plastic trays 70cm x 40cm x 30cm (L x B x H) accommodating 100 larvae counted just after 2<sup>nd</sup> moult. During each experiment larvae were fed equal amounts of leaves of the same variety. Bed spacing was performed every day before morning feeding, and bed cleaning was performed after each moult
5. Feeding criteria as follows:
  - First instar larvae were fed by leaves number 1&2, chopped at 5mm strips. (Largest Glossy Leaf [LGL] counted as leaf number 1).
  - Second instar larvae were fed by leaves

Table 1. Egg production capacities.

Egg	Egg										
	EY	Ed	E'3	E'	E3a	E'4	E4b	E3	E4	E32	E'3
<b>Hatchability (%)</b>											
Exp	95.49	95.49	95.9	95.95	95.92	95.88	95.97	95.22	95.38	95.12	95.01
Cont	95.88	94.92	95.39	95.38	95.55	95.33	95.71	95.49	95.92	95.94	95.77
TD	-0.39	0.57	-0.50	-0.04	-0.23	0.14	-0.94	-0.24	-0.07	-0.22	-0.18
<b>Incubation (%)</b>											
Exp	93.19	93.19	93.50	93.25	93.98	93.98	93.00	93.19	93.00	93.00	93.00
Cont	91.25	90.82	90.37	92.00	91.37	92.00	91.97	92.00	91.82	90.50	91.00
TD	17.19	17.51	17.83	18.25	18.51	18.98	18.83	18.19	18.98	17.50	17.00
<b>Fecundity (eggs)</b>											
Exp	539	513	574	578	532	587	545	510	515	487	454
Cont	482	441	472	492	477	450	498	449	495	370	370
TD	121	77	102	98	105	117	109	87	30	97	34

\*P<0.05, \*\*P<0.01, \*\*\* P<0.001  
Exp - Experiment, Cont - Control, TD - True Difference

Table 2. Temperature and humidity table.

Age	Temp.	Humidity	Light	Air speed
1&2 instars	27°C	85% RH	25 Lux	0.1 m/sec
3 instar	25°C	80% RH	25 Lux	0.1 m/sec
4&5 instars	23°C	70% RH	25 Lux	0.3 m/sec
Mounting & spinning	27°C	60% RH	25 Lux	0.5 m/sec

- number 3&4, chopped at 10mm strips. Beds were covered by nylon sheet to preserve humidity in the beds during the first two instars
  - Third instar larvae were fed by shootlets with the upper seven leaves.
  - Forth and fifth instar larvae were fed by whole branches, 50-70 days old.
6. Mounting by bottlebrush type plastic mountages by picking.
  7. Oviposition was done on cardboard sheets divided into 5 rows and 4 columns to give 20 equal squares 5cm X 5cm. Fertilized moths are encaged into iron funnels and allowed to oviposit. Iron funnels are having a lower diameter 5cm and upper diameter 4cm. Mother moths were put in paper bags to lay eggs when loose eggs were needed.

**Experiment (Improved Technology)**

The following modifications were superimposed on the control:

1. Application of 2<sup>ry</sup> macro-nutrients (Ca<sub>230</sub>, S<sub>50</sub> & Mg<sub>55</sub>) and micro-nutrients (Mn<sub>3</sub>, Fe<sub>4.5</sub>, Zn<sub>3.5</sub>,

Cu<sub>0.4</sub>, B<sub>0.5</sub>, Mo<sub>0.54</sub> & Cl<sub>200</sub>) to mulberry garden beside the 1<sup>ry</sup> macro-nutrients (N<sub>300</sub>, P<sub>150</sub>, K<sub>120</sub>) and adding FYM<sub>15ton</sub> only for young instar rearing garden.

2. Double disinfecting by bleaching powder 5% W/V and acetic acid 0.2% for house and appliances.
3. Eggs disinfecting by acetic acid 0.04% for 15 min. at room temperature.
4. Bed & larval disinfections by a dust mixture of:
 

Calcium Oxide	88%
Bleaching powder	10%
Benzoic acid	1%
Diathane M 45	1%

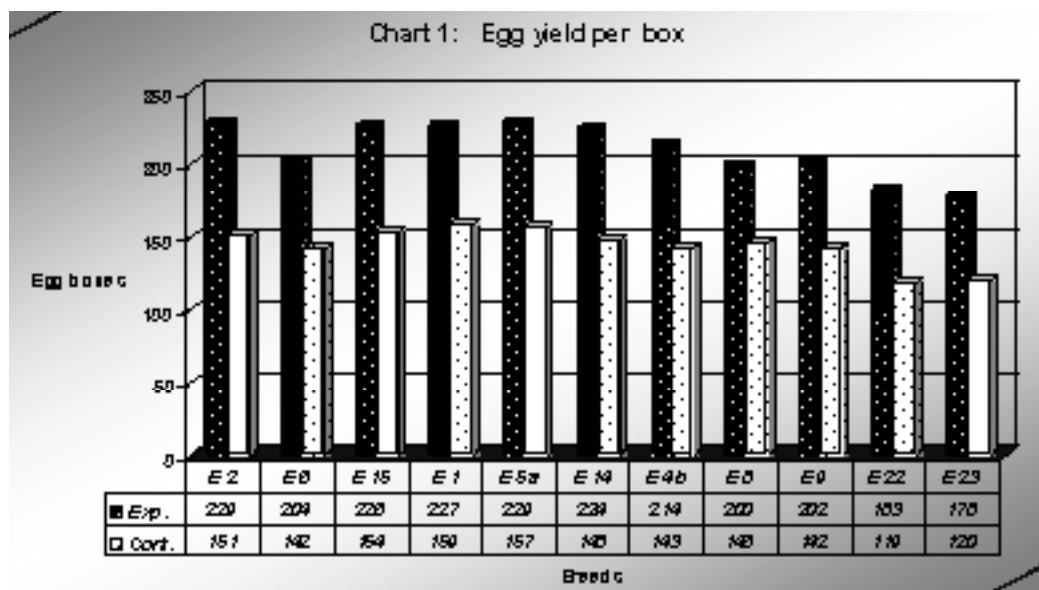
After every moult at 250gm/m<sup>2</sup> for young instars and 500 gm/m<sup>2</sup> for late instars of bed area.

5. Rearing temperature at 23°C, light intensity 25 Lux all over the larval period. Air speed and relative humidity at 0.1m/sec & 85% for young instars and 0.3m/sec & 70% for late instars respectively. During molting the humidity was reduced to 50% RH in all instars, and in the case of young instar larvae, nylon covers were removed from the beds.
6. Natural mounting and spinning at 27°C, light intensity 25 Lux, air speed 0.5m/sec and RH. 60%.

**Quantitative calculation**

Egg box yield per 1 egg box (EY): it was calculated using the formula. (Result in egg boxes of same category):

$$EY = \frac{H \times PR \times ME \times F}{2}$$



Where  $EY = \text{Egg box yield per 1 box}$ ,  $H = \text{Hatchability}$ ,  $PR = \text{Pupation rate}$ ,  $ME = \text{Moth emergence percentage}$  and  $F = \text{Fecundity}$ .

## Results and discussion

Data in table 3 show that the tested element of the new technology (egg surface disinfections with 0.04% acetic acid for 15 min at room temperature) was not of any significant influence on egg hatchability (I). Hatchability show very slight increase or decrease over the control  $\pm 0.7\%$ . This proves once again that the new method for egg surface disinfections did not exert any harmful effect on the embryos inside the eggs or the newly hatched larvae.

The new technology leads to significant increase in the pupation value. In average for the period of investigation in comparison to the traditional technology, silkworms' pupation ratio (II) was increased by 16.13-17.63%. Comparatively highest differences were recorded in breeds with Japanese origin E2, E6 & E15, and genetically Sex limited breeds (E22 & E23).

The proposed improved technology lead to significant increase in fecundity (III) and accordingly to the egg yield per 1 box of parent silkworms. On average for the period of investigation in comparison to the traditional technology, and irrespective to the genetic and geographic origin of the tested breeds the fecundity increased by 77-121egg (17.46-26.19%) for Japanese breeds, 96-117egg (19.92-26.00%) for Chinese breeds, 67-109egg (15.12-25.00%) for European breeds and 84-97 egg (22.70-26.22%) for Sex-limited breeds.

The improved technology increased the calculated yield of egg boxes per 1 egg box by 54-72egg boxes

(36.99-47.37%) chart 1, due to the increase of the pupation ratio together with fecundity. It should be noted that our formula for calculation gives the result in the same standard egg box used, irrespective of standard number of eggs per box used, as standard number of eggs per box is different in different countries.

## Conclusion

Implementation of improved technology for seed silkworm rearing lead to increase in fecundity by 67-121eggs (15.12-26.22%) and yield of standard boxes per 1 parent egg boxes by 57-78 egg boxes (48.33-51.66%).

## References

- Ajuzawa (1972) Handbook of silkworm rearing. Fuji pub. Co. Ltd. Tokyo Japan p380.
- Baburasvilli E.L., Amirashvili, L. Nonikashvili (1988) Chemical therapy of the pebrine disease of the silkworm *Bombyx mori*, "Ways for sericulture development". Tblisi 72-79p.
- Bastiani D.(1991)Il Baco da Seta come si alleva, REDA, Roma, 125 pp.
- Barman A.C. (1991) Effect of some formulated chemical composites for controlling bacterial and viral disease of silkworm *Bombyx mori* L. Bangladesh Seric. Res. And Training Inst Bangladesh, Bull.Sricult.Res. 2,62-66.
- Baig M., Pradip Kumar (1987) Silkworm diseases and pest. Chapter V. Appropriate Sericulture Techniques.

- International Center for Training and Research in Tropical Sericulture, Mysore, 127-132.
- Baig M. (1987) Silkworm disease and their prevention. A handbook for sericulture extension workers. CSR & TI Mysore 168-183
- Datta R.K., Rajan R.K., Nataraju B., Basavraju H.K., Sarkar A. (1999) Popularization of bivoltine sericulture technology in Karnataka, "XVIII International Sericultural Congress", Cairo, Egypt 12-16 October 1999, 73-80.
- Golovko V.A. (1995) Ways of increasing silkworm resistance against disease and unfavorable environmental factors Kharkov, RIP "Original" 171 pp
- Ikegami T. (1995) Effects of Chloric chemicals substitute for bleaching powder by way of disinfectant of rooms and tools for silkworm rearing. Bull. Seric. Res. Inst. Ibaraki Agric Cent, 3, 58-64.
- Kirichenko I. A. (1995) Main infectious diseases of silkworm in the Ukraine and their control methods. Kharkov, RIP "Original" 201 pp.
- Lokanath R., Shivashankar K. (1986) Effect of foliar application of micronutrients and magnesium on the growth and quality of mulberry (*Morus alba* Linn.) Indian J. Seric. 25(1) 1-5.
- Manshev M., PhD thesis Sofia (1978) Study of epizotology of Nuclear Polyhedral Virus of *Bombyx mori*. p.35.
- Muniraju E., Sekharappa B.M., Raghuraman R. (1999) Effect of temperature on leaf-silk conversion in silkworm *Bombyx mori* L. Sericologia, 39(2), 225-231.
- Michailov E. (1984) Infectious disease of the silkworm *Bombyx mori* L. Shelkoprijad, Tashkent, p. 296.
- Petkov N., Natcheva Y., (1996) Study on the economical characters of complex (tetra) hybrids of the silkworm *Bombyx mori* L. I. Biological traits of the larvae Animal Science XXXIII 4,63-67.
- Petkov, Z. (1988) Utilization of mulberry leaves from introduced in Bulgaria mulberry varieties by silkworm, Animal science, XXXV, 24-29.
- Singhvi N.R., Munizanthanam Reddy M., Srinivasan F.B. (1998) Management of Zinc in Mulberry, Indian Silk, 4, 13-14.
- Singhvi N.R., Subbaswamy M.R., Datta R.K. (1998) Magnesium management in mulberry. Indian Silk 3, 7-9.
- Tankov P (1909) New lesson in sericulture. Vratza. 714pp. Bulgaria
- Troitskaya Y.N. (1991) Epizotology of the bacterial toxicosis of the silkworm *Bombyx mori* L. Uzbekistan, Shelk, 6,11,13p.
- Tzenov P., Maldenov G. (1996) Effect of temperature during the 5<sup>th</sup> larval instar on the values of the biological and reproductive characters in the silkworm *Bombyx mori* L. Bulg. J. Agric Sci., 2, 247-257.
- Tzenov P., Petkov N. (1999) Improved labor saving technology for silkworm rearing in Bulgaria. "XVIII ISC congress" 12-16 October 1999 Cairo Egypt 92-96. Petkov N. (1995) Selection genetic investigation and results from the Silkworm (*Bombyx mori* L.) races, lines and hybrid breeding. Thesis for obtaining degree Doctor of the Agricultural Science, Sofia 305p.
- Ullal S.R., Narisimhanna .N. (1987) Handbook of practical sericulture. CSB 126-130 & 213.