## Some Factors Affecting Mating Activity of Male Moths and Egg Production of *Bombyx mori* L.

Mona M. Mahmoud and Wagiha H. Yehia<sup>1</sup>

### ABSTRACT

In order to facilitate silkworm breeding processes to produce and increase the industrial eggs, the effects of cold storage as well as the multiple mating by *Bombyx mori* L. male moths on the fecundity of female moths were studied. The obtained results indicated the possible and useful use of the cold stored male moths up to five days for mating processes. Furthermore, it can be also safely reuse the freshly and healthy males for three matings.

#### **INTRODUCTION**

It is well known that the industrial eggs are generally specific hybrids between two or more recommended lines or races of silkworms Bombyx mori L. and are reared by the sericulturists for producing cocoons on a commercial scale. In order to prepare the hvbrid eggs by crossing the parental races, Krishnaswamy et al (1973) showed that care must be taken to ensure that the moths emerge simultaneously. That could be achieved by rearing the parental races which have the same larval duration that all moths emergence at about the same time. Herein, In spite of the care taken, there should be still differences in the emergence of moths of parental races. So, male moths can be safely stored for about a week at temperature of 7°c for mating purpose after emergence, because they have more resistance for cold storage than female moths (Tajima, 1972).

On the other hand, Krishnaswamy *et al* (1973) stated that while preparing the hybrid eggs, if there is a scarcity of males, the male moths can be used for a second mating after resting for some time between the two matings. Ram and Singh (1992) found that approximately two hours of mating is sufficient, since during this period at least two ejaculations occur, the first during the first 30 minutes and the second after one or one and half hours.

Therefore, the present investigation aimed to study the effect of cold storage on the fertility of *B. mori* male moths, as well as, the efficient of multiple mating by these male moths on production of viable eggs.

#### MATERIALS AND METHODS

Experiments were performed on the Chinese hybrid 9F7X of the mulberry silkworm *Bombyx mori* L. The larvae were reared in the laboratory following the

adopted improved method of silkworm rearing by Krishnaswamy *et al* (1973). Larvae were fed on fresh mulberry leaves under the hygrothermic condition of 22  $\pm 1.5^{\circ}$ c and 82  $\pm 3\%$  R.H.

To obtain the experimental male moths, the sex discrimination was conducted at the end of the pupal stage according to the method adopted by Yokoyama (1959) on the basis of the presence of a small hole on the  $10^{\text{th}}$  abdominal segment in the female, in which absent in the male pupa.

After the emergence of moths, the experiment was divided into two sets, for the first set, one hundred healthy freshly emerged male moths were taken and kept in the fridge for cold storage at temperature of  $7^{\circ}$ c up to 8 days.

A group of ten male moths were daily taken off from the fridge and allowed to mate with freshly female moths, thereafter each couple was kept in a perforated paper bag till the end of egg deposition; the deposited eggs per female moth were counted and recorded.

The second set of emerged male moths was randomisely taken to study the effect of reuse of them in multiple mating. Freshly emerged male moths were allowed to mate with female ones. During copulation, the coupling pairs were kept in a temperature of  $24 \pm 1.5^{\circ}$ c. They were left up to one and half hours and then separated by holding the female lightly and moving the male sideways with the fingers of right hand, to facilitate easy separation without injury to the female genetalia.

These same separated male moths were reused to mate with other females. This procedure was repeated four times with an interval of one and half hour between each separation. After each separation each female moth was kept individually in perforated paper bag. After oviposition, numbers of deposited eggs per female moth were counted and recorded. A simple correlation between age of male (in hours) and number of deposited eggs of experimented female moths was considered.

Data were statistically analyzed using "F" test and least significant difference (L.S.D.) at 5% probability level (Snedecor and Cochran, 1978).

<sup>&</sup>lt;sup>1</sup>Dep. of sericulture, Plant Protection Res. Institute

ARC, Alexandria, Egypt

Received March17, 2009, Accepted March26, 2009

#### **RESULTS AND DISCUSSIONS**

# Effect of cold storage of male moth on the fecundity of *B. mori* female moth:

The results presented in Table 1 illustrate the effect of daily periodic storage of male moths at  $7^{\circ}c$  on the rate of deposited eggs by female moths.

The results indicated the insignificant differences in the counted numbers of deposited eggs between the normal mated couples (control, 363.8 eggs/female) which were left for mating just after emergence and these laid by the female moths which were mated with cold stored male moths till the 5<sup>th</sup> day of storage.

A significant decrease was obtained in the number of laid eggs by mated females with cold stored males for 6 days (312.1 eggs/female); it was somewhat lower than the above mentioned results of the first five days as well as the control treatment, (Table 1). Concerning the 7<sup>th</sup> and 8<sup>th</sup> days of cold storage, the number of deposited eggs significantly was much more decreased.

Takemura *et al.* (1999) discussed the effect of long – term storage of male moths at  $5^{\circ}$ c and showed that only 30% of moths were able to survive after 2 months of storage; moths lost their mating ability.

# Effect of reused male moths for mating on the fertility of *B. mori* female moths

A male moth is capable to mate with certain number of females without any defect on fecundity (Jadav and Gajare, 1978; Askari and Saran, 1984; Ram and Singh, 1992; Vijayan *et al* 1994), after that both fecundity and fertility were reduced.

As shown in Table 2, there were no significant differences in number of laid eggs per female moth from the first mating of male moth until the third one. (371.8, 364, 359.3 eggs/female in respect). The same male moth was capable for doing the fourth mating since the average of resulted eggs per female moth (254 eggs/female) was significantly lower than that per female in the aforementioned three matings. (Table, 2).

Arnold and Zoraida (2003), studied the productivity of male moths that were used repeatedly, they showed that a male moth can mate only two females, third and fourth mating be inevitable.

There are some factors affecting mating to ensure the success of this process; age of moth on mating, frequency and duration of mating, reuse of male moth for successive mating, impact of multiple mating (Sengupta *et al* 1973; Yamaoka and Hirao, 1981; Paul *et al*, 1993; Suzuki *et al*, 1996 and Singh *et al*, 2003). In addition to, the longevity of male moth before mating (Takemura *et al*, 1999).

# Correlation between male moth age and female moth fecundity in silkworm *B. mori* L.

The results presented in Fig. (1) clarify the calculated simple correlation coefficient value between the age of male moths (in hours) and the egg production of the mated female moths. The calculated value of significant negative correlation (r = -0.878) indicated that the number of eggs laid / female decreased by the increase of age of male moth.

Paul and Kishorekumar (1995) proved that there was no significant difference between age of males and number of eggs laid by the mated females at high temperature in comparison to the male partner's age.

From the above mentioned results it could be concluded that for producing industrial hybrid the cold storage of male moths can be necessarily used in case of delay of female moth emergence.

Reuses of male moths are also accepted due to several reasons such as scarcity of male moths, failure in emergence in the component races and presence of weak males.

Furthermore, it can be useful to use the male moths which gave significant results represented in the cold stored males for 6 up to 8 days as well as the fourth mating despite the more or less decreased number of resulted eggs per mated female moths, in order to increase silkworm seed production in case of occurring any disturbance or difficult circumstances.

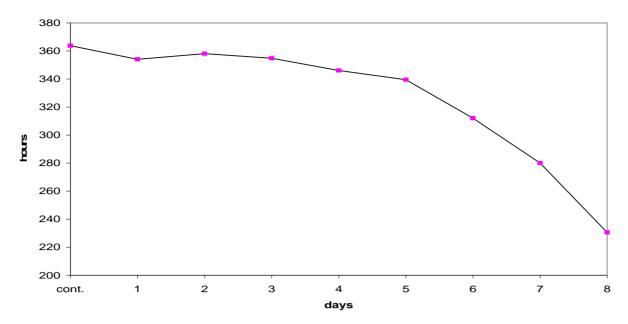
Table 1.	Effect of	f cold stor	age male	moths on	the fecun	dity of <i>Bo</i>	mbyx mo	<i>ri</i> L. fema	ale moths
Days	cont.	1	2	3	4	5	6	7	8

				-		-	-		-
No. of	а	а	а	а	а	а	b	с	d
dep.	363.8	354.1	358.1	354.8	346.1	339.5	312.1	280.1	230.6
eggs	±15.519	$\pm 24.173$	$\pm 24.164$	±20.291	$\pm 20.979$	$\pm 24.744$	$\pm 47.648$	$\pm 37.843$	$\pm 29.609$
Table 2. Effect of reuse of male moths for mating on the fertility of Bombyx mori L. female									

moths

4
a b
359.3 254
9.398 ±53.246

\* No significant differences among the means with the same letters.



### Fig. 1. Correlation between age of male moth and no. of deposited eggs / mated female moth

#### REFERENCES

- Arnold, M. Inurnpa and Zoraida, R. Espiritu. (2003). Productivity of re-used male moths of *B. mori* Textile Research Institute. Vol. (3). ISSN 1656 – 4744.
- Askari, S. and R. K. Saran. (1984). Studies of different copulating duration on pre – oviposition, fecundity and fertility of mulberry silkworm, *Bombyx mori* (Bombycidae : Lepidoptera). J. Adv. Zool. 5, 114 – 119.
- Jadav, L. D. and B. P. Gajare. (1978). Studies on the effect of mating duration on the viability of silkworm (*Bombyx mori*) eggs. Indian J. Seric. 17, 28 – 32.
- Krishnaswamy, S.; M. N. Narasimhanna; S. K. Suryanarayan and S. Kumaraj. (1973). Sericulture manual – 2 – silkworm rearing, FAO, Agriculture Services Bulletin. 15 (2).
- Paul, D. C.; C. M. Kishorekumar and S. K. Sen. (1993). Effect of age at mating on the oviposition, fertility and longevity of female silk moth, *Bombyx mori* L. (Lepidoptera : Bombycidae). Indian J. Seric. 32, 24 – 25.
- Paul, D. C. and C. M. Kishorekumar. (1995). Influence of male age on mating capacity, fecundity and fertility of mated female silk moth, *Bombyx mori* L. under high temperature and high humidity conditions. Central Sericultural Research & Training Institute, Berhampore, 742 101, west Bengal, India. (3/4) 253 – 255.
- Ram K. and Singh Darshan. (1992). Role of mating duration in the production of viable silkworm (*Bombyx mori* L.) eggs. Journal of Entomological Research. Vol. 16 no. (3).

- Sengupta, K.; R. K. Datta and S. N. Biswas. (1973). Effect of multiple crossing on the type of progeny recovered in silkworm *B. mori* L. Ind. J. Seric. 12, 31 – 38.
- Singh, T.; B. Saratchandra and H. S. Phaniraj. (2003). Physiological and biochemical modulations during oviposition and egg laying behavior in the silkworm, *Bombyx mori* L. Int. J. Indust. Entomol. 6, 115 – 123.
- Snedecor, G. W. and W. G. Cochran. (1978). Statistical methods. The 9<sup>th</sup> ed. Lower state University Press, Ames : lower, U.S.A.
- Suzuki, N.; T. Okuda and H. Shinbo.(1996). Sperm precedence and sperm movement under different copulation intervals in the silkworm *Bombyx mori* J. Insect. Physiol. 42, 199 – 204.
- Tajima, Y. (1972). Hand book of silkworm rearing. Copyright, Fuji Publishing Co., Ltd.
- Takemura Yoko; Kanda Toshio and Horie Yasuhiro. (1999). Effect of long term cold storage of male pupae and moth on reproductive ability of sperm in the silkworm, *Bombyx mori* L. Journal of sericultural Science of Japan. Vol. 68 no. (2) pp. 133 – 137.
- Vijayan, V. A.; G. Subramanya and N. B. Krishnamurthy. (1994). Differential mating capacity of male moths in six races of silkworm *Bombyx mori* L. Bull. Sericult. Res. 5, 77 – 79.
- Yamaoka, K. and T. Hirao. (1981). Mechanism of ovipositional behavior in *Bombyx mori* time gating and accumulation of the internal factor. Intl. J. Invertebr. Repod. 4, 169 – 180.
- Yokoyama, T. (1959). Silkworm genetics illustrated. Lap. Soc. Prom. Sci., Tokyo. P. 185.

# الملخص العربي

# دراسة بعض العوامل المؤثرة على نشاط التلقيح لذكور ديدان حرير القز على إنتاجية البيض من ماهر عمود، وجبهه حسين يحي

تم دراسة تأثير التخزين البارد لذكور فراشات ديدان حرير القز الذكور و التي اجرى لها تخزين بارد حتى خمسة ايام فى التلقيح و و ايضا استخدام الذكور لتلقيح أكثر من انثى و ذلك لتــسهيل ايضا يمكن استخدام الفراشات فى تلقيح اكثر من انثى حتى ثلاثة عمليات التربية لإنتاج بيض الهجن و الذى يــتم تربيتــه تجاريــا. فراشات إناث مما ادى إلى انعكاس ذلك على كفاءة خصوبة اوضحت النتائج المتحصل عليها أنه من المفيد استخدام الفراشات