PUBLICATIONS RELATED TO THIS WORK

Brief note

STUDIES ON SEED (EGG) PRODUCTION WITH REFERENCE TO MATING BEHAVIOUR OF MUGA SILKWORM, *ANTHERAEA ASSAMA* WESTWOOD*

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The experiment was conducted in Muga Research Laboratory, A.B.N. Seal College, Cooch Behar to overcome the constraints regarding seed (egg) production of Muga silkworm ***Antheraea assama*** *Ww. through manipulating their mating behaviour to standardize the mating hours and utilisation of male moth for more than one time for better supply of quality seed to farmers. Mating duration of 7 hours showed highest fecundity and hatching. Utilisation of male moth up to 3 times during March-April and up to 4 times during Sept.- Oct. had no harmful effect on fecundity as well as hatching. It could be suggested that 7 hours mating would be optimum and the same male moth could be utilised up to three / four times without any hampering effect on egg production and hatching for better supply of eggs to the farmers for muga culture.*

Keywords: Muga silkworm, egg laying, egg retention, hatching, repeated mating

INTRODUCTION

Muga Silkworm (*Antheraea assama* Ww.) is a highly heterogeneous, unique and semi-domesticated strain of Saturniidae endemic to Assam, adjacent foothills of Meghalaya, Nagaland, Arunachal Pradesh and Mizoram. Exploration of Muga culture in Terai region of West Bengal particularly in Cooch Behar and Jalpaiguri districts having contiguity with Assam faces problems related to supply of egg. Production of large number of eggs for farmers is a serious problem in Assam (Sahu *et al.,* 1998). Uneven sex-ratio, poor seed cocoon and its preservation and asynchronous moth emergence resulting in poor mating and poor number of egg production are the principal constraints other than environmental stresses (Samson and Barah, 1989). Though works related to male-female ratio and seed cocoon production through improvement of rearing technology are in progress (Annual Report, RMRS, CSB, Boko, Assam, 1997), efforts to overcome asynchronous moth emergence to ensure higher mating percentage leading to higher fecundity is very limited in muga silkworm and the approach is only to synchronise male and female through short-time preservation (Khanikor and Dutta, 1989). In mulberry silkworm (*Bombyx mori* L.), some works have been attempted by exploiting male moths for more than one time to overcome the sex ratio problem as well as asynchronous moth emergence (Benchamin *et al.,* 1990). In Tasar silkworm (*Antheraea militta*) repeated mating has been adopted as practice (Ojha *et al.,* 1996). In the present investigation, an attempt has been made to overcome the constraints in muga silkworm through manipulating their mating behaviour to standardize the mating hours and utilisation of male moth for more than one time for better supply of quality seed to the farmers.

* (Paper presented at 90th Indian Science Congress, Bangalore, 3 – 7th January, 2003.)

MATERIALS AND METHODS

Seed cocoons were collected from Extension Centre (Cooch Behar, West Bengal), Regional Muga Research Station, Central Silk Board and the experiment was conducted thrice during 2001-2002 in muga research laboratory, A.B.N. Seal College, Coot Bear. Eleven (11) treatments for standardisation of mating hours were selected from 2 hours to 12 hours with one hour interval, egg laying and retention were taken as quantity production parameters and hatching number and percentage were taken as quality production. Subsequently in another experiment males were exploited up to a maximum of 6 times during Sept.-Oct. and four times during March-April with fresh female and egg laying, retention and hatchability were taken as key parameters. All data collected were analysed statistically.

RESULTS

a. Effect of different mating hours on egg laying and hatching:

No significant differences were observed between different mating hours on egg laying, retention and hatching. However, highest fecundity was observed when mating was allowed for seven hours (287), lowest being from 12 hours (250). Egg retention was nearly nil to 11 hours. Number of egg hatched was highest from seven hours (284) which was 99.19%, lowest being from 12 hours which was 235 and 94% for egg laying and hatching respectively (Table I).

Table I. Data on egg laying and hatching as influenced by different mating hours.

Tableau I. Données sur la ponte et l'éclosion sous l'influence de différentes heures d'accouplement.

Mating Hours *Heures d'accouplement*	Egg laying (no.) *Ponte des œufs (nbre)*	Egg Retention (no.) *Rétention d'œufs (nbre)*	Hatching (no.) *Eclosion (nbre)*	Hatching percentage (%) *Pourcentage d'éclosion (%)*
2 hrs.	267	10	253	95
3 hrs.	269	9	265	99
4 hrs.	273	9	266	97
5 hrs.	280	6	277	99
6 hrs.	279	6	273	98
7 hrs.	287	2	285	99
8 hrs.	276	8	267	97
9 hrs.	273	5	266	97
10 hrs.	252	4	250	99
11 hrs.	251	3	247	98
12 hrs.	250	6	235	94
CD at 5%	Non-Significant *Non significatif*	Non-Significant *Non significatif*	Non-Significant *Non significatif*	Non-Significant *Non significatif*

Table II. Data on grainage performance as influenced by repeated mating during March-April.

Tableau II. Données sur la performance de grainage sous l'influence d'un accouplement répété en mars – avril.

Treatment ***Traitements***	**Egg laying (no.)** ***Ponte d'œufs (nbre)***	**Egg Retention (no.)** ***Rétention d'œufs (nbre)***	**Hatching (no.)** ***Eclosion (nbre)***
Fresh male × fresh female *Nouveau mâle × nouvelle femelle*	232	7	231
Single mated male × fresh female *Mâle accouplé une fois × nouvelle femelle*	207	7	197
Double mated male × fresh female *Mâle accouplé deux fois × nouvelle femelle*	194	15	164
Triple mated male × fresh female *Mâle accouplé trois fois × nouvelle femelle*	75	140	0
C.D. at 5% / *DC à 5 %*	**3**	**9**	**4**

b. Effect of number of matings utilising single male moth on egg laying and hatching:

Results obtained from repeated mating during March-April were depicted in Table II and that of during September-October in Table III.

During March-April:

During March-April, prior to second commercial crop rearing period i.e. April-May one male moth could be forced to couple with fresh female up to four times.

Egg laying:

Significant variations observed between the treatments. Highest egg laying was recorded when fresh male and female moths were coupled (232). A decreasing trend was observed from second time utilization to 4th time utilization of the same male moth. There observed significant variation in egg laying between second (207) and third time (194) exploitation of the same male moth. But significantly abrupt decreased egg laying was observed when extreme utilization of male moth for the fourth time was done (75).

Egg Retention:

Egg retention was not significantly higher inside the female body up to 3rd time utilization of a single male (7 to 15). But the egg retention in the female body when coupled with a male utilising for the 4th time was very high (140) which was even nearly double than the egg laid by that female.

Table III. Data on grainage performance as influenced by repeated mating during September-October.

Tableau III. Données sur la performance de grainage sous l'influence d'un accouplement répété en septembre – octobre.

Treatment *Traitement*	Egg laying (no.) *Ponte d'œufs (nbre)*	Egg retention (no.) *Rétention d'œufs (nbre)*	Hatching (no.) *Eclosion (nbre)*
Fresh male × fresh female *Nouveau mâle × nouvelle femelle*	264	6	262
Single mated male × fresh female *Mâle accouplé une fois × nouvelle femelle*	252	12	247
Double mated male × fresh female *Mâle accouplé deux fois × nouvelle femelle.*	254	7	249
Triple mated male × fresh female *Mâle accouplé trois fois × nouvelle femelle*	248	16	244
Tetra mated × fresh female *Accouplé quatre fois × nouvelle femelle*	237	25	198.
Penta mated male × fresh female *Mâle accouplé cinq fois × nouvelle femelle*	136	125	0
C.D. at 5% / *D. C. à 5 %*	**7**	**5**	**6**

Hatching:

Number of hatched egg was significantly highest (231) from the egg laid by the female coupled with fresh male. The number of hatched egg recorded was 197 and 164 from the egg laid by the females repeatedly utilization of males for 2nd and 3rd times, respectively. Most striking observation was that no egg was hatched when the female mated with the male utilising for the fourth time.

During September-October:

During September-October- prior to main commercial rearing i.e. October-November one male moth could be exploited up to 6th time to mate with the fresh females.

Egg laying:

Like March-April, during September-October also the egg laying was found significantly highest (264) when fresh female were coupled with fresh male. Eggs laid by the females mated with the males utilising for 2nd, 3rd and 4th time had non-significant variation among them (248– 254). Eggs laid by the females mated with the male exploited for the 5th time was significantly lower (237). But repeated utilization of the male moth for the 6th time with fresh female reflected very low egg laying (136).

Egg retention:

Egg retention was 6 to 16 inside the female body coupled with up to 4th time utilization of single male moth. Very huge amount of egg retention (125.00) was recorded inside the female body when mated with a male utilised earlier for the five times.

Egg hatching:

Significantly highest number of hatched eggs was observed by the eggs laid by the female when coupled with fresh male (262) followed significantly by the eggs laid by the female mated with the males for 2nd to 4th time (244–249). Number of hatched eggs decreased significantly in the eggs laid by the females mated by the male repeatedly used for the 5th time (199). Surprisingly no egg was hatched for the females when coupled with the male utilised for sixth time.

DISCUSSION

Success of muga culture highly depends upon seed (egg). Lack of supply of quality seeds (high hatching) not only affects commercial rearing but also disappoints farmers who are trying to adopt the muga culture in the newly adopted area of extended muga culture in terai region of West Bengal. The two main commercial crop-rearing seasons are October-November and April-May. Success of grainage during March-April and September-October is the foremost task to ensure the supply of large quantity of quality seed during commercial rearing. Asynchronous moth emergence and less number of seed cocoon productions are the principal constraints regarding egg production. Repeated mating by exploiting the same male can help to overcome this problem. For this, the optimum hours of mating should be recorded. Therefore, in the first experiment, effect of different mating hours on egg laying and hatching have been recorded (Table I). Non-significant results obtained from mating hours reflects that up to 12 hours mating any duration from second hour is sufficient for successful fertilization and egg laying which confirms the observations of Samson and Barah (1989), according to whom 3-5 hours mating is sufficient. From this observation it can be suggested that minimum mating hours can be exploited and the decoupled male can be utilized for repeated mating. However, in practice mechanical injury may damage both the male and fertilized female during decoupling within 5 hours. Moreover, mating duration of 7 hours shows highest fecundity and hatching, though the differences with others are not significant. Therefore, mating duration of 7hours can be suggested as optimum for successful mating, egg laying and hatching.

In the next experiment, repeated mating has been conducted exploiting the males for a maximum of four times during March-April (Table II) and of 6 times during September-October (Table III) because it has been reported that female fertility in silkworm depends upon their male mates (Sidhu *et al.,* 1967). During both the seasons- though potential fecundity of the female was nearly same, repeated mating changed the egg laying and hatching. From repeated mating exploiting males up to 4 times during March-April shows that utilization of male up to 3 times have no harmful effect on fecundity as well as hatching. During September-October also, no significant variation has been observed between the egg laying of females mated with males utilized for the 2nd, 3rd and 4th time and after that decreases significantly and hatching also decreases from single mating to 5th time mating. However, no egg hatched from 6th time utilization. So, utilization of male up to 4th time during this season is effective in terms of egg laying and hatching.

Both the results have clear conformity with Benchamin *et al.* (1990), according to them in *Bombyx mori* the fecundity is comparable in the first 4 matings but reduces in 5th and 6th mating and with Ojha *et al.* (1996) trying the repeated mating in *Antheraea mylitta* where they have been found that females lay eggs even when mated with 4 × males. Repeated mating lead to inadequate discharge of spermatic fluid and result in reduced fecundity (Sidhu *et al.,* 1967). This is also reflected in terms of reduced laying and fertility percentage in the present study. Earlier studies also indicate that viability of eggs is not affected significantly up to four matings of male moths (Subramanyam, 1982).

Therefore, in muga culture, as synchronization of male and female is one of the principal constraints of egg production, the problem can be overcome through repeated exploitation of male moth up to four times prior to main commercial crop rearing season and up to three times before second commercial rearing by artificial coupling for seven hours which will ultimately supply higher number of quality eggs to the farmers.

ACKNOWLEDGMENT

The financial assistance provided by University Grants Commission, New Delhi in carrying out this research work is gratefully acknowledged

REFERENCES

BENCHAMIN K.V.; MAGADUM S.B. & SHIVASHANKAR N. (1990). Mating capacity of male moths of pure breeds and hybrids in silkworm *Bombyx mori* L. Indian J. Seric. 29(2) : 182-187.

KHANIKOR D.P. & DUTTA S.K. (1998). Low temperature preservation of seed cocoons for delaying moth emergence in muga silkworm (*Antheraea assama* Westwood) (Lepidoptera Saturniidae). Proc. of The 3rd International Conference on Wild Silkmoths, Bhubeneshwar, Orissa, November 11-14.

SAHU M.; MUKHERJEE P.K. & MONDAL T. (1998). Constraints during pre-seed crop rearing of *Antheraea assama* Ww. at seed rearers level in lower Assam. Proc. of The 3rd International Conference on Wild Silkmoths, Bhubeneshwar, Orissa, November 11-14.

SAMSON M.V. BARAH A. (1989). Suggestion for Better Muga Seed Production. Indian Silk. June 1989, 15-16.

SIDHU N.S.; SREENIVASAN R. & SHAMACHARY (1967). Fertility performance of female moths depends on their male moths. Indian J. Seric. 1(2) : 77-84.

SUBRAMANYAM K. (1982). Studies on the effect of rest on the efficiency of male moths in relation to number of mating and occurrence of viable eggs in silkworm *B. mori.* L. STS dissertation submitted ICTRETS, C.S.R & T.I, Mysore, 14-15.

OJHA N.G.; SHARAN S. K.; RAVIKUMAR G.; DUBEY O.P.; SINHA B.R.R.P. & SINHA S. S. (1996). Effect of refrigeration on mating efficiency of male moths of Tasar silkworm, *Antheraea mylitta* D. Indian J. Seric. 35(2) : 155-157.

Research Paper *Indian J. Seric., 2005, Vol. 44, No. 1, 107-112.*

STUDIES ON REARING PERFORMANCE OF MUGA SILKWORM (*ANTHERAEA ASSAMA*) IN TERAI REGION OF WEST BENGAL (INDIA) - A NEWLY EXPLORED AREA

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(Received: 3 June, 2004; Accepted: 28 June, 2005)

A preliminary investigation on muga silkworm (*Antheraea assama* Westwood) rearing as well as grainage was undertaken during nine different seasons (February-March, April-May, May-June, July-August, August-September, October-November, November-December, December-February, and January-March) during 2000-2003 to identify the suitable season for commercial and seed crop rearing for successful introduction of mugaculture in Terai region of West Bengal, India, a newly explored area. Rearing and grainage parameters were better during Oct. - Nov. (Effective rate of rearing: 49.20 %, Yield: 32.07 kg, Fecundity: 283, Hatchability: 98.87 %) followed by Apr. - May and minimum during July - Aug. (Effective Rate of Rearing: 10.60 %, Yield: 6.39 kg, Fecundity: 188, Hatchability : 6.24 %).

Key words: Effective rate of rearing, fecundity, muga silkworm, seed crop, yield.

INTRODUCTION

In the perspective of rural poverty and unemployment, low productive agriculture and industrial backwardness in Terai region of West Bengal, India, the sericulture in general and mugaculture in particular may appear to have special relevance for the economic uplift of rural people. Till date, mugaculture is mainly confined to the Brahmaputra valley of Assam and foothills of East Garo hills of Meghalaya mainly due to the unique climate and natural abundane of food plants in North Eastern India (Thangavelu *et al.*, 1988; Samson and Barah, 1989). The climatic condition of Cooch Behar district and a part of Jalpaiguri district of Terai region of West Bengal is to some extent similar to that of Assam which may favour muga culture in commercial scale in this region (Isa and Samson, 1987; Singha *et al.*, 1991; Ray, 2003). Hence Central Silk Board and State Sericulture Department, Government of West Bengal try to introduce mugaculture in the region. It is well established that regional variation in climate influences the rearing of mulberry silkworm, *Bombyx mori* (Venugopala Pillai and Jolly, 1985; Muslin, 1986; Rahman and Ahmed, 1988; Roy Chowdhuri *et al.*, 1992; Ray *et al.*, 2000) which may be true for muga silkworm, *Antheraea asama* also. Since investigation has not been done till date in the zone under consideration, the present study was undertaken to identify the commercial crop and seed crop rearing seasons for successful mugaculture in the region.

MATERIALS AND METHODS

Cooch Behar district of West Bengal is under Terai region and is situated in the north eastern part of West Bengal, adjacent to Goalpara district of Assam. It lies between 26° 57'40" to 26° 32'20" North latitude and 88° 47'40" to 89° 54'35" East longitude. The altitude of the district is 43 m above MSL.

The meteorological data are given in Table 1. Muga silkworm rearing (9 crops) was conducted during all the seasons under consideration, namely, February-March, April-May, May-June, July-August, August-September, October-November, November-December, December-February and January-March. Rearing parameters such as larval period, mature larval weight (male and female), single cocoon weight (male and female), single shell weight (male and female), shell ratio, effective rate of rearing in number, absolute silk content and yield / 10,000 brushed larvae were recorded during all the 9 crops. Grainage parameters like potential fecundity, realized fecundity and hatchability (%) were

also observed in all the seasons. The experimental rearings were conducted during 2000-2003 at Khagrabari Extension centre (Cooch Behar, West Bengal), Regional Muga Research Station (Boko, Asam), Central Silk Board, India as well as at Khagrabari State Sericulture Farm (Cooch Behar, West Bengal), Govt. of West Bengal and grainage and other studies were conducted at Muga Research Laboratory, A. B. N. Seal college, Cooch Behar. Data obtained from each rearing were recorded and analyzed statistically.

RESULTS

Seasonal influence on rearing performance: Seasonal influence on rearing performances as reflected by larval

Table 1: Mean meteorological data (2000-2003).

Season	Photoperiod (h)		Temperature (°C)		Humidity (%)		Rainfall
	Light	Dark	Max.	Min.	Max.	Min.	(mm)
March - April	12.19	11.41	25.57	19.72	72.20	58.02	57.53
April - May	12.45	11.15	29.77	23.52	73.35	62.82	164.43
May - June	13.09	10.51	31.77	25.97	77.57	69.30	506.94
June - July	13.20	10.40	32.30	27.17	82.87	75.16	725.44
July - Aug.	13.04	10.56	32.24	26.84	81.17	75.69	571.17
Aug. - Sept.	12.40	11.20	31.85	25.99	80.15	75.05	381.15
Sept. - Oct.	12.50	11.10	29.40	23.98	78.76	72.98	294.18
Oct. - Nov.	11.15	12.45	26.91	19.24	75.41	70.53	42.32
Nov. - Dec.	10.45	13.15	24.14	13.97	74.02	68.45	5.77
Dec. - Jan.	10.39	13.21	20.02	10.57	75.32	64.45	9.47
Jan. - Feb.	10.51	13.09	19.80	11.27	79.10	61.37	15.57
Feb. - March	11.30	12.30	24.07	15.17	75.73	57.80	37.23

(Source: State Meteorological Department, Cooch Behar and CSB Extension Centre, Cooch Behar.)

Table 2: Effect of seasons on rearing performances of muga silkworm.

Rearing parameter / Season	Larval duration (days)	Weight of single mature larva (g)		Single cocoon weight (g)		Single shell weight (g)		Shell %		ERR/ No.	Absolute silk content (kg)	Yield/ 10,000 larvae brushed (kg)
		♂	♀	♂	♀	♂	♀	♂	♀			
Feb.-March	25.00	7.02	11.00	4.75	7.02	0.330	0.430	7.06	6.12	31.20	1.19	18.35
April - May	24.00	6.96	11.30	4.49	7.77	0.350	0 .444	7.86	5.71	43.40	1.74	26.61
May - June	23.90	6.38	10.24	4.23	7.23	0.330	0.440	7.75	6.11	35.40	1.36	20.23
July - Aug.	23.60	6.47	10.70	4.38	7.66	0.324	0.436	7.40	5.69	10.60	0.40	6.39
Aug. - Sept.	24.40	7.05	11.17	5.01	7.72	0.354	0.450	7.08	5.99	27.80	1.12	17.70
Oct. - Nov.	27.20	7.61	11.97	5.19	7.84	0.338	0.496	6.45	6.32	49.20	1.95	32.07
Nov. - Dec.	35.60	7.24	11.51	4.62	7.27	0.340	0.444	7.35	6.10	39.60	1.55	23.54
Dec. - Feb.	58.80	7.10	10.11	3.98	5.95	0.306	0.408	7.69	6.85	24.20	0.87	12.02
Jan.- March	55.60	6.83	9.75	3.50	5.50	0.282	0.356	8.05	6.47	19.80	0.63	8.93
CD at 5 %	10.94	0.14	0.24	0.17	0.19	0.041	0.049	0.60	0.41	5.01	1.02	2.94

duration, single cocoon weight, single shell weight, shell ratio, absolute silk content, effective rate of rearing and yield are depicted in Table 2.

Table 3: Effect of seasons on grainage performances of muga silkworm.

Season	Potential fecundity (no.)	Realised fecundity (no.)	Laying (no.) up to 3rd day	Egg retention (no.)	Hatchability (%)
Feb.-Mar.	240.00	238.00	204.00	2.00	97.06
April - May	254.40	251.20	216.80	3.20	44.79
May - June	235.20	230.00	204.40	5.20	20.88
July - Aug.	235.40	188.00	162.80	47.40	6.24
Aug. - Sept.	241.80	230.20	179.80	11.60	63.03
Oct. - Nov.	285.60	283.80	222.80	1.80	98.87
Nov. - Dec.	250.00	245.00	200.40	5.00	97.37
Dec. - Feb.	200.80	193.00	151.80	7.80	97.80
Jan - March	203.80	200.00	146.80	3.40	98.20
CD at 5 %	11.48	12.13	3.18	4.29	2.52

Larval duration

The larval duration was the longest during Dec.-Feb. (58.80) days) followed by that of Jan.-Mar. (55.60 days), Nov.-Dec. (35.60 days) and Oct.-Nov. (27.20 days). The larval duration for the remaining months were observed between 23.60-27.20 days. No significant difference was observed between that of Feb.-Mar., Apr.-May, May-June and Aug.-Sept. (23.90-25.0 days). The shortest larval duration was observed during July-Aug. (23.60 days) having non-significant difference with that of Aug.-Sept. (24.40 days).

Larval weight

As male and female mature larval weight vary greatly in muga silkworm, the larval weight for male and female was recorded separately.

Weight of mature male larvae: Significantly high weight was observed during Oct.-Nov. (7.61 g) followed by Nov.-Dec. (7.24 g). No significant difference was observed between values recorded during Dec.-Feb. (7.10 g), Aug.-Sept. (7.05 g), Feb.-Mar. (7.02 g) and Apr.-May (6.96 g). Also, there was no significant variation between that of Apr.-May and Jan.-Mar. (6.83 g). The lowest weight was observed during May-June (6.38 g) having non-significant difference with that of July-Aug. (6.47 g).

Weight of mature female larvae: The highest mature female larval weight was recorded during Oct.-Nov. (11.97 g) followed by Nov.-Dec. (11.51 g). Non-significant difference was observed between that of Nov.-Dec. and Apr.-May (11.30 g). Mature female larval weight was higher during Feb.-Mar. (11.00 g). Significantly lowest female larval weight was observed during Jan.-Mar. (9.75 g).

Single cocoon weight

Cocoon weight also varied between male and female population in muga silkworm.

Single cocoon weight (male): Male cocoon weight varied from 3.5 to 5.19 g during different seasons having significant variations. The highest male cocoon weight was obtained during Oct.-Nov. (5.19 g) followed by Aug.-Sept. (5.01 g) and Feb.-Mar. (4.75 g). However, there was no significant difference between that of Feb.-Mar. and Nov.-Dec. (4.62 g). Significantly lowest male cocoon weight was recorded during Jan-Mar. (3.5 g).

Single cocoon weight (female): Female cocoon weight varied from 5.50 to 7.84 g. The highest female cocoon weight was recorded during Oct.-Nov. (7.84 g). During Jan.-Mar., a low female cocoon weight was recorded (5.50 g).

Shell weight

Male shell weight varied from 0.282 to 0.354 g having non significant variation during different seasons. Female shell weight varied from 0.356 to 0.496 g having non-significant difference during different seasons. The highest female shell weight was observed during Oct.-Nov. (0.496 g) followed by Aug.-Sept. (0.450 g). During Jan.-Mar., both male (0.282 g) and female (0.356 g) shell weights were recorded the lowest. Regarding average shell weight, non significant differences were observed during all the seasons (0.402 - 0.378 g) except during Dec.-Feb. when it was 0.357 g and during Jan.-Mar. when it was 0.319 g which was the lowest.

Shell %

Shell % was observed higher in male (6.45 - 8.05 %) than in female (5.68 - 6.85 %). The highest SR % was obtained during Jan.-Mar. (8.05 %) for male cocoon and during Dec.-Feb., for female cocoon (6.85 %).

Effective rate of rearing (ERR / No.)

The economic output of muga silkworm rearing as reflected by effective rate of rearing in number (ERR) showed significant variations among the seasons. ERR during Oct.-Nov. recorded the best (49.20 %) followed by that in Apr.-May (43.40 %). The ERR was also good during May-June (35.40 %). Lowest ERR was observed during July-Aug. (10.60 %) followed by Jan.-Mar. (19.80 %).

Absolute silk content

From the qualitative point of view, absolute silk content was calculated (per 10,000 larvae brushed) and it was observed the highest during Oct.-Nov. (1.95 kg) followed by Apr.-May (1.74 kg). Absolute silk content during Nov.-Dec. was also better (1.55 kg) having non-significant difference with that in Apr.-May. The silk content during May-June was also good (1.36 kg). However, the lowest performance was obtained during July-Aug. (0.40 kg) followed by winter months.

Yield

Cocoon yield was calculated per 10,000 larvae brushed. The highest cocoon yield was recorded during Oct.-Nov. (32.07 kg) followed by Apr.-May (26.61 kg). Yield during May-June (20.23 kg) and Nov.-Dec. (23.54 kg) was also good. The lowest yield was observed during July-Aug. (6.39 kg).

From the above results it was found that almost all the rearing parameters were observed better during Oct.-Nov. and the worst during July-Aug.

Seasonal influence on grainage performance: Seasonal influence on grainage performances as reflected by potential fecundity, realized fecundity and hatching percentage are depicted in Table 3.

Potential fecundity

Significant variation was observed among the seasons. Significantly highest potential fecundity was recorded during Oct.-Nov. (285.60) followed by Apr.-May (254.40) and Nov.-Dec. (250.00). It was found nearly the same during Mar.-Apr. and Aug.-Sept. (240.00 and 241.80, respectively) and also during May-June and July-Aug. (235.20 and 235.40, respectively). During Dec.-Feb., the entiality was the lowest (200.80) followed by Jan.-(203.80).

Realized fecundity

Realized fecundity was also observed significantly highest during Oct.-Nov. (283.80) followed by Apr.-May (251.20), Nov.-Dec. (245.00). The lowest realized fecundity was observed during July-Aug. (188.00) when the egg retention inside the female body was maximum (47.40). During Dec.-Feb. (193.00) and Jan.-Mar. (200.00), realized fecundity was also low. However during May-June and Aug.-Sept., the realized fecundity was at par (230. 00 and 230.20, respectively).

However, as per usual recommendation regarding effective collection of eggs up to three days to minimize the length of rearing and to synchronize the mating, data were recorded up to three days which reflected the same trend like realized fecundity *i.e.*, the best during Oct.-Nov. (222.80) followed by that in Apr.-May (216.80) and the lowest during Jan.-Mar. (146.80).

Hatchability

Hatching percentage was observed the highest during Oct.-Nov. (98.87 %) followed by Jan.-Mar. (98.20 %), Dec.-Feb. (97.80 %), Nov.-Dec. (97.37 %) and Mar.-Apr. (97.06 %). During May-June, the hatching percentage was very poor (20.88 %) and during July-Aug. almost no egg hatched (6.24 %).

Seasonal influence on grainage parameters was also observed better during Oct.-Nov. and the worst during July-Aug.

DISCUSSION

After careful observation it was found that the larval duration was observed longer during Dec.-Mar., which may be due to low temperature and shorter light period in winter seasons (Ochieng'-Odero, 1992; Sweeney and Vannote, 1981). During the rest of the seasons, the larval duration varied from 23.60 days to 27.20 days. Both male and female larval weights were higher during Oct. to Dec., may be due to longer larval period. Effective rate of rearing, the ultimate economic output to the farmers, was better during Oct.-Nov. followed by Apr.-May due to low disease occurrence. Regarding ERR and cocoon weight, Oct.-Nov. was the best followed by Apr.-May., Nov.-Dec. also showed better yield. Yield of May-June was also good.

From the meteorological data, it can be noticed that during Oct.-Nov., the temperature ranges from 26.91 to 19.24°C and humidity from 75.41 to 70.53 % and during Apr.-May it is 29.77 to 23.52°C and humidity from 73.35 to 62.82 % which are the optimum temperature and humidity ranges for muga culture (Thangavelu *et al.*, 1988). Fall of temperature and humidity after Oct.-Nov. and rise of temperature and humidity after Apr.-May affect effective rate of rearing as well as cocoon yield. Both ERR and yield were the highest during Oct.-Nov. in Assam also followed by May-June (Sahu *et al.*, 1998).

So, the ideal season for commercial rearing in Terai region of West Bengal is Oct.-Nov. and Apr.-May. The food plants may be utilized in Nov.-Dec. and May-June also if leaf and seed are available. Winter months from mid Dec. to mid Feb. should be avoided. The season, Feb.-Mar. may be utilized mainly as for seed production crop for supply during Apr.-May rearing. During rainy months (June to August), the rearing should be avoided. Only multiplication and maintenance of stock can be undertaken during rainy and winter months.

Grainage parameters (fecundity and hatchability) were also superior during Oct.-Nov. due to optimum temperature (19.24 to 26.91 °C), humidity (70.53 to 75.41 %) and photoperiod (11.15 L : 12.45 D) (Thangavelu *et al.*, 1988; Mahanta and Goswami, 1986). But during Apr.-May, though fecundity was high, hatchability was not good which may be due to longer light period as compared to hatchability during winter months having longer dark period.

However, for supply of seed during Apr.-May and Oct.-Nov., rearing seasons of Feb.-Mar. and Aug.-Sept. should be utilized. During Apr.-May rearing, sufficient number of seed can be supplied from Feb.-Mar. (231 hatched larvae/laying), but during main commercial crop rearing season, seeds from Aug.-Sept. rearing result in 140 hatched larvae from one dfl due to poor hatchability. This problem is also faced by Assam (Sahu *et al.*, 1998; Khanikor and Dutta, 1998) and for that reason, rearing status of Aug.-Sept. should be improved, or large scale rearing should be done during Aug.-Sept. for steady supply of seed for main commercial crop rearing.

From the results it can be suggested that for successful mugaculture in Terai region of West Bengal especially in Cooch Behar and Jalpaiguri districts, commercial crop should be reared during Oct.-Nov. and April-May and seed crop during Feb.-March and Aug.-Sept.

ACKNOWLEDGEMENT

The investigators are grateful to University Grants Commission, New Delhi for financial assistance through Major Research Project on Mugaculture, to Central Silk Board, Govt. of India for supply of seed and seed cocoon and to State Sericulture Farm, Govt. of West Bengal for providing muga food plantation for this experiment.

REFERENCES

Khanikor, D. P. and Dutta, S. K. (1998) Low temperature preservation of seed cocoons for delaying moth emergence in muga silkworm (*Antheraea assama* Westwood) (Lepidoptera: Saturniidae). *3rd International Conference on Wild Silkmoths*, 11-14 November, Bhubaneshwar, Orissa, India, p.73.

Mahanta, H. C. and Goswami, M. C. (1986) Some aspects of influence of photoperiod on muga silkworm, *Antheraea assama* Westwood (Lepidoptera: Saturniidae). *Uttar Pradesh Journal of Zoology*, **6(1)**: 98-107.

Isa, M. and Samson, M. V. (1987) Muga culture finds new fields in West Bengal. *Indian Silk*, **26(4)**: 47-48.

Muslin, M. M. (1986) Trials on multiple silkworm rearing in spring-post spring seasons in Pakistan. *Pak. J. For.*, **36(1)**: 5-8.

Ochieng'-Odero, J. P. R. (1992) The effect of three constant temperatures on larval critical weight, latent feeding period, larval maximum weight and fecundity of *Cnephasia jactatana* (Walker) (Lepidoptera: Tortricidae). *J. Insect Physiol.*, **38(2)**: 127-130.

Rahman, S. M. and Ahmed, S. U. (1988) Stability analysis for silk yield in some promising genotypes of silkworm *Bombyx mori* L. *International Congress on Tropical Sericulture Practices*, Bangalore, February, 18-28.

Ray, N. (2003) Muga steps in West Bengal. *Recent Advances in Animal Science Research*, Orion Press International, India, **2**: 284-288.

Ray, N.; Senapati, S. K. and Deb, D. C. (2000) Studies on the combined effect of mulberry varieties and seasons on performances of bivoltine silkworm, *Bombyx mori* in Terai zone of West Bengal. *Indian J. Enotomol.*, **62(1)**: 60-65.

Roy Chowdhuri, S.; Rajanna, L.; Subba Rao, G. and Mani, A. (1992) Impact of climatic factors on the rearing performance of bivoltine at Nilgiri hills, Tamilnadu. *National Conference on Mulberry Sericulture Research*, December, 10-11.

Sahu, M.; Mukherjee, P. K. and Mondal, T. (1998) Constraints during pre-seed crop rearing of *Antheraea assama* Ww. at seed rearers level in lower Assam. *3rd Internaional Conference on Wild Silkmonths,* 11-14 November, Bhubaneswar, Orissa, India, p.32.

Sahu, A. K.; Singha, B. B. and Das, P. K. (1998) Phenological studies in muga silkworm *Antheraea assama* Ww. (Lepidoptera: Saturnidae) in relation to its rearing and grainage behaviour. *3rd International Conference on Wild Silkmoths,* 11-14 November Bhubaneswar, Orissa, India, p.3.

Samson, M. V. and Barah, A. (1989) Suggestions for better muga seed production. *Indian Silk,* **28(2):** 15-16.

Singha, B. B.; Lepcha, S. T. and Pramanik, A. T. (1991) Muga finds its way to West Bengal. *Indian Silk,* **30(5):** 41-43.

Sweeney, B. W. and Vannote, R. L. (1981) *Ephemerella* mayflies of White Clay Creek: bioenergetic and ecological relationships among six coexisting species. *Ecology,* **62:** 1353-1369.

Thangavelu, K.; Chakraborty, A. K.; Bhagowati, A. K. and Md. Isa. (1988) *Hand book of muga culture,* Central Silk Board.

Venugopala Pillai, S. and Jolly, M. S. (1985) An evaluation of quality of mulberry varieties under hill conditions and crop results of *Bombyx mori. Indian J. Seric.,* **24:** 48-52.

J. Exp. Zool. India Vol. 10, No. 2, pp. 271-276, 2007 *ISSN 0972-0030*

STUDIES ON SHORT-TERM COLD PRESERVATION FOR SUPPLY OF SEED DURING MAIN COMMERCIAL CROP REARING OF MUGA SILKWORM

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(Accepted 30 May, 2006)

ABSTRACT – An attempt has been made to avoid rearing during mid June to mid August reflecting poor supply of seed during main commercial season (October-November) by refrigerating the cocoons from previous commercial crop (May-June) to supply sufficient seed for commercial rearing. From the results it can be said that 15 days cocoon preservation from proceedings commercial crop rearing can delay the moth emergence for 10 days, subsequent adult moth preservation for 5 days show almost no deleterious effect on fecundity and hatchability. The eggs thus obtained if preserved after 24 hrs. for 21days can delay the hatching for 24 days. This experiment provided a schedule of rearing for better supply of seed during the main commercial crop rearing during October-November by maintaining the schedule of preservation package collecting the cocoons from commercial crop of May-June avoiding the adverse seasons. Moreover, only 35 paise/dfl. additional expenditure is required to supply adequate amount of healthy seed to the farmers for main commercial crop rearing during October-November.

Key words- muga silkworm, cold preservation, seed.

INTRODUCTION

For the success of sericulture industry, proper supply of silkworm eggs (seed) is essential. The hatching period of eggs must coincide with the availability of suitable leaves as well as environmental conditions. This problem is more acute in muga culture as the environmental conditions during mid June to mid August is quite adverse for rearing and as a result main commercial crop rearing during October-November suffers badly due to poor supply of seed (Samson and Borah, 1989; Sahu, 2003). Therefore, the hatching of larva has to be controlled, accelerated or postponed by artificial treatment under the refrigerated condition. In the life cycle of silkworm refrigeration is usually restored to all the four steps of development viz. egg (Yokoyama, 1962; Dutta *et al*, 1972; Tazima, 1978), larva (Yokoyama, 1962; Tazima, 1978), cocoon/pupa (Yokoyama, 1962; Tanaka, 1964; Kovala, 1970) and moth (Tazima, 1978; Ayuzawa *et al*, 1972). In muga silk worm, cocoon refrigeration at 7±1°C can delay moth emergence upto 30-40 days (Khanikor and Dutta, 1998), preservation of adult moth at 10±1°C can delay the coupling upto 7 days (Biswas and Ray, 2003) and cold preservation of 36 hours eggs can delay the hatching up to 36 days (Singha *et al*, 1998). Keeping these findings i.e. refrigeration of cocoon, moth and egg can delay the egg hatching under consideration, an attempt has made to avoid rearing during mid June to mid August reflecting poor supply of seed during main commercial season (October-November) by refrigerating the cocoons from previous commercial crop (May-June) to supply sufficient seed for commercial rearing.

MATERIALS AND METHODS

Cocoons were collected from Extension Centre, Regional Muga Research Station, Central Silk Board, Cooch Behar from commercial crop of May-June. Cocoons were preserved at 10±1°C upto 21 days and emergence was recorded. Subsequently the moths were preserved at 10±1°C upto 7 days and their grainage activities were recorded. Eggs laid by the females were collected for low temperature preservation at 10±1°C after 12, 24, 36 and 48 hrs. of egg laying for 7, 15, 21 and 30 days. Simultaneously a batch of cocoons and eggs were allowed to emerge and hatch respectively in normal condition and treated as control. Larva thus obtained then reared for supply of seed during October-November. Larvae thus obtained then reared for supply of seed during October-Nov. The experiment was conducted at Muga Research Laboratory, A.B.N. Seal College, Cooch Behar during 2001-2005. Data thus obtained were analysed statistically.

RESULTS

On Cocoon Preservation :

Effect of short term cold preservation on cocoon was recorded as pupal period and emergence percentage. (Table-1)

Pupal Period

In natural condition (Control), pupal period was

The success of this experiment can able to provide a schedule of rearing for better supply of seed during the main commercial crop rearing during October-November.

Cocoon Collection	**Commercial Crop Rearing**	:	**May-June**
	Spinning	:	9th June
	Cocoon Preservation at 10±1°C	:	14th June
Preservation	Moth Emergence	:	6th July
	Moth Preservation at 10±1°C	:	6th July to 11th July
	Coupling and egg laying	:	12th July to 15th July
	Egg preservation at 10±1°C	:	13th July to 16th July
	Egg hatching	:	14th Aug. to 17th Aug.
	Seed crop rearing	:	14th Aug. to 10th Sept.
Seed Crop Rearing	Moth Emergence	:	29th September
	Coupling and egg laying	:	30th Sept. to 3rd Oct.
	Brushing for commercial rearing	:	8th Oct. to 11th Oct.
Main commercial rearing		:	**From 9th October**

Additional electrical expenditure calculated for 20,000 dfl seed for commercial rearing which can be affordable.

Additional expenditure : (as electricity charge)

Input			
Cocoon Preservation (2500 cocoon)	15 days	180.00/day x 15	2700.00
Adult Preservation (90% Emergence) (@ 2200)	5 days	540.00 / day x 5	2700.00
Egg Preservation (70% coupling & 210 fec.) @ 1000 dfl.	21 days	45.00 / day x 21	945.00
Total electricity charges (Rs. 3.00 per unit)	6345.00		
Output			
Seed crop rearing of 1000 dfl (having 70% hatching, 30% ERR and 230 realised fecundity)		20,000 dfl. Seed	
Additional expenditure (6345.00 . 20,000) or 32 paise @ 35 paise. / dfl.			

found 17.33 days for male and 18.33 days for female. No adult was emerged after 18 days. After short term preservation of seed cocoon at 10±1°C, the pupal period has been lengthened for both male and female. It was observed that longer the preservation days longer the pupal period upto 18th day. Longest pupal period was observed for 17 days preservation which was 9.33 days and 8.67 days more over control for male and female respectively. However, there observed no significant difference between 11 to 18 days in male and 13 to 18 days in female.

Emergence Percentage

Emergence percentage was found higher over control than preserved for more than 2 days to 8 days. However, 90 to 96% emergence was observed upto 15 days preservation and in control. A drastically decreased emergence was observed after 16 days (85%) when it was 30% in 17th days and only 10% in 18 days preservation and after that emergence was nil.

Table-1 : Effect of seed cocoon preservation at 10±1°C on pupal period and emergence percentage of muga silkworm.

Cocoon Preservation Days	Pupal period after preservation		Emergence %
	Male	Female	
1	18.33	18.67	92.00
2	19.00	19.67	92.00
3	19.67	21.00	96.00
4	21.00	22.00	96.00
5	22.00	22.33	96.00
6	23.67	23.67	93.00
7	24.33	24.67	93.00
8	24.33	24.67	93.00
9	24.33	25.00	90.00
10	24.67	25.67	90.00
11	25.33	25.67	90.00
12	25.33	25.67	90.00
13	25.67	26.33	90.00
14	26.33	26.67	90.00
15	26.33	26.67	90.00
16	26.67	27.00	85.00
17	26.67	27.00	30.00
18	26.00	26.33	10.00
Control (Fresh)	17.33	18.33	92.00
CD at 5%	**1.432**	**1.444**	**0.739**

Table 4 : Rearing performance during different seasons.

Seasons	ERR. No.	Pupal Period (days)	Realized Fecundity (no.)	Incubation Period (days)	Hatchability (%)
June-July	8.20	16.67	170.00	8.00	7.21
July-August	10.60	16.89	188.00	8.00	6.24
Aug.-Sept.	30.80	17.67	230.00	8.33	63.03
C. D at 5%	2.090	1.196	10.22	1.505	1.70

Taking emergence percentage as well as delayed pupal period under consideration, adult moths from 15 days preserved cocoon was selected for adult preservation.

On Adult Preservation

Effect of adult preservation was recorded as coupling efficacy, realized fecundity and hatchability. (Table-2)

Coupling efficacy : No significant variation was observed between the coupling efficacy of fresh coupling (control) (96.67%) and upto 3 days of adult preservation (90%-96.67%). Significant variations were observed in all other cases and from 6 days and above the coupling efficacy were observed very low.

Table-2 : Effect of adult preservation at 10±1°C on coupling efficacy, fecundity and hatchability of muga silkworm.

Adult Preservation Period	Coupling efficacy	Fecundity	Hatchability
1 day	96.67	242	88.66
2 days	96.78	240	89.40
3 days	90.00	225	87.16
4 days	80.00	220	80.18
5 days	70.00	210	78.30
6 days	36.67	200	65.28
7 days	23.33	189	63.18
Fresh x Fresh (Control)	96.67	245	88.79
CD at 5 %	**9.53**	**3.53**	**8.48**

Table 3 : Effect of seed preservation of different hours of laying at at 10±1°C on incubation period and hatching percentage of muga silkworm.

Hrs. after laying	Preservation period (days)	Incubation period (days)	Days delayed	Hatching %
12 hours	7	18.33	10.33	86.33
	15	27.00	19.00	85.00
	21	32.33	24.33	42.75
	30	–	–	–
24 hours	7	18.33	10.33	83.76
	15	27.67	19.67	73.30
	21	32.33	24.33	70.28
	30	–	–	–
36 hours	7	18.67	10.67	79.33
	15	28.33	20.33	70.67
	21	31.33	23.33	60.33
	30	–	–	–
48 hours	7	19.33	11.33	75.67
	15	24.33	16.33	68.33
	21	29.33	21.33	49.67
	30	–	–	–
C.D. at 5%	Hrs. after laying	**0.263**	**0.263**	**0.201**
	Preserve Period	**0.228**	**0.228**	**0.151**
	Hrs. after laying x	**0.457**	**0.457**	**0.604**
	preserve period			

Fecundity

As the preservation day increased fecundity decreased. Highest fecundity was observed in control (245) having nonsignificant difference with 1 day and 2 days preservation, lowest being from 7 days (189).

Hatchability

Hatchability also followed the similar trend. Significantly highest hatchability was obtained from control (88.79%) and upto 3 days of adult preservation (87.16% - 88.66%) followed by preservation of 4 and 5 days (78.30% - 80.18%) adult preservation. Significantly lowest hatchability was observed from 7 days preservation (63.18%).

From overall results, it can be said that eggs from 5 days preserved adult can be selected for preservation so far as delayed hatching is concerned.

On Egg Preservation

Eggs obtained from 5 days adult preservation was collected after 12 hrs., 24 hrs., 36 hrs. and 48 hrs. and preserved for 7, 15, 21 and 30 days. Incubation period, days delayed (control : 8 days) and hatching percentage were recorded. (Table-3)

Incubation period

Incubation period could not be recorded for 30 days preservation as no larva was hatched. Highest incubation periods was observed when 12hrs./24hrs. eggs were preserved for 21 days (32.33 days) followed by 36hrs. for 21 days (31.33 days). Lowest incubation period was observed when 12hrs./24hrs. eggs were preserved for 7 days (18.33days)

Days delayed

When 12hrs./24hrs. eggs were preserved for 21 days hatching delayed for 24.33 days followed by 36 hrs egg preserved for 21 days (23.33 days) and by 48 hrs egg preserved for 21 days (21.33). Lowest delay was observed from the eggs of 12hrs./24hrs when preserved for 7 days (10.33 days).

Hatching Percentage :

Hatching percentage showed significant difference. Highest hatching percentage was found from the eggs of 12hrs. and preserved for 7 days and lowest from 12hrs. preserved for 21 days. Eggs of 24 hrs. when preserved for 21 days showed 70.28% hatching while in the eggs of 36 hrs preserved to 21 days showed 60.33% hatching. However 48hrs. egg when preserved for 21 days showed only 49.67% hatching.

Muga Silkworm survivability & egg production during adverse months

Larva thus hatched were reared during August-September and ERR No., fecundity and hatchability were recorded, alongwith the performances (Table-4) during adverse months.

Effective Rate of Rearing

ERR was observed very poor during June-July and July-August. However, ERR increased during August-September (30.80%)

Realised fecundity

Fecundity was observed highest during August-September (230.00) followed by July-August (188.00) and June-July (170.00).

Hatchability

Hatching percentage during June-July (7.21%) and July-August (6.24) was very poor, nearly no larva could hatch. However, 63.03% larvae were hatching during August-September.

DISCUSSION

Successful muga culture needs sufficient supply of seed during commercial rearing. Pre-seed crop rearing season for main commercial crop rearing suffers very much due to adverse environmental conditions i.e. very high temperature and humidity alongwith heavy rainfall during mid June to mid August. Attempts should be made to skip these months by any means. In this investigation short term cold preservation at 10±1°C can formulate a package to overcome this stress investigating all the loop holes including the economic investment.

From the results it can be said that 15 days cocoon preservation from proceedings commercial crop rearing can delay the moth emergence for 10 days, subsequent adult moth preservation for 5 days show almost no deleterious effect on fecundity and hatchability. The eggs thus obtained if preserved after 24 hrs. for 21days can delay the hatching for 24 days. These results have conformity with the earlier findings of Khanikar and Dutta, 1998; Biswas & Ray, 2003 and Singh *et al*, 1998. Though they exploited only one stage for preservation and cold shock was maintained much lower than this experiment (10±1°C). The eggs thus brushed will be reared for supply of seed for commercial rearing. The grainage performance was also observed satisfactory.

So, by maintaining the schedule and preservation package collecting the cocoons from commercial crop of May-June the adverse seasons can be avoided as well as only 35 paise/dfl. additional expenditure/dfl is required to supply adequate amount of healthy seed to the farmers for main commercial crop rearing during October-November.

ACKNOWLEDGEMENT

Authors are grateful to the University Grants Commission, New Delhi for financial assistance through Major Research Project.

REFERENCES

Ayuzawa C, Sekido I, Yamakawa K, Sakuri U, Kuruva W, Yaginuma Y and Tokovo Y (1972) *Hand book on Silkworm Rearing*. Fuji Publishing Company, Toyo, Japan.

Biswas I and Ray N (2003) Exploitation of muga culture in terai region of West Bengal, a non-traditional area. *Proc. of 4th all India people's technology Congress*, February 23-24, Kolkata. pp. 86-90.

Dutta R K, Sengupta K and Biswas S (1972) Studies of preservation of multivoltine silk worm eggs at low temperatures. *Indian J. Seric.* 11, 20-27.

Khanikar D P and Dutta S K (1998) Low temperature preservation of seed cocoons for delaying moth emergence in muga silkworm. (*Antheraea assama* Westwood) (Lepidoptera: Saturniidae). *The 3rd International Conference on Wild Silk moths*. Bhubaneswar, Orissa. November 11-14.

Kovalev P A (1970) *Silkworm breeding stocks*. Central Silk Board. 112 (108-133)

Sahu A K (2003) Muga silkworm seed production : Constraints and strategies. *Indian Silk* 42(2), 14-17.

Samson M V and Barah A (1989) Suggestions for Better Muga Seed Production. *Indian Silk* 28(2), 15-16.

Singha B B, Sahu A K and Das P K (1998) Identification of low temperature registrant embryonic stages(s) in muga silkworm, *Antheraea assama* Ww. The 3rd International conference on Wild Silk moths, Bhubaneswar, Orissa, Nov. 11-14.

Tanaka Y (1964) *Sericology*. Central Silk Board, Bombay, India. pp 20-35.

Tazima (1978) *The silkworm: An important Laboratory Tool*. Published by Kodansha Ltd. Tokyo, Japan.

Yokayama (1962) *Synthasized sciences of sericulture*. Central Silk Board, Bombay, India. pp 210-223.

eggs longer preservation period have adverse effect on hatching and also needs lower temperature stress to get positive performance.

ACKNOWLEDGEMENT

Authors are grateful to University Grants Commission for their financial support through Research Project on mugaculture.

REFERENCES

Ayuzawa C, Sekido I, Yamakawa K, Sakuri U, Kuruva W, Yaginuma Y and Tokovo Y (1972) *Hand book on Silkworm Rearing*. Fuji Publishing Co. Tokyo.

Biram Saheb M N and Puttaswamy Gowda (1987) Silk worm seed technology, In: *Appropriate sericulture techniques*, Dr. M.S.Jolly (Ed.), Mysore, India.

Dutta R K, Sengupta K and Biswas S (1972) Studies on preservation of multivoltine silk worm eggs at low temperatures. *Indian J. Seric.* **11**,20-27

Furusawa T, Shimizu K and Yano T (1987) Polymol accumulation in Non-Diapause eggs of the silkworm, *Bombyx moriL. J. Sericult. Sc. Japan* **56**, 150-156.

Hiratsuka (1975) *Text book of Tropical Sericulture*. Japanese Overseas co-per. Volunters, Tokyo.

Jolly M S (1983) *Organization of Bivoltine Grainage for Tropics. Sericulture Project 3*.CSRTI, Mysore.

Khanikar D P and Dutta S K (1989) Low temperature preservation of seed cocoons for delaying moth emergence in muga silkworm (*Antheraea assama* Westwood) (Lepidoptera: Saturniidae).*The 3rd. Int. Conf. Wild Silk moths*, Bhubaneswar, Orissa..

Kovalev P A (1970) *Silkworm breeding stocks*. Central Silk Board, Bangalore, India.

Krishnaswamy S, Narasimhanna M , Suryanarayana S K and Kumara Raj S (1978) Manual on sericulture -2, Silkworm Rearing, F.A.O. Rome.

Narayanswamy T K and Govindan R (1987) Effect of refrigeration of eggs of pure Mysore of Silkworm *Bombyx mori* L, at blue stage. *Entomon* **12**, 105-107.

Pearse V, Pearse J, Buchsbaum M and Buchsbaum R (1987) In: *Living Invertebrates*. Blackwell Scientific Publication, Boston.

Sahu A K, Singha B B and Das K K(1997) Preservation of seed cocoon of muga silkworm, *Antheraea assama* Ww, at low temperature and high altitude. In: *Current Technology on non-Mulberry* Service, Ranchi.

Sahu A K, Singha B B and Das P K (1998) Identification of low temperature resistant embryonic stage(s) in muga silkworm, *Antheraea assama* Ww. *The 3rd. Int. Conf. Wild Silk moths*, Bhubaneswar, Orissa. November 11-14.

Sahu A K, Singha B B and Das P K(1998) Phenological studies in muga silkworm *Antheraea assama* Ww, Lepidoptera: Saturniidae) in relation to its rearing and grainage behaviour. *The 3rd. Int. Conf. Wild Silk moths*, Bhubaneswar, Orissa.

Sahu M, Mukherjee P K and Mondal T (1998) Constraints during pre-seed crop rearings of *Antheraea assama* Ww. at seed rearers level in lower Assam.

Tanaka Y (1964) *Sericology*. Central Silk Board. Bombay, India.

Tazima (1978) *The silk worm: An important Laboratory Tool*. Published by Kodansha Ltd. Tokyo, Japan.

Ullal S R and Narasimhanna M N (1978) *Hand book of Practical sericulture*. Central Silk Board, Bangalore, India.

Wang san-ming (1994) *Silkworm Egg Production*. Vol-III. Published by Food and agricultural Organization, United Nations.

Wei-hua Xu, Sato Y, Ikeda M and Yamashita O (1995) *J. biol. Chem.* **270**, 3804-3808

Yokayama (1962) *Synthasized sciences of sericulture*. Central Silk Board, Bombay, India.

Yuyin C (2000)Technical standards for commercial egg production of bivoltine silkworm varites used in China. *Sericologia* **40**, 185-201

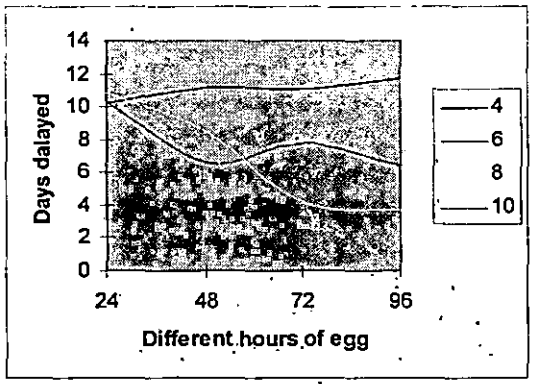
which was below 50%. Moreover 24 hours laying for 21 days at 4 °C and 6°C also performed poorly which was only 50%.

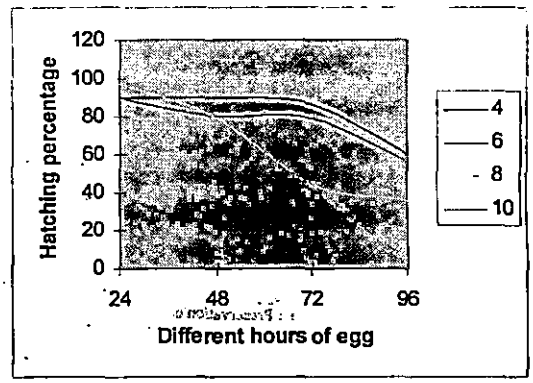
When eggs of the 96 hours were preserved for 15 days, no egg was hatched at 8°C and 10°C. At the same time at 4°C and 6°C when eggs were hatched, the hatching percentage nearly 20% only. The result was also same for 12 days preservation of 96 hours eggs at 8oC and of 72 hours laying both at 8°C and 10°C. Better performances (99.90 to 88.90%, having non-significant difference) were obtained from most of the other treatments excepting 12 and 15 days preservation of 48 hours laying at 10°C and 96 hours laying at any temperature for 7 and 12 days preservation and 3 days preservation at 8°C.

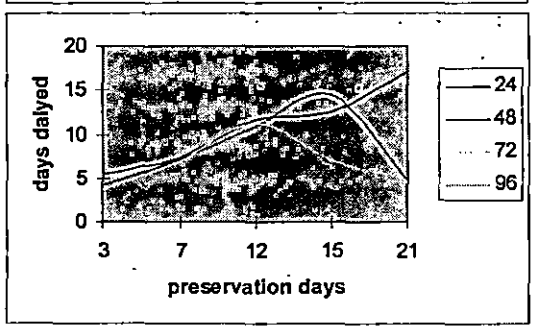
DISCUSSION

Muga culture needs sufficient supply of seed during commercial rearing which is far below the target (Sahu *et al,* 1998a,b). Moreover the hatching period of eggs must coincide with the favourable environmental condition of commercial rearing. In this regard the hatching of larvae has to be controlled which may be done by low temperature stress on eggs. In mulberry sericulture a lot of research work has been initiated in this aspect which shows that eggs of 16-24 hours should be utilized for cold stress at 5°C for a maximum period of 8 days (Biram Saheb *and* Gowda , 1996; Wang San-ming, 1994; Jolly,1983; Tazima,1978; Ayuzawa *et al,* 1972) . Results from the present investigation in muga silkworm (*Antheraea assama*) also shows similarity with the works of mulberry silkworm. Among the four age group considered (24hours, 48 hours, 72 hours and 96 hours) eggs of 24 hours perform best in respect to delayed hatching with higher hatching percentage. When compared with the controlled incubation period of 7 days all the preservation period from 3 days to 21 days show longer incubation period, highest being from 21 days. Eggs of 24 hours when preserved for 21 days hatch after 26.10 days which is 19.10 days more than normal condition and hatching percentage is also better (99.10%) than in normal condition (82 %). Among all the temperature stress condition (4,6,8 and10°C), 10°C is the optimum for 24 hours egg, 8°C for 48 hours, 6°C for 72 hours and 4°C for 96 hours. Therefore from the result it can be said that higher the age of eggs, lower the temperature stress needed for successful cold preservation (fig.1&2). Moreover, as the age of the eggs progresses preservation period become shorter ¾ a maximum of 15 days for 48 hours egg and a maximum of 12 days and 96 hours egg (fig.3&4). Result reflects that if 48 hours egg are necessary for preservation, 15 days preservation at 8°C shows 18 days delayed hatching with higher hatching percentage and at 6°C shows 15 days delayed hatching with higher hatching percentage. A delayed hatching with high hatching percentage of 2 weeks can be done by 72 hours eggs when preserved for 15 days at 6°C. Delayed hatching (9-13 days) with satisfactory hatching percentage can be obtained from 96hours egg at 4°C when preserved for maximum of 12 days.

So it can be concluded that so far as egg preservation is concern, low temperature stress on muga silkworm egg can be exploited to control hatching period as and when needed.24 hours old egg at 10°C preserved for 21 days can delay hatching for 19 days having no deleterious effect on hatching percentage. More over for older







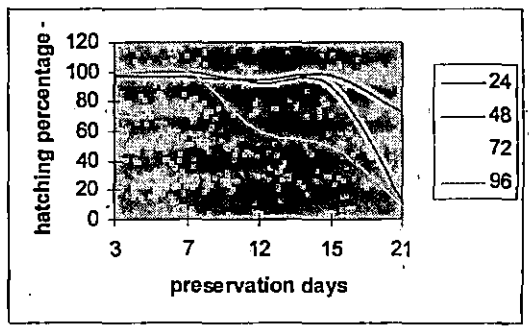


Fig. 1 : Effect of different age group of egg and preservation temperature on days delay of Hatching.
Figs. 2 : Effect of different age group of egg and preservation temperature on percentage of hatching.
Figs. 3 : Effect of different age group of egg and preservation period on days delay of Hatching.
Figs. 4 : Effect of different age group of egg and preservation period on percentage of hatching.

6°C and 96 hours 4°C showed better delay of egg hatching. Simultaneously delay was found higher with increased preservation days, highest being from 21 days, but hatching was only found for 24 hours laying at any temperature. In case of 48,72and 96 hours laying only at 4°C the eggs were hatched when preserved for 21 days. Highest delay for 19 days were obtained from 24 hours laying for 21 days preservation at 10°C followed non-significantly by 72 hours laying preserved for 21 days at 4°C (18.13 days), 48 hours and 96 hours laying for 21 days preservation at 4°C (18.06 days) and significantly by others. Days delay was minimum when eggs of 72 hours were preserved for 3 days at 10°C (3.03 day) having non-significant differences with layings of 48hours, 72 hours and 96 hours eggs for 3 days at any temperature (3.10-4.46 days).

Effect of laying, preservation period and preservation temperature on hatching percentage:

Hatching percentage, the ultimate reflection of cold preservation, was depicted in Table-3 and in the control batch hatching percentage is 82%.

Only the eggs of 24 hours laying for 21 days preservation at 8°C(98.93%)and 10°C(90.10%) showed better hatching percentage, but eggs of other hours were failed to show better hatching percentage

Table-1 : Effect of refrigeration at different temperature for different days on eggs of different days laying on incubation days.

°C	24 hours Egg				48 hours Egg				72 hours Egg				96 hours Egg			
D*	4	6	8	10	4	6	8	10	4	6	8	10	4	6	8	10
3	11.96	12.16	12.30	13.20	11.36	11.00	11.46	11.33	10.96	11.06	10.46	10.03	10.5	10.40	10.10	0.00
7	14.03	14.13	14.36	15.06	14.46	13.96	14.50	14.03	14.10	14.63	14.03	13.60	16.03	13.53	13.53	0.00
12	18.00	18.10	18.50	20.10	18.13	17.03	18.96	17.46	19.43	20.10	17.50	15.96	20.06	16.96	16.06	0.00
15	18.96	18.96	20.10	20.20	22.10	18.96	25.16	20.06	21.03	21.03	0.00	0.00	22.16	19.03	0.00	0.00
21	22.96	23.30	24.10	26.10	25.06	0.00	0.00	0.00	25.13	0.00	0.00	0.00	25.06	0.00	0.00	0.00

CD at 5% : Egg laying x Preservation Day= 0.028; Egg laying x Temperature=0.024; Preservation day x temperature=0.028; Egg laying x Preservation temperature x Preservation day=0.512

Table-2 : Effect of refrigeration at different temperature for different days on eggs of different days laying on days delay of hatching..

°C	First day laying				Second day laying				Third day laying				Fourth day laying			
D*	4	6	8	10	4	6	8	10	4	6	8	10	4	6	8	10
3	4.96	5.16	5.30	6.20	4.36	4.00	4.46	4.33	3.96	4.06	3.46	3.03	3.50	3.40	3.10	0.00
7	7.03	7.13	7.36	8.06	7.46	6.96	7.50	7.03	7.10	7.53	7.03	6.60	9.03	6.53	6.53	0.00
12	11.00	11.10	11.50	13.10	11.13	10.03	11.96	10.46	12.43	13.10	10.50	8.96	13.06	9.96	9.06	0.00
15	11.96	11.96	13.10	13.20	15.10	11.96	18.16	13.06	14.03	14.03	0.00	0.00	15.16	12.03	0.00	0.00
21	15.96	16.30	17.10	19.10	18.06	0.00	0.00	0.00	18.13	0.00	0.00	0.00	18.06	0.00	0.00	0.00

CD at 5% : Egg laying x Preservation Day= 0.118; Egg laying x Temperature=0.073; Preservation day x temperature=0.113; Egg laying x Preservation temperature x Preservation day=1.487

Table-3 : Effect of refrigeration at different temperature for different days on eggs of different days laying on percentage of hatching.

°C	First day laying				Second day laying				Third day laying				Fourth day laying			
D*	4	6	8	10	4	6	8	10	4	6	8	10	4	6	8	10
3	99.46	99.50	99.50	90.13	99.90	99.56	98.33	99.66	98.80	99.63	99.60	99.83	99.56	99.56	80.16	0.00
7	99.30	99.30	99.56	89.30	99.73	99.63	99.60	99.76	98.10	99.73	99.72	99.46	80.16	80.36	80.06	0.00
12	99.66	99.53	90.26	88.90	99.83	99.73	99.70	69.70	98.50	99.56	20.00	20.06	80.03	79.36	20.03	0.00
15	99.70	99.70	99.70	89.96	99.36	99.83	99.43	59.90	98.66	98.73	0.00	0.00	19.93	19.46	0.00	0.00
21	50.30	50.10	98.93	90.10	49.80	0.00	0.00	0.00	39.93	0.00	0.00	0.00	20.13	0.00	0.00	0.00

CD at 5% : Egg laying x Preservation Day= 0.132; Egg laying x Temperature=0.065; Preservation day x temperature=0.092; Egg laying x Preservation temperature x Preservation day=11.510

preservation temperature on delay of hatching:

Control batch showed incubation period for 7 days and after 7 days eggs were hatched. So the delay of egg hatching was calculated by deducting these 7 days from the total incubation period of different treatments and results were furnished in Table-2.

Delay of hatching followed the same trend of incubation period i.e. for 24 hours laying at 10°C, for 48 hours 8°C, for 72 hours

Therefore the hatching of larvae has to be controlled, accelerated or postponded by artificial treatment under the refrigerated condition. In the life cycle the silkworm refrigeration is usually restored to all the four steps of development viz. egg (Yokoyama, 1962; Dutta *et al,* 1972; Tazima 1978), larva (Yokoyama, 1962; Tazima 1978) cocoon / pupa (Yokoyama, 1962; Tanaka, 1964; Kovalev, 1970), moth (Tazima 1978; Ayuzawa *et al,* 1972; Krishnaswami *et al,* 1978; Jolly, 1983; Ullal and Narasimhanna, 1978). But the cold storage is to be restricted to any one of the developmental stages of the silkworm for avoiding deleterious effects (Jolly, 1983). Hiratsuka (1975) opined that the eggs after oviposition can be preserved for 20 days with out affecting the rearing characteristics. Moreover, an increase in the chilling duration beyond 20 days results in a decrease in the hatching percentage of non-hibernating eggs (Furusawa *et al,* 1897; Narayanaswamy and Govindan, 1987). According to Chen Yuyin (2000) for silkworm the safety time for cold inhibition is 3-5 days and at 5°C with an intermediate temperature of 10-13°C for 2-3 hours. Any information about induction of delayed hatching of muga silkworm (*Antheraea assama* Westwood) through refrigeration is not available except Sahu *et al* (1998) according to whom 36 hours eggs can be induced for delayed hatching up to 36 days. Keeping this in view under consideration an attempt has been made to delay the hatching of Muga silkworm eggs of different age by cold temperature preservation at different temperatures for different days with an ultimate objective to find out the optimum condition of temperature stress on muga silkworm eggs to coincide the hatching with suitable environmental condition for commercial rearing.

MATERIALS AND METHODS

Seed cocoons were collected from Extension Centre, Regional Muga Research Station, Central Silk Board, Coochbehar, West Bengal. After moth emergence, coupling was done and egg laying of female were collected for low temperature preservation at 4, 6,8,10°C in BOD incubating first, second, third and fourth day laying for 3,7,12,15,21 days; simultaneously, a batch of eggs were allowed to hatch in normal condition to measure delayed hatching due to low temperature preservation. All data analysed statistically.

RESULT

Effect of laying, preservation period and preservation temperature on incubation period :

Incubation period increased with the increased duration of preservation period, highest being from 21 days of preservation. Regarding cold temperature preservation, 10°C was found best for the 24 hours eggs, 8°C for 48 hours, 6°C for 72 hours and 4°C for 96 hours egg. Highest incubation period was found on 24 hours egg at 10°C for 21 days, which was 26.10days and lowest being from 72 hours eggs at 10°C when preserved for 3 days, which was 10.03 days. However no egg was hatched in different treatments which were at 10°C on 96 hours eggs for all the preservation days, on 72 hours eggs for 15 and 21 days preservation and 48 hours eggs for 21 days, at 8°C 72 hours and 96 hours eggs for 15 and 21 days preservation, 48 hours eggs for 21 days preservation and at 6°C 48 hours, 72 hours and 96 hours eggs for 21 days preservation. No harmful effect was observed for 4°C preservation.(Table1)

Effect of laying, preservation period and

J. Exp. Zool. India Vol. 9, No. 1, pp. 249-254, 2006 *ISSN 0972-0030*

EFFECT OF LOW TEMPERATURE PRESERVATION ON DELAYED HATCHING OF EGG OF MUGA SILKWORM, *ANTHERAEA ASSAMA* WESTWOOD

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(Accepted 25 May 2005)

ABSTRACT - Among most successful animal groups in terms of species, insects are predominant one and estimates of total number of living species ranges from 10 to 30 million. Many factors must contribute to the profusion of insect species, but one key element is probably the developmental plan. Particular set of environmental stimuli such as temperature, photoperiod, humidity and nutrients play important roles for success of insect development. Developmental plan of insects can be manipulated purposefully by altering temperature. Insect eggs are one of the suitable stages for the study of the effect of environmental stimuli on the development of insect embryo. Keeping this in view under consideration, an attempt has been made to delay the hatching of eggs of muga silkworm (*Antheraea assam* Westwood), by cold temperature preservation with an objective to find out the maximum delay of hatching so that eggs can be supplied to the farmers as and when required. In the present investigation eggs were collected after 24, 48, 72 and 96 hours after laying and preserved at 4, 6,8,10°C for 3,7,12,15 and 21 days. Results show that when eggs are collected after different hours after laying and are preserved for 12 days at 6±1°C shows 100% hatching. Eggs collected 24 hours after laying and preserved at 10±1°C for 21 days shows delay of hatching for 19 days with satisfactory hatching percentage. From overall result it can be said that it is possible to induce delay of hatching without affecting hatching percentage using low temperature preservation as stress.

Key words : *Antheraea assama,* preservation, temperature, hatching.

INTRODUCTION

Among most successful animal group in terms of species insects are predominant one and estimates of total number of living species ranges from 10 to 30 million (Pearse *et al,* 1987). Many factors must contribute to the profusion of insect species, but one key element is probably the developmental plan. Particular set of environmental stimuli such as temperature, photoperiod, humidity and nutrients play important roles for success of insect development (Wei-hua Xua *et al*, 1995). Developmental plan of insects can be manipulated purposefully by altering temperature. Insect eggs are one of the suitable stages for the study of the effect of environmental stimuli on the development of insect embryo.

For the success of sericulture industry, proper supply of silkworm eggs (seed) is essential. The hatching period of eggs must coincide with the availability of suitable leaves as well as environmental conditions.

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