BIONOMICS OF INDIAN OAK TASAR SILKMOTH, ANTHERAEA ROYLEI MOORE AND ITS POTENTIAL FOR BREEDING IN NORTH EAST INDIA

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ABSTRACT: During the survey of oak fed silkmoth fauna in the forest of North-Eastern part of India, the Indian oak fed silkmoth, *Antheraea roylei* Moore was recorded from oak growing areas of this region and it constitutes a part of the wild silk moth genetic resources of this region. It is distributed in the Indian Sub-Himalayan range and South East Asian countries. It feeds on about 12 varieties of oak tasar food plants. The bionomics and potential of the silkmoth was studied in the laboratory condition and the results are presented in this paper. *A. roylei* behaves as bivoltine in nature. The female moth lays 185-230 eggs. The average hatching percent, larval period, cocoon yield, effective rate of rearing (ERR %), cocoon weight, cocoon shell weight, cocoon shell ratio % and average single filament length in the laboratory condition were 61.40 %, 38 days, 30 cocoons/disease free laying (dfl), 20.36 %, 7.94 g, 0.85 g, 10.71 % and 250 m respectively. *A. roylei* shows high genetic compatibility with *A. pernyi* and *A. proylei* in interspecific hybridization which shows the genetic potential of this species. The F₁ hybrids of *A. roylei* with *A. pernyi* and *A. proylei* showed high vigour in all the yield contributing characters. The present study will be very helpful for future breeding programs in oak tasar culture.

KEY WORDS: Antheraea roylei, bionomics, characterization, genetic resource, conservation.

The oak tasar silk is produced in large scale by the countries like China, India and Japan. The commercially exploited oak tasar silkmoths are Antheraea pernui of China, Antheraea proylei of India and Antheraea yamamai of Japan. Many species reared for production oak tasar silk are univoltine or bivoltine in nature. The wild silk moths play an important role in the conservation and utilization of biodiversity (Frankel, 1982; Peigler, 1993). The conservation links genetic diversity to utilization, protecting diverse gene pool, habitat or ecosystem for human socio-economic needs (Metzler & Zebold, 1995). The ecorace conservation is must for utilizing their valuable genes in enhancing productivity and to build variation in new population through hybridization (Kumaresan et al., 2004; Mirhoseini et al., 2004). The intercross between A. pernui and A. yamamai was made but hybrids were sterile (Shimada & Kobayashi, 1992). The different interspecific crosses were also made in Antheraea but majority of them were sterile except A. roylei × A. pernui, A. mylitta × A. pernui and their reciprocals [Jolly et al., 1969; Jolly et al., 1973). The Indian Sub-Himalayan belt extending from Jammu & Kashmir in the North West and Manipur in the North East is the natural abode of many oak fed Antheraea species which feeds on nature grown oak plants available in this region. Out of the thirty five species of Antheraea recorded so far, thirty one species have been belonging to Indo-Australian biographic region (Seitz, 1933; Crotch, 1956). A few workers have studied some aspects of wild silkmoths in general and Antheraea species in particular in North east India during last some decades (Jolly et al., 1976; Thangavelu, 1991; Singh &

Singh, 1998; Baruah et al., 2000; Singh et al., 2000; Singh & Maheswari, 2003). Many important genetic resources of sericigenous insects may become extinct due to large scale deforestation, threat from various pests and predators, soil & air pollution by chemical insecticides and adverse climatic condition. Hence, proper conservation and utilization of the wild silk moths are the need of the hour to boost the silk production thereby helping in the conservation and utilization of biodiversity. The Indian oak fed wild silk moth, *Antheraea roylei* is one of such wild silkmoth found in the oak forest of North-Eastern India, which can be exploited for production of oak tasar silk. The successful performance of rearing and grainage of this wild silkmoth in the laboratory and its characteristic features reveal its genetic potential and need for conservation as a good genetic resource. Therefore, the present study was undertaken to know the bionomics of *A. roylei* and its genetic potential in Manipur, North East India and the results are presented in this paper showing the potential of the silkmoth for future breeding programmes.

MATERIALS AND METHODS

The survey and collection of Antheraea roylei were conducted in Manipur and its adjoining states in North East India and the seed cocoons were collected and consigned for conducting grainage in the laboratory of Regional Tasar Research Station, Imphal. The characteristics of the cocoons were recorded and compared with other races maintained in the station. The behavior of moth emergence, coupling and oviposition were recorded. The moths were allowed to couple for 10-12 hours inside wire mesh cages $(6' \times 6' \times 6.5')$ and after decoupling moths were kept individually for egg laying inside nylon net netted bags (15×20 cm) for 72 hr. The prepared disease free layings (dfl) were disinfected in 3% formalin for 10 minutes followed by washing in plain water prior to incubation. The dfls were incubated at 22+2°C and 70-80 % R.H. with slight modifications of the method in A. pernui [Jiang et al., 1992]. Eggs were spread in a thin layer prior to hatching and exposed to light in the morning hour. A few twigs of *Lithocarpus dealbata* were placed on the hatched larvae. The worms crawl over the leaves within half an hour and then the twigs were directly shifted in outdoor on *L. dealbata* foliages. The rearing field was well cleaned and disinfected before 10-15 days of rearing. The nylon bags containing eggs may be directly shifted on branches when the hatching starts so that the worms can crawl over the leaves. The rearing was conducted on raised plantations of L. dealbata following RBD with four replications successively for six generations in outdoor condition under nylon net cover in two seasons *i.e.*, April-May and July-August for three years in Imphal, India (785 m above MSL; 93.94°E longitude and 24.83°N latitude). While transferring of the worms, the branches with the worms are cut with the help of a secateure and shifted to another bush having quality foliages. Worms are not touched by hand as far as practicable. Transfer of worms is always carried out in the morning and evening hours only. Harvesting of the cocoons is done after 6-7 days of spinning. The newly hatched worms of the F_1 hybrids of the different crosses were reared cellularly and recorded the different rearing parameters. The grainage and rearing performance along with salient features of different life stages of the silk moth were recorded and analyzed. The conservation in the exsitu condition of the moth was continued for six generations to acclimatize in the semi-domesticated condition and to explore the possibility of future programs. This species was crossed with A. proulei and A. pernui and rearing performances were analyzed. The important yield contributing characters viz., fecundity,

hatching percentage, number of cocoons per dfl, effective rate of rearing (ERR %), average cocoon weight, single cocoon shell weight, cocoon shell ratio (SR %), single cocoon filament length were recorded.

RESULTS AND DISCUSSION

Regular field survey revealed that *Antheraea roylei* is distributed in the undisturbed oak growing forest of North-East India at the altitude of 780-2000 m AMSL. It was particularly more abundant in Senapati district (1500 m AMSL) of Manipur and West Kameng of Arunachal Pradesh while sparsely at other parts. The cocoons and larvae are found mostly on *Lithocarpus dealbata* and less on *Quercus serrata* and *Quercus grifithii* showing more preference on foliages of *L. dealbata*.

Voltinism: Voltinism is one of the important self regulating character in the life cycle of *Antheraea roylei*. *A. roylei* is purely bivoltine in nature and it undergoes diapause at the pupal stage after second crop. Normally first crop and second crop were reared during April-May and August-September seasons from the natural emergence. The natural emergence of moths starts from the third week of March during first crop and rearing is completed during April-May. During second crop, moth emergence starts from the second week of July and conducted grainage in the captive condition. The *A. roylei* moths do not undergo self coupling under captive condition in the early generations. However, it undergoes self coupling (60 %) in captive condition in the later generations. The second crop rearing is completed during August -September. The life cycle (egg to adult) of *A. roylei* under fluctuating environment varies from 70-80 days. The pupal diapause of this wild silk moth may be a physiological mechanism to withstand the cold climate of the winter.

Moth emergence and coupling: The emergence of moth occurs in the night from 18.00 to 22.00 hr. Self coupling of moths just after emergence is not allowed by keeping male and female moths separately in the cages for about 2-3 hr so as to allow the sperms and ova to mature. Coupling starts in the night time mostly from 19.00 hr and continued up to 23.00 hr. Coupling (45-60 %) takes place in the captive condition inside cages and the coupling of the remaining moths was induced mechanically. The optimum temperature and relative humidity for moth emergence and coupling are 22-24°C and 75-85 %. On reaching the female moth, the male moth starts courtship behavior with raised antennae, fluttering wings around the female followed by mating. Mating lasts for 10-12 hr but it continues up to 24 hr of the next day if not disturbed. Similar finding of mating on A. pernui was reported by some workers (Kuang-Ming & Ta-Yuan, 1958). Copulation by one male moth is enough for complete fertility of the female moth. Male moths are utilized for second time mating when there is shortage. In the natural condition the male moth flies long distances in search of females and the female moth also flies particularly after mating to lay the eggs on the leaves and branches of the food plants. However, they usually do not fly at day time. The moths do not lay all eggs at one place only but in a scattered way. The coupled moths detach at the slight mechanical disturbance. The life span of the adult moths is 7-10 days.

Oviposition: After decoupling, the individual female moths are kept for egg laying in nylon net bags (15×20 cm) in dark condition at $22\pm2^{\circ}$ C and 75 ± 5 % R.H. Eggs are collected after 72 hr of oviposition but egg laying continues up to 4-

5 days. The average fecundity ranges from 185-230 eggs in both the cropping seasons.

Incubation and Hatching: After proper disinfection the eggs are incubated at $22\pm 2^{\circ}$ C and 70-80 % R.H. Hatching takes place after 9-10 days of incubation. Profuse hatching does not occur initially and it continues up to late hours of the day. The average hatching varies from 60-75 %.

Larval Development: The larva spins the cocoon after about 35 days of active feeding through five larval instars but it is prolonged up to 40 days depending upon the climatic condition. The brushing and rearing of *A. roylei* need special care. The duration of development of first, second, third, and fourth instars took 6 days, 5 days, 7 days and 8 days respectively while the fifth instar took prolong time (12 days) where the temperature and relative humidity ranges from 23-31°C and 64-78 % R.H.

Laboratory studies have shown that the optimum favourable temperature and relative humidity for larval development is 28°C and 80 % for first and second instars, 27°C and 75 % for third instar and 25°C and 70 % for fourth and fifth instars respectively. The optimum environmental conditions are more or less similar to that of *A. proylei* (Singh & Singh, 1998).

Feeding behavior: The larvae prefer tender leaves to mature and hard leaves irrespective of the instars. Just after hatching the larvae crawl in search of food and are in the habit of eating bits of egg shell when hatched. It is observed that the worms crawl up to the tip of the branches and start eating tender leaves. It is desirable to provide tender leaves to the chawki worms (1st-3rd instars) while semi-mature and mature leaves should be provided to the 4th and 5th instars respectively for healthy growth of the larvae. The larvae feed on the entire leaves including midrib. It stops feeding at the slight disturbance. A larva consumes about 75-80 g of leave during the entire larval development.

Pupation and Adult development: The mature larva completes the pupation inside the cocoon within 5-7 days. The pupa develops into adult moth after about 20-35 days. After second crop the pupa undergoes diapause for 5-6 months. However, a few stray emergence of moths is also observed during late autumn.

Cocoon yield and survivability: The data of the *ex-situ* rearing in three consecutive years (2 crops/year) reveal that the average cocoon yield varies from 30-35 cocoons/dfl. The survivability is recorded as 25-30 % during both the cropping season. The low cocoon yield is mainly attributed due to the variations in climatic condition and quality of leaves fed to the silkworms. The low survivability may be due to non-acclimatization of the silkmoth under captive condition.

Morphological characteristics

Egg: Egg shell is brown and oval in shape. The size of the egg varies from 2.8-3.0 mm in length and 2.2-2.4 mm in breadth. The weight of the egg ranges from 0.8-0.9 mg.

Larva: The newly hatched larva is dark green in colour. The size varies from 5-6 mm in length and 1.0-1.5 mm in breadth and it is 5-6 mg in weight. There is colour polymorphism in the late instar larvae. The larval colour varies from light green to dark green (Fig. 1 a & b). The weight of mature larvae varies from 12.40-

16.64 g in male and 15.86-25.75 g in female. There are 462 numbers of elongated tubercular setae are present in the mature larva arranged in irregular fashion. The mature larva forms cocoon on the tree itself by making a hammock. The peduncle supports the cocoon in hanging from the branch.

Cocoon: Cocoons are oval in shape, length and breadth varies from 4.4-5.4 cm and 3.2-4.3 cm respectively, white in colour, double layered of which the outer layer is unreelable and the inner layer is reelable (Fig. 2 a & b). The cocoon weight ranges from 4.0-5.9 g in male and 6.1-9.9 g in female. The shell weight ranges from 0.5-0.7 g in male and that of female ranged from 0.7-1.0 g. The shell ratio ranges from 12-13 % in male while in female it ranges from 10-11%. The length of the peduncle ranges from 3.2-5.4 cm and it has a ring at the terminal for hanging on the twigs. The raw silk content varies from 06-09 % and the filament length varies from 180-204 m. The denier ranges from 5.7-6.3 and the reelability varies from 16-25 %. The boil-off loss is 17-20 %.

Pupa: The pupa is light metallic in colour. The length of the pupa varies from 2.8-3.6 cm in male and 3.4-4.2 cm in female. The breadth varies from 1.5-1.9 cm in male and 1.7-2.8 cm in female. The pupal weight varies from 3.5-5.3 g in male and 7.0-9.3 g in female respectively.

Moth: There are variations in the colour of both male and female moths. Males are greenish gray while the females are reddish brown in colour (Fig. 3 a & b). The body length of the moth varies from 2.9-3.6 cm. The wing span varies from 11.5-13.4 cm in male and 11.8-14.5 cm in female. The eye spot in both forewings and hind wings of both the sexes is very distinct and is round to oval with a large transparent round to elliptical fenestra. The eye spot of the hind wing is smaller than that of the forewing. The ground colour of the moth is grayish brown. The colour of ante median line, oblique line and post-median line are reddish, black with yellow and reddish white respectively.

Genetic potential: A. roylei can be utilized in interspecific hybridization for heterosis breeding and creation of genetic variability in oak tasar silkworms. The interspecific hybrids and reciprocal crosses of A. roylei (n=31) with A. pernui (n=49) and A. proylei (n=49) showed high hetero-beltiosis (improvement of the F_1 hybrid over better parent in percentage) in the yield contributing characters in spite of wide differences in chromosome numbers (Table 1). The rearing performance of the F₁ hybrids of A. roulei with A. pernui and A. proulei revealed that the fecundity, cocoon shell weight and cocoon shell ratio (%) were highest in A. pernyi \times A. proylei whereas the hatching (%) and cocoon weight were recorded highest in A. roylei × A. proylei. However, cocoon per dfl and ERR (%) was found maximum in A. roylei $\times A$. pernyi. The resulting fertile and vigorous F₁ offspring may be due to the high degree of homology between the chromosomal complements of these three species (Singh et al., 1984; Nagaraju & Jolly, 1986). The studies carried out in the silkworm have shown that the characters could be pooled to suit the breeder's choice since selection for one trait has a correlation with genetic changes for other traits (Gamo, 1976; Tazima, 1984).

Biodiversity conservation is increasingly recognized as a fundamental component of sustainable development of natural resources by protecting and using biological resources in the ways that do not diminish the world's variety of genes and species or destroy important habitat or ecosystem. *Antheraea roylei* constitutes a significant component of wild silk moth genetic diversity that cannot be ignored in the assessment of quantitative and qualitative characters, conservation and utilization of silk moth biodiversity. The present study reveals that *A. roylei* can be utilized in heterosis breeding and for enhancing genetic variability in oak tasar culture.

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Name of the spp. / breeds	Fecundity (no.)	Η%	Cocoon Per dfl (no.)	ERR (%)	Cocoon Weight (g)	Shell Weight (g)	SR %	Filament Length (m)
A. roylei	198	61.4	30	20.36	7.94	0.85	10.71	250
A. pernyi	133	69.92	45	48.38	5.94	0.61	10.27	626
A. proylei	121	70.24	42	49.41	5.91	0.69	11.67	628
A. roylei × A. pernyi	174 (30.83)	81.03 (15.89)	91 (102.22)	64.53 (33.38)	7.94 (33.67)	0.84 (37.70)	10.57 (2.92)	810 (29.39)
A. pernyi × A. roylei	142 (6.76)	74.65 (6.76)	79 (43.04)	74.53 (54.05)	7.84 (31.98)	0.99 (62.29)	12.92 (25.80)	925 (47.76)
A. roylei × A. proylei	187 (54.54)	75.94 (8.12)	74 (76.19)	52.12 (5.48)	8.80 (48.90)	1.08 (65.00)	12.27 (51.41)	990 (57.64)
A. proylei × A. roylei	132 (9.09)	66.66 (-5.09)	64 (52.38)	72.72 (47.17)	8.61 (45.68)	0.91 (31.88)	10.69 (-8.40)	905 (44.10)

Table 1. Average rearing performance of *Antheraea roylei* with other oak tasar silkworm species and their hybrids.

*Figure within parentheses indicates hetero-beltiosis in percentage of the F1 hybrid



(a) (b) Figure 1 (a & b). Colour variations of *Antheraea roylei* larvae feeding on *Lithocarpus dealbata*.



Figure 2. Antheraea roylei cocoon (a) garlands for grainage, (b) male & female cocoon.



Figure 3 (a & b). Moths of Antheraea roylei (Male and Female).