Ecologia

ISSN 1996-4021

Academic
Journals Inc.

www.academicjournals.com

Ecologia 2 (2): 43-51, 2012
ISSN 1996-4021 / DOI: 10.3923/ecologia.2012.43.51

Evaluation of *Antheraea mylitta* Cocoons Preservation for Synchronize Seed Production Through Eco-tasar-friendly Technique

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ABSTRACT

Tropical tasar silkworm, *Antheraea mylitta* produces tasar silk having vast demand in national and international market due to unique quality. During the long period of diapause and quiescence, the cocoons face extremes of temperature (10-46°C) and humidity (30-80% RH) which cause pupal mortality, decreased vigour of moths during grainage and unseasonal emergence. All these losses during the long adverse period of diapause and quiescence require to be reduced to the minimum, so that maximum utilization of seed cocoons is ensured and optimum production of disease free layings is achieved. Cocoon loss in diapausing stock sometimes attains serious proportion. As normal practice, seed cocoons of *A. mylitta* are used to preserve in different conditions during period of diapause to minimize the loss. In the present study, evaluations of *A. mylitta* cocoons preservation in concrete grainage house and agro shade net grainage house have been done for synchronize seed production through eco-tasar-friendly technique. Comparative grainage behavior of tasar silkworm in these two modes of cocoons preservation revealed highly significant differences in all grainage parameters. Higher synchronized moth emergence, more coupling percentage, less span of emergence peak was found in cocoons preserved in agro shade net grainage, in comparison to concrete grainage house where higher pupal mortality, abnormal moths and less coupling percentage were observed. Our result clearly indicates that, preservation of seed cocoon under agro shade net grainage house is suitable for seed production.

Key words: *Antheraea mylitta,* cocoon preservation, grainage, eco-tasar-friendly, seed cocoons

INTRODUCTION

The Indian tropical tasar silkworm, *Antheraea mylitta* Drury (Lepidoptera: Saturniidae) used to rear twice or thrice in a year (June-August, September-October and in November-January) depending on location and period of rearing. The trivoltine pupae of *A. mylitta* remains in diapause condition from January to May whereas, bivoltine stocks remain in diapause from November to mid-June. After the commercial crop (diapausing) rearing, the pupae undergo diapause for several months to avoid unfavorable condition of winter. It is reported that long period of diapause and exposure to varied climatic conditions results in loss of substantial percent of seed cocoons due to pupal mortality and erratic moth emergence (Kapila *et al*., 1992).

In tropical insects, environmental regulators, such as day length, temperature and rainfall regulate their diapause (Denlinger, 1986) and low temperature as experienced in winter, increases diapause incidence (Hackett and Gatehouse, 1982; Kikukawa and Chippandale, 1983). Similarly, higher temperature blocks initiation of development (Hackett and Gatehouse, 1982) and lack of rainfall, however, may have an indirect effect by altering composition of the food source (Denlinger, 1986). Onset of rains is directly linked to diapause termination and synchronized adult insects availability (Wolda and Denlinger, 1984; Denlinger, 1986).

Impact of temperature on growth, development and reproduction of insect has been studied by several researchers in different insects (Malik, 2001; Mahar *et al.*, 2005; Pandey *et al.*, 2008; Ahmad *et al.*, 2008) and its relation with endocrine causes of early embryonic development has been also investigated (Bajaj and Sharma, 2011). Developmental response of cabbage butterfly, *Pieris brassicae* L. on different cole crops under laboratory and field condition has been also evaluated (Ali and Rizvi, 2007). Good impact of nitrogen treated mulberry trees on the larval, development and cocoon weight of *Bombyx mori* was found (Mahmood *et al.*, 2002). Impact of inter-specific biodiversity of 51 peanut cocoon strains of Iran silkworm germplasm based on reproductive traits was found (Nezhad *et al.*, 2010). Recently, Reddy (2011) identified the impact of trait selection in optimizing the egg and silk yields of *A. mylitta* Drury for deed and commercial crop seasons. Preceding literature suggest that evaluations of *A. mylitta* cocoons preservation for synchronize seed production through eco-tasar-friendly technique have not been studied much and its correlation with temperature is not fully established.

As a traditional practice, seed cocoon preservation and grainage used to perform inside the mud-walled grainage house, concrete (concrete) grainage house, country tiles and arbutus roof grainage house to offer better protection to the seed cocoons, particularly against low humidity and high temperature. Percentage of normal emergence has been reported to range in between 55 to 78% in different modes of indoor preservation with longer span of emergence (Kapila *et al.*, 1992). Singh *et al.* (2004) reported seed cocoons preserved in outdoor under wire net at Ranchi (24.21° North, 659 meters AMSL) is comparatively better than indoor preserved seed cocoons. It is felt that, to enhance production and productivity of tasar silk, successful grainage management is necessary. For successful grainage, synchronized moth emergence, good coupling percentage, less span of grainage peak, less pupal mortality and normal moth's emergence are important parameters. But studies on these parameters in correlation with behavioral response of *A. mylitta* moth emergence under two different cocoon preservation conditions i.e., concrete grainage house and agro shade net grainage house have not been reported till date. Hence, in the present study, the behavior of moth emergence of *A. mylitta* cocoons, preserved in concrete grainage house and agro shade net grainage house has been evaluated to see impact of preservation conditions and its suitability for synchronize seed production through eco-tasar-friendly technique.

MATERIALS AND METHODS

Project duration: Present study was conducted during the financial year 2010-2011 at Pilot Project Centre, Bengabad, Jharkhand India and Central Tasar Research and Training Institute (Central Silk Board) Ranchi, India.

Insect material: The larvae of tropical tasar silkworm, *A. mylitta* Drury (Bivoltine Daba ecorace) were reared during commercial crop (diapausing generation) in outdoor conditions (Kumar *et al.*, 2011). After spinning stage, cocoons were collected from field and preserved in concrete grainage house and agro shade net grainage house located at Pilot Project Centre, Bengabad, Jharkhand India. These cocoons were allowed to get ready for adult emergence and utilized for present study.

Experiment design: The seed cocoon preservation experiments were designed using Complete Randomized Design (CRD). The healthy diapausing cocoons of stocks which were free from uzi and Ichneumon infestations, were consigned to the two modes of preservation since 1st of January after completion of pupation. The seed cocoons of stocks were used in the experiments and were preserved under concrete (Pakka) grainage house and agro shade net grainage house. Soon after harvest from the second crop, 5 replications of 50000 cocoons were preserved in respective grainage house. Total 500 cocoons from each replication of both the preservation conditions were randomly selected to study grainage parameters. The *A. mylitta* moth emergence behavior under these two preservation conditions has been evaluated to see storage condition impact and suitability.

Grainage parameters: Data was recorded for male and female insect pupal mortality, abnormal moth, erratic emergence, regular emergence period (the period during when the leaves of host plants are ready for consumption by the silkworms), span of emergence peak (the period during which 70% population emerges in concentrated form), coupling percentage, disease percentage (pebrine) and the number of dfls (Disease free laying) obtained.

Temperature and relative humidity: Pattern of temperature and relative humidity during cocoon preservation and grainage period of inside the agro-shade net grainage house and concrete grainage house have been recorded.

Data analysis: The data was subjected to the statistical analysis to see the impact of different modes of preservation on aforesaid grainage parameters by using Student's' t' test. Microsoft Excel 2003 software was used to analyze the data.

RESULTS

The *A. mylitta* moth emergence behavior under two preservation conditions i.e., agro shade net grainage and concrete grainage house and its correlation with temperature, humidity and grainage parameters has been evaluated to see cocoon preservation conditions impact and its suitability. Data revealed that, comparative grainage behavior of tasar silkworm in two mode of cocoons preservation showed highly significant differences in all grainage parameters of tasar silkworm. Higher synchronized moth emergence, more coupling percentage, less span of emergence peak was found in cocoons preserved under agro shade net grainage, in comparison to concrete grainage house. The higher pupal mortality, abnormal moths and less coupling percent were observed in seed cocoon preserved concrete grainage house leading to affect the low seed production. Prolonged moth emergence period 15th June 2011 to 1st August 2011 was recorded in the concrete grainage house with major peak period (above 70%) 13th July to 27th July 2011 whereas, in agro shade grainage house, major peak period (80%) was observed on 23rd June to 5th July 2011 and more synchronized with host plant leaf availability (Fig. 1).

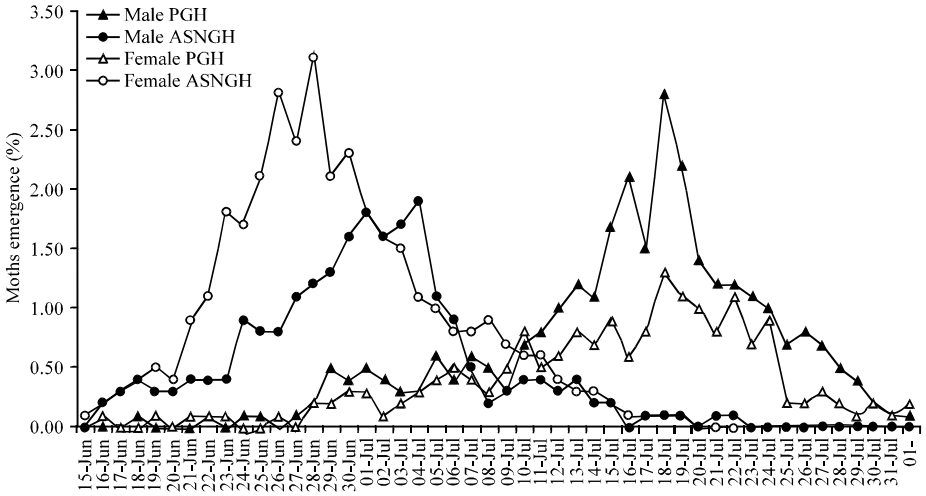

Fig. 1: Comparative evaluation of moth emergence and behavioral pattern of *A. mylitta* cocoons stored in concrete grainage house and agro shade net grainage. ASNGH: Agro shade net grainage house, PGH: Pakka grainage house

Table 1: Comparative evaluation based on grainage parameters of *A. mylitta* seed cocoons stored in concrete (Pakka) grainage house and agro shade net grainage house

Grainage parameters	Mode of preservation of seed cocoons	Mean±SE	Variance	t-value
Male moths (%)	PGH*	30.00±1.581	12.500	-6.045**
	ASNGH**	35.20±1.158	6.700	
Female moths (%)	PGH	18.40±0.980	4.800	-3.641**
	ASNGH	22.80±1.200	7.200	
Total moths emergence (%)	PGH	48.40±0.812	3.300	-6.857**
	ASNGH	58.00±1.897	18.000	
Pupal mortality (%)	PGH	51.60±0.812	3.300	6.857**
	ASNGH	42.00±1.897	18.000	
Abnormal moths (%)	PGH	41.39±1.768	15.626	10.466**
	ASNGH	20.49±1.856	17.215	
Coupling (%)	PGH	53.31±1.764	15.557	-15.382**
	ASNGH	82.39±2.192	24.024	
Peak moth emergence span (days)	PGH	20.60±0.927	4.300	9.731**
	ASNGH	14.000±0.707	2.500	
Total moth emergence span (days)	PGH	39.400±1.030	5.300	13.054**
	ASNGH	23.200±0.860	3.700	

*PGH: Pakka (Concrete) house grainage, **ASNGH: Agro shade net grainage house

Comparative grainage behavior of tasar silkworm in two mode of cocoons preservation is presented Table 1. Data reveals that the highly significant differences ($p<0.01$) were observed in all grainage parameters of tasar silkworm cocoons kept in concrete grainage house and agro shade net grainage. In Agro shade net grainage, higher synchronized moth emergence, more coupling percentage (82.39±2.19%), less span of peak (14.00±0.71 days) and regular emergence (23.20±0.86 days) were recorded in comparison to concrete grainage house. The higher pupal mortality (51.60±0.812% and abnormal moths (41.39±1.76%) were observed in concrete

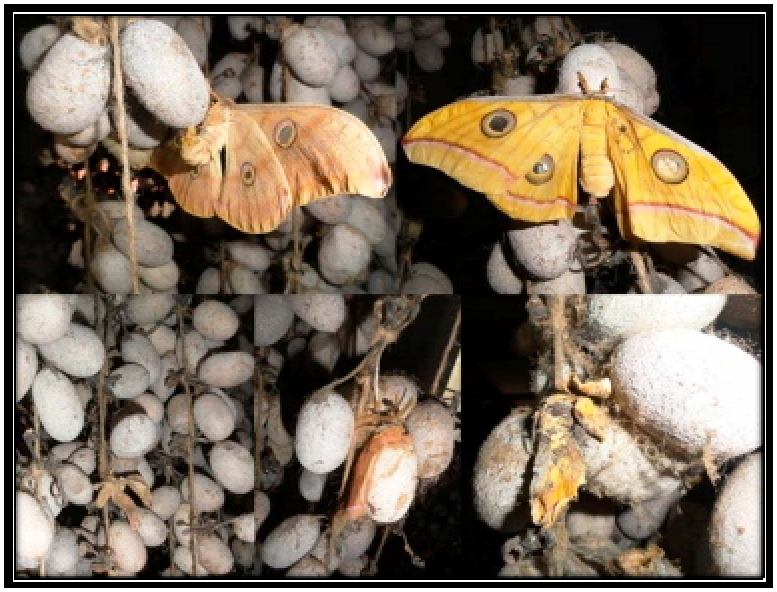

Fig. 2: Showing abnormal moths emergence i.e., no egg inside abdomen, crippled winged, half emerged moths in the concrete grainage house

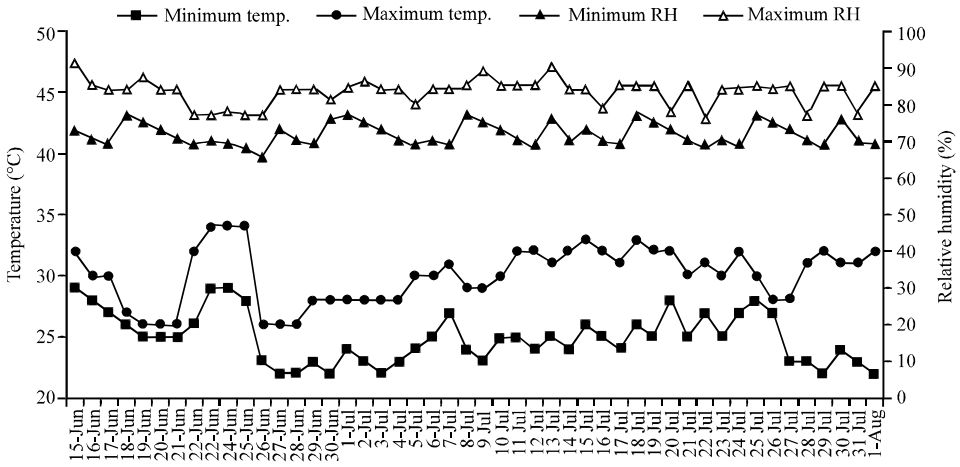

Fig. 3: Pattern of temperature and relative humidity during grainage period inside the agro shade net grainage house

grainage house. In concrete grainage house coupling percent was adversely affected due to abnormal moths and prolonged emergence span leading to low seed production in seed (Fig. 2). Variation in maximum and minimum temperature (°C) and relative humidity (%) was recorded in both the grainage houses (Fig. 3-4). The near to optimum temperature and RH was observed in the agro shade net grainage house in comparison to concrete grainage house i.e., higher temperature and lower relative humidity.

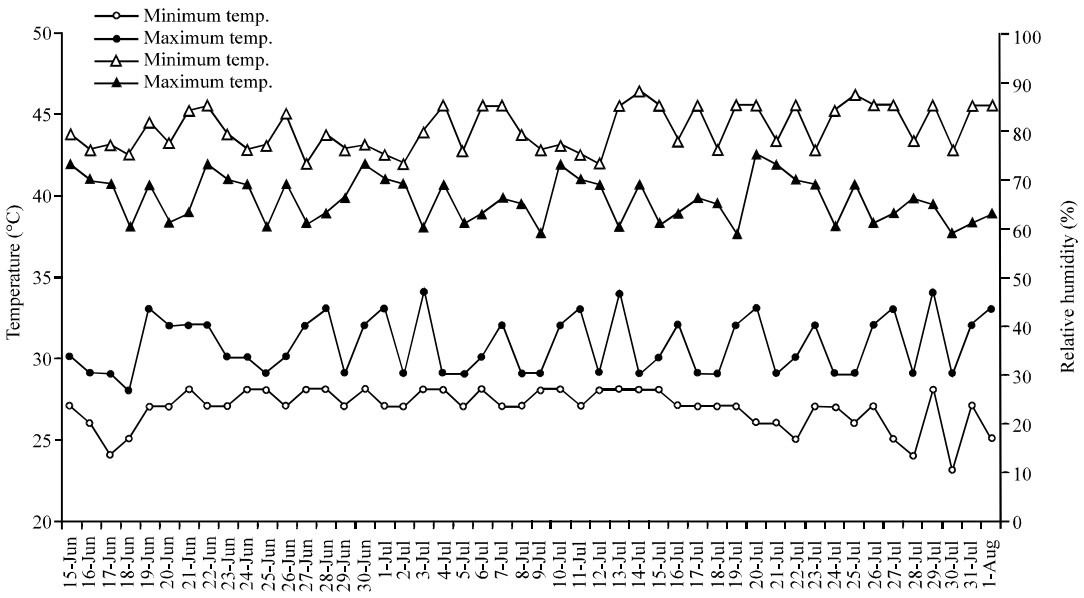

Fig. 4: Pattern of temperature and relative humidity during grainage period inside the concrete grainage house

DISCUSSION

In present study, comparative grainage behavior of tasar silkworm in two mode of cocoons preservation revealed highly significant differences in all grainage parameters of tasar silkworm cocoon preserved in concrete grainage house and agro shade net grainage. In agro shade net grainage, higher synchronized moth emergence, more coupling percentage, less span of peak in comparison to concrete grainage house. The higher pupal mortality and abnormal moths were observed in concrete grainage house. The coupling percent was adversely affected by the abnormal moths and prolonged emergence span in concrete grainage house leading to low seed production. We propose that differences in grainage parameters of both the preservation condition is due to temperature, relative humidity and photoperiod. Since, outdoor preservation in agro shade net is near to nature therefore, insect grainage performance is comparatively better than concrete grainage house preserved seed cocoons. Our finding corroborates the finding of Singh *et al.* (2004) revealed that cocoons preserved under outdoor condition emergence period was significantly reduced and emerged male and female moths were more synchronized for coupling. Peak period of emergence recorded in outdoor preserved stock was of 7 days, whereas it was 15 days in concrete grainage house and 19 days in mud-wall grainage house. Impact of temperature on growth, development and reproduction of insect has been studied by several researchers in different insects (Malik, 2001; Mahar *et al.*, 2005; Pandey *et al.*, 2008; Ahmad *et al.*, 2008) and its relation with endocrine causes of early embryonic development has also been investigated (Bajaj and Sharma, 2011). Developmental response of cabbage butterfly, *Pieris brassicae* on different cole crops under laboratory and field condition has also been evaluated (Ali and Rizvi, 2007).

Based on our finding it is evident that agro shade net grainage house is comparatively suitable for the optimum temperature and relative humidity which is essential for tasar cocoons preservation and synchronized moth emergence in comparison to concrete grainage house i.e., higher temperature and lower relative humidity. These findings elaborated by the Singh *et al.* (2004) who

reported that variation in total effect of light intensity, temperature and humidity consequent upon mode of preservation has influenced the survival, emergence duration, synchronization between sexes and pebrine percent. In tropical insects, environmental regulators, such as day length, temperature and rain-fall regulate their diapause (Denlinger, 1986) and low temperature as experienced in winter, increases diapause incidence (Hackett and Gatehouse, 1982; Kikukawa and Chippandale, 1983). Similarly, higher temperature blocks initiation of development (Hackett and Gatehouse, 1982) and lack of rainfall, however, may have an indirect effect by altering composition of the food source (Denlinger, 1986). Onset of rains is directly linked to diapause termination and synchronized adult insects availability (Wolda and Denlinger, 1984; Denlinger, 1986). The impact of temperature on immune responses of insect has also been investigated (Pandey *et al.*, 2010). Recently, Pandey *et al.* (2012) evaluated the seed cocoons preservation conditions based on hemocyte and enzyme based method. Good impact of nitrogen treated mulberry trees on the larval, development and cocoon weight of *Bombyx mori* was found (Mahmood *et al.*, 2002). Impact of inter-specific biodiversity of 51 peanut cocoon strains of Iran silkworm germplasm based on reproductive traits was found (Nezhad *et al.*, 2010). Recently, Reddy (2011) identified the impact of trait selection in optimizing the egg and silk yields of *A. mylitta* Drury for deed and commercial crop seasons.

In the existing practice, seed cocoon preservation and grainage are done inside mud walled grainage house to offer better protection to the cocoons, particularly against low humidity and high temperature. Percentage of normal emergence has been reported to range in between 55 to 78% in different modes of indoor preservation with longer span of emergence (Kapila *et al.*, 1992). The prolonged emergence in indoor condition leads to early production of eggs when suitable leaves and environmental conditions are not available for feeding of silkworm and prolonged requirement of labour. Ecoraces of tropical tasar silkworm, like Modal and Nalia which are collected from nature, shows very little pebrine incidence. According to Nayak *et al.* (2001) this may be because in nature, temperature and relative humidity is harmoniously balanced by the very habitat itself without needing any human endeavour. Such trials to correlate the major physiological effects of climate to thermal balance and micro climatic interaction with temperature etc. have been made in other insects also (Willmer, 1982).

Since, Daba ecorace of *A. mylitta* is semi-domesticated but its nature is still wild, therefore, outdoor preservation in agro shade net grainage house is preferable for insect due to it is near to nature in comparison to cocoon preserved concrete grainage house. The present study may lead to change of seed cocoon preservation method incurring low cost, lower disease incidence and increased seed production during more appropriate time.

CONCLUSION

It is concluded that, tasar silkworm, *A. mylitta* Daba BV Ecoraces diapausing seed cocoon preservation under agro shade net grainage house is better than concrete grainage house. Higher synchronized moth emergence, more coupling percentage, less span of emergence peak was found in cocoons preserved under agro shade net grainage, in comparison to concrete grainage house where higher pupal mortality, abnormal moths and less coupling per cent were observed leading to low seed production. Agro shade net grainage house found suitable for the optimum temperature and relative humidity. The present study may lead to change of seed cocoon preservation method incurring low cost, lower disease incidence and increased seed production during more appropriate time to enhance production and productivity of tasar silk. Evaluations of *A. mylitta* seed cocoon

preservation results clearly indicates that for synchronize seed production through eco-tasar-friendly technique, seed cocoons should be preserved in Agro shade net grainage house as a substitute of Concrete grainage house.

ACKNOWLEDGMENTS

Authors are thankful to Sri Pitamber Jha, Pilot Project Officer, PPC, Bengabad, Giridih, Jharkhand for providing necessary facilities to conduct experiment. Financial support of CSB is also acknowledged.

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