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MICROSTRUCTURE AND LIFE CYCLE OF METISA *PLANA* WALKER (LEPIDOPTERA: PSYCHIDAE)

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Abstract: *The Metisa plana (Lepidoptera: Psychidae), is an important defoliator of oil palm in Southeast Asia. Severe defoliation on palm consequently affects the productivity. The rearing protocol, microstructure, behaviour and life cycle of all stages of M. plana are described. The life cycle of M. plana from egg to adult in this controlled rearing environment was 103.5 days. The larvae of M. plana in this study undergo six larval instars. The larva started feeding immediately upon being laid on the leaf surface. In the larval period, the 1st instar took 9 – 16 days to change into 2nd instar. As the larva developed, it used lichen to build a case that it wore as it fed and grew. The development duration from 2nd instar to 3rd instar was 14 – 17 days. The 3rd instar took the longest period at 16 – 18 days to develop into 4th instar. Development of subsequent instar stages 4th to 5th and 5th to 6th were slightly shorter, between 10 – 15 days and 12 – 16 days respectively. In total, development of 1st through 6th instar stage of M. plana was 71.5 days.*

KEYWORDS: *Metisa plana, oil palm, microstructure, life cycle*

Introduction

Oil-palm pests have been controlled mainly by the use of insecticides, with their attendant environmental hazards (Morin and Mariau, 1971). The use of an Integrated Pest-Management (IPM) strategy holds promise for the sustainable management of the oil-palm pests. The components of this strategy are phytosanitary surveillance, cultural, biological control and the selective use of insecticides. The IPM approach requires a good reference of the biology of the pest.

In view of its importance, one of the very initial steps towards effective bagworm control would be to establish and understand the fundamental information on biology of the bagworm, such as behaviour, structure and life cycle. Knowledge on the biology and full life cycle of *M. plana* is essential for sound management of this pest. Understanding the life cycle of this insect would elucidate the appropriate times of spraying, an important factor for effective treatment (Sidhu and Saikia, 1999). In different stages of *Spodoptera frugiperda,* instars produced different levels of feeding and damage to cotton (Luttrell and Mink, 1999). The number of instars increased in adverse conditions when larvae fail to reach a specific threshold size for metamorphosis (Esperk, *et al*., 2007).

Bagworm outbreaks of unprecedented severity occurred in Malaysian oil-palm plantations from the late 1950s to early 1960s. Until today, the outbreak of bagworm is still continuous even at the same historical area. Complete life cycle of *Metisa plana* ranges from 80 to 113 days (Khoo *et al*., 1991) with a mean of 90 days (Basri and Kevan, 1995). Initial information on *M. plana* biology was reported by Yunus and Balasubramaniam (1975), but detailed information on the behaviour, microstructure and life cycle of *M. plana* are still lacking. Therefore,

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this study was undertaken to establish the rearing protocol, microstructure, behaviour and life cycle of *M. plana* for strategic control and management of the pest.

Materials and Methods

Source of Insects and Eggs Sterilisation Process

Cultures of *M. plana* were established by collecting cocoons from several oil-palm estates having *M. plana* outbreaks in Kepala Batas, Penang. The outbreak of *M. plana* was fairly synchronised, thus the emergence of the first instar larvae were reasonably uniform.

The cocoons were collected from the field and reared to adults inside a plastic container. Eggs laid by the mated females were collected. The black-headed eggs were surface sterilised in 1% formaldehyde solution for 15 minutes and rinsed under slow-running tap water for 10 minutes. A hundred surface-sterilised eggs were picked randomly for hatchability study and replicated three times. Non surface-sterilised eggs were used as a control. The eggs were placed inside a 20ml vial.

Preparation for Scanning Electron Microscopy (SEM)

The internal organs of larvae of different instars were observed under the scanning electron microscope (JOEL 6310). The 1st to 6th instars larvae, eggs and pupae were fixed in FAA for 24 hours, washed in 1% cacodylate buffer and post-fixed in 1% cacodylate-buffered osmium tetroxide solution for two hours. The samples were dehydrated in graded series of alcohol (30, 50, 70, 90, 95 and 100%), and Critical Point Drier Balzers using carbon dioxide liquid as an intermediate fluid. Critical-point dried samples were mounted on cylinder stubs and sputter-coated with gold, and then observed with a scanning electron microscope JEOL 6310 at an acceleration voltage of 10 or 15kV.

Results and Discussion

The Microstructure and Behaviour of *Metisa plana*

(a) Larvae of *Metisa plana*

Early Development of the Larvae

The larvae were lichen, forming a portable case, which they carried around with them as they fed. The rearing study showed that the average number of neonates (new born larvae) hatched from a single egg mass ranging from 140 – 210 neonates. The difference between mean number of eggs laid and emerged neonates could possibly be due to cannibalism by sibling neonates or be unfertile eggs (Rhainds and Ho, 2002). Newly-hatched neonate was approximately 1 mm and moved actively out through the case of female parent. Some of the weaker neonates died and failed to develop into 1st instar larvae. *Metisa plana* larvae were polyphagous. Within 1 – 2 hours of emergence, the neonates used plant material to construct a small case around the posterior region of their body. They were most often seen attached to the adaxial of oil-palm leaf in an upright position. In field condition, more often the larvae were hung on long silken threads for dispersal by wind or carriage by animals and men. The case was correspondingly enlarged and developed, either by widening at the frontal aperture, or by cutting the side walls and patching over the incision with a piece of cut leaf. According to Basri and Kevan (1995), larvae use 66.8% of the leaf tissue removed from the oil palm for growth and development and 33.2% for case construction. The case was cone-shaped at the 1st instar stage of the neonate (early stage instars), with closed distal end and open base for the neonate to feed and discharge waste. The study showed that 60 – 90% of the neonates survived and developed successfully into 2nd instar larvae. Hence, a mean between 60 – 190 larvae successful developed to 2nd instar larvae per egg mass.

The instars of the larvae can be identified by the morphological architecture and length of the case, which corresponds to the length of the caterpillar. The larvae carried the case as it moved around to feed. The case surface of instar 1 was relatively smooth with an average length of 1.6 mm.

In instar 2, two-to-three tiny, rounded leaf pieces were loosely attached at the basal end of the case. The average length of the case was 4.6 mm, which had enlarged more than 100% from the 1st instar case. The average length of the case at instar 3 was 5.9 mm. The surface of distal half of the case had a smooth surface, but the surface of the case toward the posterior end was disarranged, with four to six rectangular leaf pieces attached at the proximal half of the case. At instar 4, many loosely-attached large round-to-rectangular leaf pieces were on the case surface. The average length of the case was 9.5 mm. At instar 5, most of the loose leaf pieces were plastered onto the case forming a relatively smooth surface case, except for a few semi-rounded leaf pieces attached at the proximal base. The average case size was 11.3 mm. At instar 6, the case surface was smooth and no loose leaf piece were attached, and they were whitish-grey in colour. The case expanded in length and circumference and became cylindrical with an average length of 13.0 mm. As pupation commenced, the proximal opening of the case was sealed, and remained attached to the abaxial surface. The growth of development of *M. plana* from neonate to instar 6 took a mean of 71.5 days, and the length of the case and larva increased by 800% from 1.6 mm to 13.0 mm and 1.1 mm to 8.5 mm respectively (Table 1).

Morphological Development of Larvae

In Metisa plana at the early instar stage, the abdominal region was enclosed in the case while head and thoracic legs protruded from the proximal opening of the case to allow the larva to feed and move over the plant (Plate 1). The larva was moderately sclerotised and brownish-coloured with dark patches all over the head and thorax (Plate 2). The entire body was covered with sensory setae (Plate 3). Stemmata was well-differentiated, convex, approximately 50 µm diameter and located laterally on either side of the head capsule (Plate 4). The mouthpart of the larva consisted of the labrum, mandibles and maxillae. The mandibles bit off plant material and tore it into small, easily-digestible pieces. Inside the mouth was a pair of accessory jaws maxillas, sensory organ and a maxillary palp was attached to them for tasting the food (Plate 5). The labium was a pair of plates, fused together to form the lower lip of the mouth. The labrum functioned as the upper lip of the insect mouth.

Table 1: Mean length of 10 cases of respective instar stage with corresponding length of larva.

Instar	Mean Length of Case (mm)	Mean Length of Caterpillar (mm)
1st	1.6 ± 0.2	1.1 ± 0.1
2nd	4.6 ± 0.3	2.0 ± 0.1
3rd	5.9 ± 0.4	3.3 ± 0.2
4th	9.5 ± 0.4	5.7 ± 0.4
5th	11.3 ± 0.7	7.1 ± 0.1
6th	13.3 ± 0.7	8.5 ± 0.3

Note: 10 larvae per instar.

The thorax of *M. plana* larva was easily distinguished as compared to caterpillars such as *Sibine megasomoides*, *Spodoptera frugiperda*, etc. The thorax was immediately behind the head and possessed three pairs of true legs on the underside (Plate 6). The thoracic legs were jointed with hooks for mobility of the larvae. The wings in adults were formed directly above these legs.

The abdomen was the largest segment of the *M. plana* larva, segmented and contained the major part of the digestive system. The larva had three pairs of thoracic legs and five pairs of prolegs, which performed different respective functions. Four pairs of prologs were located along both sides of the ventral part of the abdomen (Plate 7) with the fifth pair near the anus (Plate 9). The prolegs were short, stout with circular terminal pad. The internal wall of the pad was lined by a uniserial layer of hook or crochet, about 200 µm long (Plate 8). The prolegs helped the larva to grip the leaf surface, even vertical or upside down surfaces. These prolegs disappeared in the adult. Along the side of the larva abdomen were nine pairs of small openings, spiracles and a respiratory organ (Plate 10).

(b) Pupae of *Metisa plana*

At pupation, the larva attached the case to the abaxial surface via a thin holder and closed the proximal open case. The pupa reoriented itself within the case with the head pointing down to distal end of the case. In contrast, the pupa of *Brachygyna incae* (Psychidae) did not invert

its position prior to pupation, with the adult consequently emerging from a sub-apical opening in the case (Davis, 2001). The average of full-grown pupa was about 6.1 mm long which was 2 times smaller than the case at the inception of pupation at average of 13.6 mm. There was a difference in appearance of the male (Plate 11) and female (Plate 12) pupa. The male pupa had a big blackish head and was approximately 4 mm in length, whiles the female pupa had a smaller creamy yellowish-coloured head, about half the size of the male pupa, and approximately 2 mm in length. During pupation, sexual segregation of pupation sites had been recorded in *Oiketicus kirbyi* (Psychidae). Female pupate away from the host where they fed or in higher locations (Stephens, 1962) and on the youngest leaves (Mexzon *et al*., 2003). Selection of a site for pupation depends on the female size (Rhainds *et al*., 1995).

(c) Adults of *Metisa plana*

Sexual dimorphism of the developed adults *M. plana* are very distinct. The adult female resembled a maggot, wingless and legless, with a small dark-brown head and yellowish-brown body, located at the anterior end of the case (Plate 13). The average female adult size was 5.5 mm in length and 2.0 mm diameter.

In contrast, adult male wriggled out of the distal end of the case and emerged from pupal case as a moth. The male moth was about 10 – 12 mm long with clear wings and a smoky-brown colour, with a hairy black body (Plate 14). Head was chubby with feathery bipectinate antennae. The adult males flew and sought for a female to mate. The males cut the distal end (lower end) of the female's case and mate with the female by inserting two-third of it extended abdomen into the female case. The females laid a large clutch of eggs in the case and left the case and died a few hours later. The fertilised eggs were left to develop to the 1st instar neonates in the case. The male adult lived for about 3 – 4 days.

In *Oiketicus kirbyi* (Psychidae), the ratio of bagworm females to males ranged from 10:1 to 2:1 (Rhainds *et al*., 1995). The probability of mating was limited by the low number of males. Possibly, a similar situation may occur in *M. plana* as many empty cases without eggs were found in the rearing cage. The females possibly failed to mate when they left the cases.

(d) Eggs of *Metisa plana*

Freshly-laid eggs were yellowish-coloured in clusters of 200 – 300 eggs (Plate 15), partly enclosed in a sheath. The egg was oblong-shaped,

Plate 1: *Metisa plana* head and thoracic legs protruded from the anterior opening of the case, allowing the larva to feed and move over the plant.

Plate 2: Larva, side view, brownish colour with dark patches all over the head and thorax.

Plate 3: 2nd instar larva, dorsal view, setae (white arrows) were all over the larva's body. Stemmata (white arrows) located laterally on either side of the head capsule (x 70).

Plate 4: Larval stemmata (white arrow), side view, disc-shaped and approximately 50 µm in diameter (x 300).

Plate 5: Larval head, ventral view. Mouthparts (m); maxillary palp (p) (x 90).

Plate 6: Larva head and thorax, ventral view, three pairs of true legs on the thorax (white arrows) (x 60).

Plate 7: 2nd instar larva, ventral view, five pairs of stumpy prolegs on the abdomen (white arrow) (x 27).

Plate 8: Larval crochet (blue arrow), side view. The crochet are small hook-like structures arranged in a circle and approximately 200 µm in length (x 370).

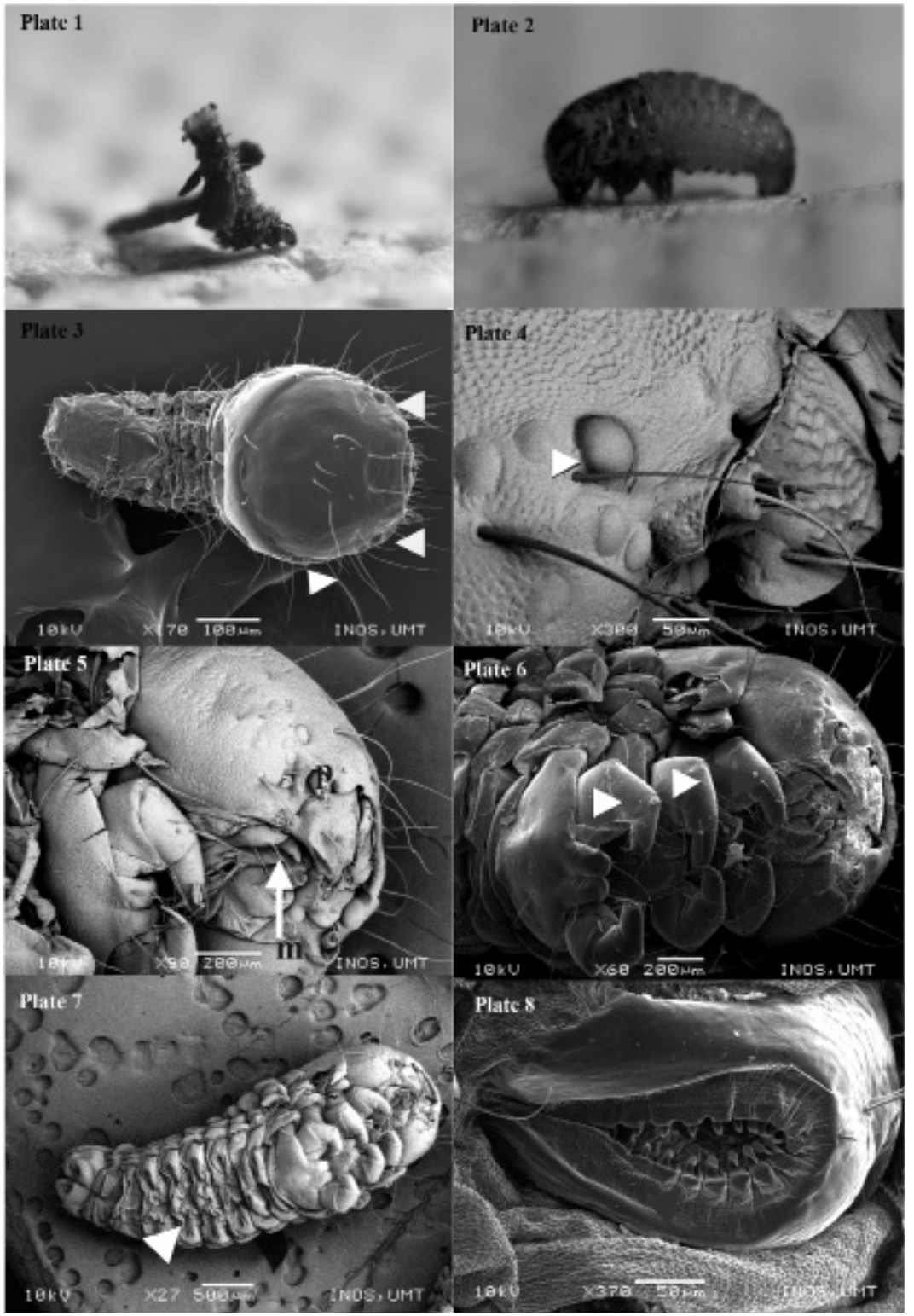
Plate 1
Plate 2
Plate 3
10kV X170 100µm INOS, UMT
Plate 4
10kV X300 50µm INOS, UMT
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Plate 8
10kV X370 50µm INOS, UMT

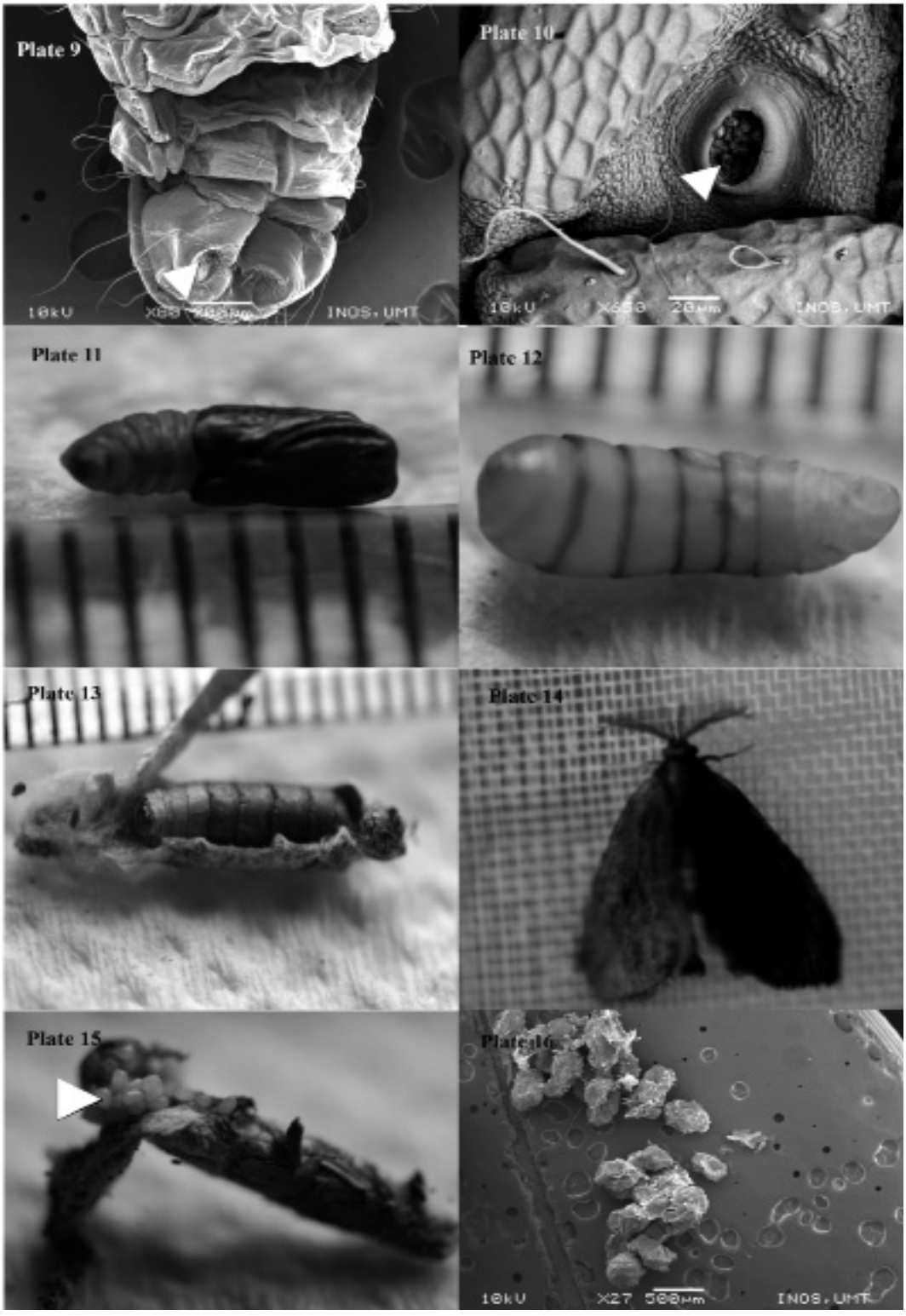
Plate 9
Plate 10
Plate 11
Plate 12
Plate 13
Plate 14
Plate 15
Plate 16

Plate 9: Larval anal proleg (white arrow), ventral view (x 80).

Plate 10: Larval spiracle (white arrow), approximately 50 µm, side view (x 650).

Plate 11: Male pupa removed from the case, showing the characteristic big blackish head. The pupa was approximately 6 mm in length.

Plate 12: Small, creamy yellowish-coloured head of female pupa, about half the size of the head of male pupa. The pupa was about 7 mm in length.

Plate 13: The case is split open into half to expose the adult female in the case. The adult female did not develop into a moth, was wingless and legless with a small dark-brown head and, located at the anterior end of the case, the yellowish-brown body. The adult female resembled a fusiform maggot.

Plate 14: The adult male was a fully-developed moth, between 10 – 12 mm with long clear wings and smoky-brown colour with a hairy black body.

Plate 15: The case is split longitudinally to expose a cluster of 200 – 300 eggs in the case.

Plate 16: Freshly-laid eggs were oblong, approximately 300 µm in width and 500 µm in length. (x 27).

approximately 300 µm in diameter and 500 µm long (Plate 16). The egg surface was covered with mucilaginous materials. After 5 – 8 days of incubation, the egg became translucent, containing the developing neonate. The developing neonate in the egg had a dark-brown body with a distinct black round spot in the middle. The local oil-palm planters referred to the eggs as black-headed eggs or "telur gelap" (in Malay).

The fecundity of *M. plana* was relatively low compared to other species of bagworm. For example, *Mahasena corbetti* laid between 2000 – 3000 eggs per female (Syed, 1978), while *Oiketicus kirbyi* between 3500 – 6000 eggs per female (Mexzon, *et al.*, 2003) and *Pteroma plagiophleps* an average of 1774 eggs per female (Howlader, 1990). The hatching rate of the eggs of *M. plana* was about 70%.

Life Cycle of *Metisa plana*

The average life cycle from eggs to adult in this controlled-rearing environment was 103.5 days (Plate 17). The larvae of *M. plana* in this study underwent six larval instars. The neonate started feeding immediately upon landing on the leaf surface, and built cases from dead plant materials, which they carried around with them as they fed. The 1st instar took approximately 9 – 16 days before molding into 2nd instar. As the larva fed and developed, the larva continuously enlarged the case by adding more plant material. The 2nd instar took around 14 – 17 days to mold into the 3rd instar. The 3rd instar took the longest period at approximately 16 – 18 days to develop into 4th instar. Development of subsequent instar stages 4th to 5th and 5th to 6th were slightly shorter, between 10 – 15 days and 12 – 16 days respectively. In total, the development of 1st to 6th instar stage was approximately 71.5 days. Apparently, the crucial period of effective control was the first 28 days after hatching or from 1st to 3rd instars larval period. After 4th instar, the larvae (5th and 6th instars) reduced their feeding activities and remained inactive in their case. The case was more solid and the larva was not easily reached by the chemical.

The larva was enclosed in the case, to pupate. Pupation duration was between 8 – 12 days, before the male adults emerged from the case, and flew to seek for the female. The gravid female remained inside the case (cocoon). Upon successful mating, the adult male remained active and died 3 to 4 days later. The adult female laid the eggs in the cocoon after mating, and left the case and died after a few hours.

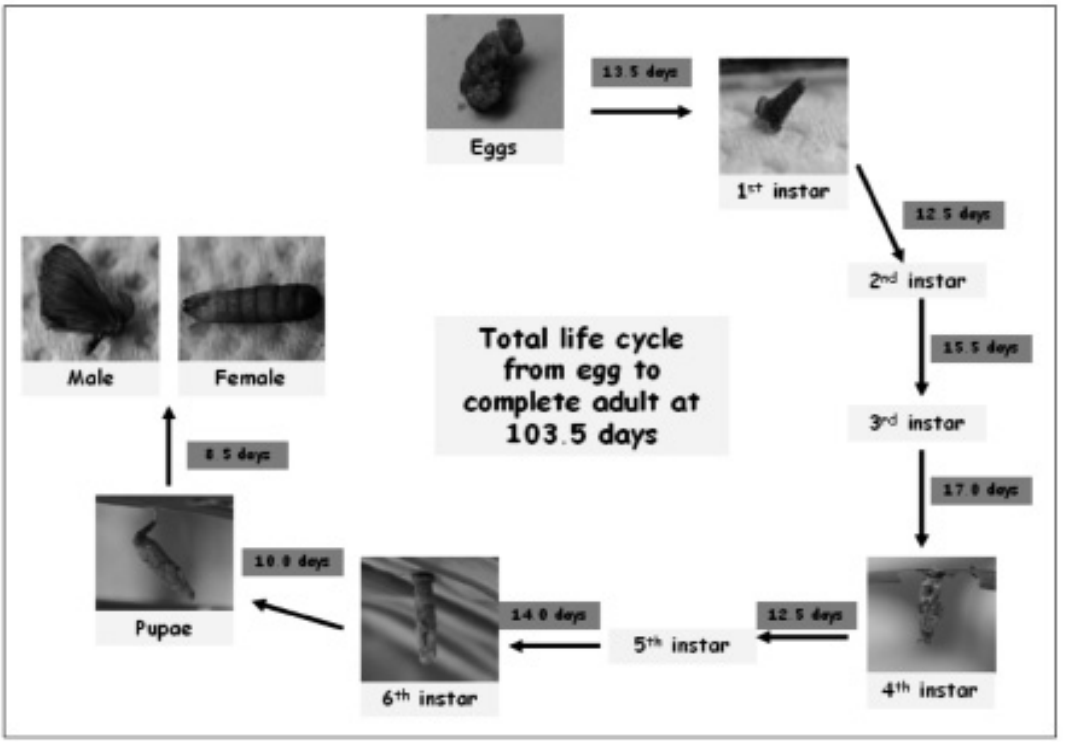

Plate 17: The complete life cycle of *Metisa plana*.

The freshly -laid eggs were yellowish in colour and contained a black spot after about 5 – 8 days, when the neonate was formed. The development duration from freshly-laid egg to hatching was between 12 – 15 days. The neonates (the young larvae) dropped onto the bottom frond or adaxial leaf surface. The neonates then started constructing their primary case.

Conclusion

Understanding the life cycle of *M. plana* is very crucial in pest management. It provides good indication for planning the strategy of *M. plana* control, and provides better understanding of the biology of the pest, besides helping to exploit the weaknesses in its life cycle for effective control of the pest.

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