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Life cycle of oil palm bunch moth, Tirathaba mundella walker (Lepidoptera: Pyralidae) reared under laboratory conditions on artificial diet

Su Chong Ming[1], Patricia King Jie Hung[2]*, Calvin Tan Zhe Khai[3], Kwan Yee Ming[4], Zakri Fitri Bin Ab Aziz[5], Joseph Bong Choon Fah[6], Ong Kian Huat[7]

[1-7] Faculty of Agriculture and Food Sciences, Universiti Putra Malaysia, Bintulu Sarawak Campus, Bintulu, Sarawak, Malaysia

Abstract
Tirathaba mundella Walker is an important pest in many oil palm plantations especially those established on peatland. The life cycle of *T. mundella* has not been described in detail despite its economic importance. This study aimed to describe the bionomic characteristics of *T. mundella*, rearing on an artificial diet, at 25 ^{O}C. The duration of a complete metamorphosis cycle of *T. mundella* was about 51.63 ± 3.80 days. The incubation period was the shortest (3.33 ± 0.80 days.), the most detrimental stage of the pest to oil palm is at larval stage, which took 33.90 ± 2.60 days before pupating. The pupal period took about 5 to 11 days with a mean ± SD period of 8.53 ± 1.48 days. The average of the adult's longevity was about 6.05 ± 1.81 days. There is no statistical difference between the duration of male and female longevity. Adult male and female moths reared in captivity feeded with 10% sucrose solution would mate readily and produce fertile eggs. The mean preoviposition period were 4 days. Successful of rearing *T. mundella* using artificial diet under laboratory conditions promises laboratory assessment for pesticide development which is less tedious and time consuming than conventional field experiments.

Keywords: oil palm bunch moth, *Tirathaba mundella*, life cycle, artificial diet

Introduction
The oil palm bunch moth species, *Tirathaba*, is classified under Pyralidae family. It was first recorded by Malaysian Agricultural Department in 1934 and has been mistakenly referred to as *Tirathaba fructivora* Megr. and *Melissoblaptes fructivora* Megr (Wood & Ng, 1974) [9]. There are only two recorded species from this family in Malaysia, namely *Tirathaba mundella* and *Tirathaba rufivena*, former is referring to oil palm (*Elaeis guineensis*) and latter is related to coconut (*Cocos nucifera* L.). *Tirathaba mundella* infestation in oil palm estate can be identified with the presence of moist reddish and dark brown faces over the surface of infested fruit bunches (Su, 2016; Calvin *et al*., 2018) [7, 1]. Possible of areca fruit, *Areca cathechu* and inflorescences of nipah, *Nipah fruticans* as alternate host for this pest has been proposed by Wood & Ng, 1974 [9].

In initial years, the level of infestation of *Tirathaba mundella* in Malaysia was mild and little damage was noticed, but of late, it becomes serious. Plantations with more than 30% trees infested has been reported and the severity could rise to 50% if no proper control programs are being implemented (Yaakop & Manaf, 2015) [10]. Moderate to severe pest infestations for oil palm estate with palms approaching maturity which include young mature palms of 3 to 6 years old (Chan, 1973; Su, 2016) [2, 7]. Indeed, major outbreaks concentrate to oil palm estates with huge areas of young mature palms and established on peat (Lim *et al*., 2012; Lim, 2012; Su, 2016; Su & Bong, 2017) [4, 5, 7, 8]. The conditions of such estates provide favorable conditions for the pest's regular multiplication and spreading. Moreover, often than not, potential natural predators and parasites are absence (Chan, 1973; Su, 2016) [2, 7].

Economic damage initiated at the inflorescence stage. The larvae of *Tirathaba mundella* can attack both male and female inflorescences at any stage of the inflorescence (Wood & Ng, 1974; Ng, 1977; Chan, 1973; Yaakop & Manaf, 2015; Su, 2016) [9, 6, 2, 10, 7]. In the cases of severe attack on female inflorescences, early bunch abortion can be resulted (Wood & Ng, 1974; Ng, 1977; Khoo *et. al*., 1991; Su, 2016) [9, 6, 3, 7]. Despite the economic importance of this pest, not many detailed descriptions regard the life cycle aspect of *Tirathaba mundella* has been published. This study aimed to describe the bionomic characteristics of *T. mundella*, rearing under laboratory condition on an artificial diet.

Materials and Methods
Insect source
The adult moth of *Tirathaba mundella* were captured from oil palm estates situated between latitudes (4.01625) and longitudes (114.22799) in Miri Sarawak, Malaysia. The captured male moths were separated from the female before oviposition. For oviposition, the male and female were kept in a container measuring 14.5 cm (height) × 26 cm (length) × 26 cm (breadth) and covered with a white cloth. The male: female ratio was at 4: 6 for oviposition. Moths were provided with 10% sucrose solution in moistened cotton. Non-woven gauze was placed at the bottom of the container for oviposition purpose. The non-woven gauze was changed daily. The mating period was recorded, and the moths were kept until death. The laid eggs were kept in a separate plastic

container that was maintained at room temperature. Mesocarp fibre was added on top of the artificial diet (Figure A) to provide hiding space for the newly hatched larvae while maintaining easy access to feed on the diet for their successful development. Hatched neonatal larvae were used for subsequent experiments.

Fig A: Feeding ground for neonatal larvae

Insect Rearing

An artificial diet developed in Laboratory of Plant Health and Management, Universiti Putra Malaysia Bintulu Sarawak Campus and was used to rear *Tirathaba mundella* at room temperature. About 7.5 mL of diet were poured into a 6.0 cm (height) × 5.5 cm (diameter) cup. Two active neonate larvae upon hatching were transferred to each plastic cup with diet. The diets were changed weekly. All the larvae were reared at room temperature. Larval survival and development progress were monitored daily until pupation and adult emergence. Daily examination under a stereoscopic binocular microscope were carried out to detect moulting of larvae by observing present of old capsule, take measurements of head capsules at its broadest points, measurements of larvae length, and detect pupation, followed by taking measurements for the length and breadth of the pupae.

During pupation period, each individual insect was kept in a bigger plastic cup at 13 cm (height) × 9 cm (diameter), monitored every alternate day until adult emergence. When the adult insect emerged, similar procedures were repeated for the next generation of insect rearing.

Results and Discussions

The life cycle of oil palm bunch moth, *Tirathaba mundella* Walker has been studied by Wood and Ng as early as 1974 using natural oil palm loose fruits as food source. And it is the only general description published on *T. mundella* life cycle. In order to give a complete picture on the bionomic characteristics of *T. mundella* with regards to its life cycle, *T. mundella* were reared under laboratory condition on an artificial diet in this study. *T. mundella* Walker fed on artificial diet developed by UPMKB, two successive generations were recorded. The duration of a complete metamorphosis cycle of *T. mundella* was about 51.63 ± 3.80 days as shown in Table 1. The incubation period was the shortest (3.33 ± 0.80 days.), the larval stage was also the longest, 33.90 ± 2.60 days. There were a total of seven larvae instars identified (Table 1). Instars changed to their successive instar in less than 6 days. Instar 1 to instar 2 took 3.88 ± 0.56 days, instar 2 to instar 3 took 4.18 ± 0.78 days, instar 3 to instar 4 took 5.20 ± 1.11 days, instar 4 to instar 5 took 4.45 ± 1.18 days, instar 5 to instar 6 took 5.17 ± 0.75 days, instar 6 to instar 7 took 5.07 ± 0.74 and instar 7 took 5.93 ± 1.05 days before turning into pupae. This is different from Wood & Ng (1974) [9]. They only reported five instars with mean duration of larvae range from 14 to 17 days and the pupa duration range from 9 to 12 days (Wood & Ng, 1974) [9]. Our study shows larval stage was around 33.90 ± 2.60 days and there are seven instars. The differences could be due to different diet used and methods to detect metamorphosis. Ng (1977) [6] had noted the duration of larvae period varied among larvae reared on different food sources. He noted the larval stage lasted 21.3 days, 25 days, 27 days and 28.9 days for larvae fed on male inflorescence, ripe fruitlets, young fruitlets and immature fruitlets respectively, longer than he first reported in Wood & Ng (1974) [9]. However, the fate of nutritional spectra in *Tirathaba mundella* during its life cycle phases is still poorly understood. Worth noting, pupal stage for *Tirathaba mundella* generally ranged from 7 to 12 days. Our observation has shown that pupal stage took average 8.53 ± 1.48 day (Table 1). Our study has shown that the average of *Tirathaba mundella* adult's longevity was about 6.05 ± 1.81 days. There is no statistical difference between the duration of male and female longevity.

T. mundella larvae were able to complete their life cycle with the nutrition provided by the artificial diet developed by UPMKB and reared under laboratory conditions. The characteristics of adult moths has been recorded and shown in Figure 1 and Figure 2. Adult moths had silvery-grey forewings with a slight greenish tinge and row of black spots separates the yellowish-brown outer wing fringe. The hind-wings and abdomen were light yellowish brown in colour while the ventral abdomen and the legs were silvery-grey. The wingspan of the males (Figure 1) were slightly smaller (9.96 ± 0.57 mm) than the female moth (Figure 2) (12.48 ± 0.46 mm). The preoviposition period ranged from 3 to 5 days with an average of 4 days. Both male and female adult moth live for about the same duration ranging from 5 to 9 days with mean ± SD of 6.05 ± 1.81 days. The adult reared in captivity with 10% sucrose solution would mate readily and produce fertile eggs.

Ratio male: female adult moth at 4: 6 offered good mating outcomes. The mating period was noticed around two to three days after mixing the correct ratio of male and female moth in a container. Both male and female moth live for a period of 5 to 9 days counted from the day emerged from pupa and eggs started to be seen from third day onwards.

The eggs of *T. mundella* were laid in masses and placed side by side to each other. Eggs were oval in shape and with 0.40 ± 0.06 mm x 0.27 ± 0.04 mm in dimensions (Table 3). Fertile eggs were orange-red in colour (Figure 3), while infertile eggs were pale yellow in colour (Figure 4). A day before hatching,

a black head capsule of the larva could be readily seen in the egg (Figure 5). The incubation period ranged from 3 to 5 days with a mean duration of 4 days.

The larvae of *T. mundella* were light brown in colour at younger stage. The anterior were darker than posterior. Generally the colour grow darker upon maturity. There were 7 instars identified for larvae feed on artificial diet keep in captivity under laboratory conditions. The body length, head capsule measurements and period of days for each larva instar were shown in Table 1, 2 and 3 respectively. The mean ± SD larva period for instar 1 and 2 were 3.88 ± 0.56 and 4.18 ± 0.78 days respectively and carried a mean ± SD head capsule measurement of 0.18±0.01mm (Figure 6) and 0.25 ± 0.02 mm (Figure 7); the mean ± SD body length measurement were 0.95 ± 0.10 mm (Figure 8) and 2.00 ± 0.43 mm (Figure 9). Larvae instar 3 had 0.35 ± 0.04 mm of head capsule (Figure 10) with a body length of 3.66 ± 0.42 mm (Figure 10) and the mean larva period for 3rd instar was 5.20 days.

From 4th instar onwards and until 7th instar, the growth rate of the larvae accentuate, almost double the initial growth rate within a mean larva period of about 5 to 6 days. The mean ± SD head capsule measurement obtained from 4th, 5th, 6th and 7th instar were 0.54 ± 0.07 mm (Figure 12), 0.75 ± 0.06 mm (Figure 13), 0.95 ± 0.07 mm (Figure 14) and 1.25 ± 0.14 mm (Figure 15) respectively. On the other hand, the mean ± SD body length obtained from 4th, 5th 6th and 7th instar were 4.80 ± 0.50 mm (Figure 16), 7.44 ± 0.96 mm (Figure 17), 9.52±0.58 mm (Figure 18) and 13.67 ± 2.20 mm (Figure 19) respectively. The total period of all the 7 instars larva ended in about 33.90 ± 2.60 days and it was longer than the larval period for larvae fed on male inflorescence, ripe fruitlets, young fruitlets and immature fruitlets as reported in Ng (1977) [6]. This indicates the diet source influences the length of larval period. The artificial diet developed by UPMKB used in this study extended the larval period and this would allow detailed study of potential pesticides against the most detrimental stage of *Tirathaba mundella*.

Pupation normally take place in a cocoon that consist of tough fibrous membrane and incorporated with faecel matter and plant fibres. The cocoon was ovoid and reddish in colour with a mean ± SD body length of 11.90 ± 0.94 mm and a mean ± SD breadth of 3.21 ± 0.26 mm (Figure 20). Pupa period ranged from 6 to 11 days with a mean period of 8.53 ± 1.48 days before the emergence of the adult moth (Table 1). This is however in agreement with Ng (1977) [6] and indicates that diets used in rearing *Tirathaba mundella* in both Ng (1977) [6] and this study have little impact on the pupa period.

Conclusion

The findings of this study suggested that full life cycles could be obtained by rearing the oil palm bunch moth *T. mundella* Walker on artificial diet developed by UPMKB. The mean ± SD duration of the life cycle of oil palm bunch moth, *Tirathaba mundella* Walker from egg stage until adult moth took about 51.63 ± 3.80 days, which is approximately one and a half month. The artificial diet used in this study extended the larval period as compared to the pest fed on natural diets. This could be a useful tool to study the most detrimental stage of *Tirathaba mundella* in pesticide development research without affecting the duration of pupa stage and production of fertile eggs. The use of this artificial diet was able to produce successive generations and provides the detail description of the life cycle of *Tirathaba mundella* for in depth understanding of the bionomic characteristics of this pest.

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Table 1: Life cycle period of *Tirathaba mundella* fed on artificial diet under laboratory conditions

Food source	Larvae instars duration (Mean ± standard deviation in days)							Total larvae period (Mean in days)	Pupa duration mean (Range)	Adult moth period duration mean (Range)	Total life cycle period duration mean (Range)
	1st	2nd	3rd	4th	5th	6th	7th				
Artificial Diet	3.88 ± 0.56	4.18 ± 0.78	5.20 ± 1.11	4.45 ± 1.18	5.17 ± 0.75	5.07 ± 0.74	5.93 ± 1.05	33.90 ± 2.60	8.53 ± 1.48	6.05 ± 1.81	51.63 ± 3.80

Table 2: Life cycle period of *Tirathaba mundella* fed on artificial diet under laboratory conditions

Food source	Larvae instars duration (Mean ± Standard deviation in days)							Total larvae period (Mean in days)	Pupa duration mean (Range)	Adult moth period duration mean (Range)	Total life cycle period duration mean (Range)
	1st	2nd	3rd	4th	5th	6th	7th				
Artificial Diet	3.88 ± 0.56	4.18 ± 0.78	5.20 ± 1.11	4.45 ± 1.18	5.17 ± 0.75	5.07 ± 0.74	5.93 ± 1.05	33.90 ± 2.60	8.53 ± 1.48	6.05 ± 1.81	51.63 ± 3.80

Number of studied subjects=30

Table 3: Morphometrics (mm) for different larvae instars of *T. mundella* walker fed on artificial diet under laboratory conditions

Morphometrics measurement (mm)	Larvae Instars of *Tirathaba mundella* Walker						
	1st	2nd	3rd	4th	5th	6th	7th
Mean Body Length	0.95 ± 0.10	2.00 ± 0.43	3.66 ± 0.42	4.80 ± 0.50	7.44 ± 0.96	9.52 ± 0.58	13.67 ± 2.20
Mean Head Capsule	0.18 ± 0.01	0.25 ± 0.02	0.35 ± 0.04	0.54 ± 0.07	0.75 ± 0.06	0.95 ± 0.07	1.25 ± 0.14

Number of studied subjects=40

Table 4: Morphometrics (mm) for Egg, pupa and adult moth of *Tirathaba mundella* walker

Different life stage	Morphometrics measurement (mean ± SD)	
	Length (mm)	Breadth/wingspan (mm)
Egg	0.40 ± 0.06	0.27 ± 0.04
Pupa	11.90 ± 0.94	3.21 ± 0.26
Adult Moth (Male)	10.28 ± 0.29	9.96 ± 0.57
Adult Moth (Female)	11.85 ± 0.23	12.48 ± 0.46

Fig 1: Male adult moth of *Tirathaba mundella* Walker

Fig 2: Female adult moth of *Tirathaba mundella* Walker

Fig 3: Fertile egg of *Tirathaba mundella* Walker (reddish yellow in colour)

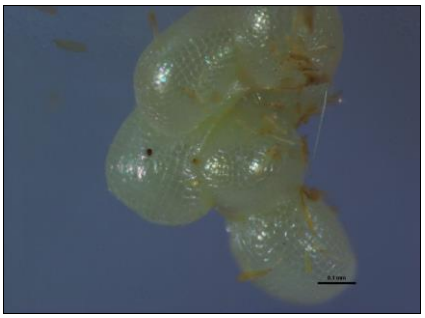

Fig 4: Cluster of infertile eggs of *Tirathaba mundella* Walker (whitish yellow in colour)

Fig 5: Cluster of fertile eggs with readily seen black head capsule of *T. mundella* Walker one day before hatching

Fig 6: Head capsule for 1st Instar larvae of *T. mundella Walker*

Fig 7: Head capsule for 2nd instar larvae of *T. mundella* Walker

Fig 8: 1st Instar larvae of *T. mundella* Walker

Fig 9: 2nd Instar larvae of *T. mundella* Walker

Fig 10: Head capsule for 3rd Instar larvae of *T. mundella Walker*

Fig 11: 3rd Instar larvae of *T. mundella* Walker

Fig 12: Head capsule for 4th instar of *T. mundella* Walker

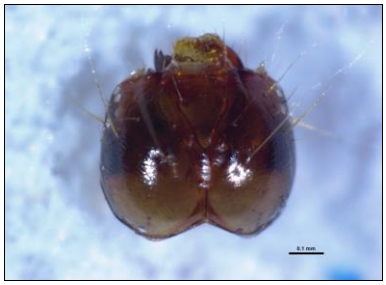

Fig 13: Head capsule for 5th instar larvae of *T. mundella* Walker

Fig 14: Head capsule for 6th instar larvae of *T. mundella* Walker

Fig 15: Head capsule for 7th instar larvae of *T. mundella* Walker

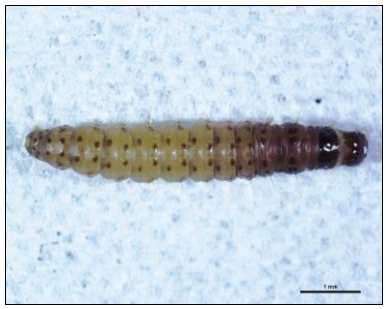

Fig 16: 4th instar larvae of *T. mundella* Walker

Fig 17: 5th instar larvae of *T. mundella* Walker

Fig 18: 6th instar larvae of *T. mundella* Walker

Fig 19: 7th instar larvae of *T. mundella* Walker

Fig 20: Ovoid and reddish brown colour cocoon of *T. mundella* Walker

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