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LIFE CYCLE AND MORPHOLOGICAL DESCRIPTIONS OF *Helopeltis theivora* WATERHOUSE (HEMIPTERA: MIRIDAE)

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ABSTRACT

The tea mosquito bug, *Helopeltis theivora* Waterhouse, is a serious pest of tea that damages buds, young shoots, and tender leaves mainly in Asian countries, including Malaysia. It is essential to understand the life cycle of this insect pest in order to apply the most effective management techniques. Therefore, the study aimed to gain a better understanding of the life cycle of *H. theivora*. The life cycle study of *H. theivora* showed that the incubation period was 8.40±0.15 days and the nymphal duration was 15.20±0.96 days, underwent five nymphal instars characterized by the length of the antenna. The mean longevity of adult females was 37.23±1.65 days, whereas for adult males was 29.11±0.92 days. The total life cycle of the females and males lasted 60.83±3.45 days and 52.71±4.33 days, respectively. On average, each female produced 156.47±16.33 eggs, with the sex ratio male to female was 1:1.09. The length and width of eggs were 1.134±0.02 mm and 0.283±0.01 mm, respectively, with two unequal respiratory horns at the tip. The first and second instar nymphs were light orange, and later instars were greenish to green. The full-grown fifth nymphal instar measured at 4.65±0.03 mm body length and 7.20±0.02 mm antennal length. Males and females of *H. theivora* were differentiated by the size of the body. The pronotal collar was bicoloured in both sexes. The findings of this study could be useful to formulate an effective management program against this pest.

Keywords: Pest, tea; *Helopeltis theivora*; life cycle; morphology

ABSTRAK

Serangga perosak teh, *Helopeltis theivora* Waterhouse, merupakan perosak utama tanaman teh yang merosakkan tunas, pucuk muda dan daun lembut, terutamanya di negara-negara Asia termasuk Malaysia. Ianya adalah penting untuk mengetahui kitaran hidup serangga perosak ini bagi tujuan aplikasi teknik pengurusan yang paling berkesan. Oleh itu, kajian ini bertujuan untuk mendapatkan kefahaman yang lebih baik tentang kitaran hayat *H. theivora*. Kajian kitaran hayat *H. theivora* menunjukkan bahawa masa pengeraman adalah 8.40±0.15 hari manakala tempoh nimfa ialah 15.20±0.96 hari dan mempunyai lima instar nimfa yang dicirikan oleh panjang antena. Purata hayat betina dewasa ialah 37.23±1.65 hari, manakala bagi jantan dewasa ialah 29.11±0.92 hari. Kitaran hayat betina dan jantan masing-masing adalah 60.83±3.45 hari dan 52.71±4.33 hari. Secara purata, seekor betina menghasilkan 156.47±16.33 telur, dengan nisbah jantan kepada betina ialah 1:1.09. Panjang dan lebar telur masing-masing adalah 1.134±0.02 mm dan 0.283±0.01 mm, dan terdapat dua tanduk pernafasan yang tidak sama panjang di hujungnya. Nimfa instar pertama dan kedua berwarna jingga muda, dan instar seterusnya berwarna kehijauan ke hijau. Nimfa instar kelima berukuran 4.65±0.03 mm panjang badan dan 7.20±0.02 mm panjang antena. Jantan dan betina *H. theivora* dibezakan mengikut saiz badan. Kolar pronotum adalah dwiwarna pada kedua-dua jantina. Dapatan kajian ini berguna untuk merangka program pengurusan yang berkesan terhadap perosak ini.

Kata kunci: Perosak; teh; *Helopeltis theivora*; kitaran hayat; morfologi

INTRODUCTION

Tea mosquito bug, *Helopeltis theivora* is a species under the genus *Helopeltis* belongs to the family Miridae (Hemiptera). It is an insect pest that significantly reduces the economic value of tea both quantitatively and qualitatively (Roy & Prasad 2018). According to Roy et al. (2015), the tea mosquito bug is found mainly in the Asian countries. The damage is estimated as high as 80% (Pandey et al. 2021), occurs all year round, and is a major constraint to tea production in South and Southeast Asia (Roy et al. 2015). *Helopeltis theivora* is also one of the significant sucking insect pests in Malaysia (Latip et al. 2010; Pandey et al. 2021; Roy et al. 2015).

The study on the life cycle of *H. theivora* has been carried out in different countries (Antony et al. 2018; Roy et al. 2015). Previous studies found that *H. theivora* went through incomplete metamorphosis with four (Kalita et al. 2018; Tea Research Association 2010) to five nymphal instars (Antony et al. 2018; Mamun 2022; Thube et al. 2019) which is influenced by the environmental condition, geographical region, and host plants. Hence, the rate of development and reproduction of *H. theivora* are not uniform and regular among countries and tea varieties. In Malaysia, the life cycle study of *H. theivora* was done by Miller (1941) and Tan (1974) on cocoa only. However, there is no study on the life cycle of *H. theivora* on tea in Malaysia.

Therefore, understanding the life cycle and determining the nymphal instar of *H. theivora* are crucial for providing comprehensive information on their biology, which can be used to formulate an effective pest management strategy (Antony et al. 2018; Srikumar & Bhat 2013). Knowledge of the morphological characteristics helps accurately identify different growth stages of *H. theivora*, which is very important for determining the extent of damage to tea plants (Roy et al. 2015; Serrana et al. 2021). Laboratory studies can provide significant information on the development, survival, longevity, and reproduction of insects (Rostaee et al. 2021; Roseli et al. 2019). Therefore, this study was conducted to determine the life cycle of *H. theivora* as well as to describe the morphology and morphometric measurement of this pest at every life stage.

MATERIALS AND METHODS

Helopeltis theivora Collection

The nymphs (no. 200) and adults (no. 50 males and no. 50 females) of *H. theivora* were collected by hand picking from the infested tea plantation sections of Bharat tea plantation, MARDI tea plantation, and Sungei Palas tea estate (BOH Plantations) in Cameron Highland, Pahang, Malaysia, in November 2019. Cameron Highland is the main tea growing area in Malaysia. Hence, insect sampling was done to focus on this area. Nymphs and adults were collected for laboratory rearing instead of eggs because they were easier to find and collect. Eggs were typically embedded within plant tissues, making them difficult to locate. Collecting nymphs and adults allowed for immediate rearing, saving time and ensuring a steady supply of insects for research (Serrana et al. 2021; Srikumar & Bhat 2012).

Insects were collected between 7.00-9.00 am and 5.00-7.00 pm on the same day and this was continued for three consecutive days. *Helopeltis theivora* were collected during this time to align with their natural feeding behavior and favorable environmental conditions, as they were more active during these cooler periods of the day (Roy et al. 2015). High temperatures and strong sunlight at midday could have reduced their movement and feeding activity. Collecting them during these times helped capture more bugs while minimizing stress or mortality caused by excessive heat. The collected nymphs and adults were kept in plastic aquariums separately covered with muslin clothes. Cut tea shoots in glass vials filled with water were supplied as food sources. All the insects were taken to the insectary under the Department of Plant Protection, Faculty of Agriculture, UPM for rearing.

Laboratory Culture and Rearing of *H. theivora*

The culture and rearing of *H. theivora* in the laboratory condition were adopted and modified from Sudhakaran & Muraleedharan (2006). Cultures were reared and maintained in the insectary, Department of Plant Protection, Faculty of Agriculture, Universiti Putra Malaysia, under laboratory conditions with a temperature of 25.0±2.0°C and 70.0±5.0% humidity with 12h:12h photoperiod.

Nymphs were transferred to a plastic aquarium of 29 x 19 x 18 cm and fresh tea shoots in a glass vial (28 ml) filled with water were supplied as food (tea shoots were collected from the Tea Plot of the University Agriculture Park, UPM). The vial was replaced with new vials containing fresh tea shoots every alternate day. While replacing the vial, the nymphs on the tea shoots were carefully removed and released onto the new shoots. This was continued till the emergence of adults.

The males and females were kept for pairing (5 pairs per rearing cage, 1:1 sex ratio) in a separate plastic aquarium measuring 35 x 20 x 24 cm. Fresh tea shoots in glass vials with water were supplied for feeding and oviposition. Copulation was observed when the male mounted the female, and directed its abdomen downwards along the female's right side. Successful copulation was confirmed when the pair remained attached in fully opposite directions for several minutes, indicating proper genital alignment and sperm transfer (Serrana et al. 2021). Every alternate day, the tea shoots were removed and examined for eggs with the help of a magnifier hand lens (10X). The presence of a pair of fine terminal respiratory filaments projecting from the surface of the plant tissue was indicative of the existence of eggs. The egg-laden shots were kept in a glass vial filled with tap water in a separate plastic aquarium (40 x 23 x 28 cm). Another glass vial containing fresh tea shoots was placed close to the glass vial containing egg-laden shoots so that the newly emerged nymphs could feed immediately. The water of the glass vial containing shoots with eggs was changed every alternate day to avoid fungus growth. Immediately after hatching, nymphs were transferred to nymphal rearing cages (29 x 19 x 18 cm). The fresh tea shoots kept in a water-filled vial were supplied as feed on every alternate day. This process was continued until all nymphs turned into adults. Further, the newly emerged males and females were paired, allowing females to oviposition. This process was repeated to maintain the tea mosquito bug stock culture throughout the experimental period.

Determination of the Life Cycle of *H. theivora*

Determination of the developmental time of *H. theivora* involved egg, nymph, and adult. The life cycle of *H. theivora* was investigated in the laboratory from December 2019 to March 2020 for one generation using tea shoots as a food source. Different biological parameters, such as the incubation period of eggs, nymphal period, sex ratio, longevity of adults, and fecundity, were recorded.

Incubation period

Ten to fifteen eggs with 2-3 tea shoots were kept in a plastic aquarium measuring 29 x 19 x 18 cm and labeled with the first-day egg date. A glass vial filled with fresh tea shoots was placed in each cage. The eggs were observed daily until all eggs were hatched. This was replicated five times, and 50 eggs were observed.

Nymphal period

Ten newly hatched nymphs of *H. theivora* were transferred individually into each nymphal rearing cage (21 x 13 x 12 cm). The container was labelled with the date of the first day of the first instar. The newly hatched nymphs were fed on fresh tea shoots in a water-filled vial. The nymphs were observed daily to notice and record the moult of each instar. To avoid mixing with the previous cast skin, the cast skin was removed with a fine camel hair brush immediately after each moult. Tea shoots were changed every alternate day for the first and second instar nymphs, while they were changed daily for the third instar until the fifth instar nymph. The period from instar to instar and nymphal stage to adult emergence were recorded. This was replicated five times.

Longevity, fecundity, and sex ratio

Five pairs of newly emerged males and females were put in separate plastic aquariums (29×19×18 cm) containing tea shoots for feeding and egg-laying. The mouth of each plastic aquarium was covered with a piece of muslin cloth and tightened with a lid. The number of eggs produced by each pair and the adult's longevity were recorded. This was replicated five times. The sex ratio was calculated using the number of males and females that emerged from five replicates (each replicate included ten nymphs).

Determination of Nymphal Instar Stages

Ten pairs of *H. theivora* were released in a cage (40×23×28 cm) for 24 hours for oviposition on tea shoots placed in a glass vial filled with water. Then, the shoots were removed from the cage and replaced by new shoots. Shoots containing eggs were placed in a plastic container (29×19×18 cm) with fresh tea shoots in another glass vial. All eggs were observed until hatching, and all first instar nymphs were transferred into new plastic containers (21×19×18 cm) with fresh tea shoots in water-filled glass vials. The container was labelled with the date of the first day of the first instar. The nymphs were reared in laboratory conditions until the last nymph moult. This process was repeated until the experiment was finished.

One-day-old first instar nymphs were taken to determine morphometric measurement. Every day, five nymphs were taken out and then put in a glass vial with a cotton ball soaked in ethyl acetate for 2-3 minutes to make them motionless. Then, the nymphs were put on a Petri dish and spread with fine camel brush and forceps to take the morphometric measurement. The determination process continued until every nymph in the plastic containers had undergone a moult. Measurements were taken by a Dino-eye microscope eyepiece (ANMO Electronics Corporation, Taiwan) attached to a Wild Heerbrugg Microscope (M3Z Switzerland). The software Dino-capture 2.0 was used to take pictures of the specimens. The morphometric characteristics of nymphs in this study were body and antennae lengths of different instars.

Morphological Description and Morphometric Measurement of *H. theivora* at Every Stage

The external morphology of *H. theivora* was observed and distinguished based on its colour, shape, and marking of the body. The morphometric measurements were taken from 10 individuals for each stage. The measurements include (Table 1, Figure 1):

Table 1. The morphometric measurements of the egg and different body parts of *H. theivora*

Sl. No.	Parameter	Description	Figure	Reference
1	Length of egg	From the top to bottom end of the egg	1a	Sudhakaran & Muraleedharan (2006)
2	Width of egg	The maximum wide of the egg	1b	Sudhakaran & Muraleedharan (2006)
3	Length of respiratory horn	From the base to the apex of the respiratory horn	1c,d	Serrana et al. (2021)
4	The body length of each nymphal stage	Distance from the edge of the head capsule until the edge of the posterior end	1e	Serrana et al. (2021)
5	The body length of the adult	Distance between the apex of the tylus to the posterior tip of the folded forewings along the middorsal line	1f	Yeshwanth (2013)
6	The antennal length of nymph and adult	Distance between the base to the apex of individual segments I, II, III, and IV, respectively, of the antenna	1g,h	Roy et al. (2015) Yeshwanth (2013)
7	Stylet length	Distance between the bases of the baculla to the apex of the stylet	1i	Sudhakaran & Muraleedharan (2006) Yeshwanth (2013)
8	length of the scutellar horn	From the base to the apex of the scutellar horn	1j	Sudhakaran & Muraleedharan (2006)
9	length of wing pad of nymph	Distance between articulatory points to the tip of the wing pad	1k	Sudhakaran & Muraleedharan (2006)
10	Wing length of adult	Distance between articulatory points of forewings to the tip of the wing membrane	1l	Yeshwanth (2013)

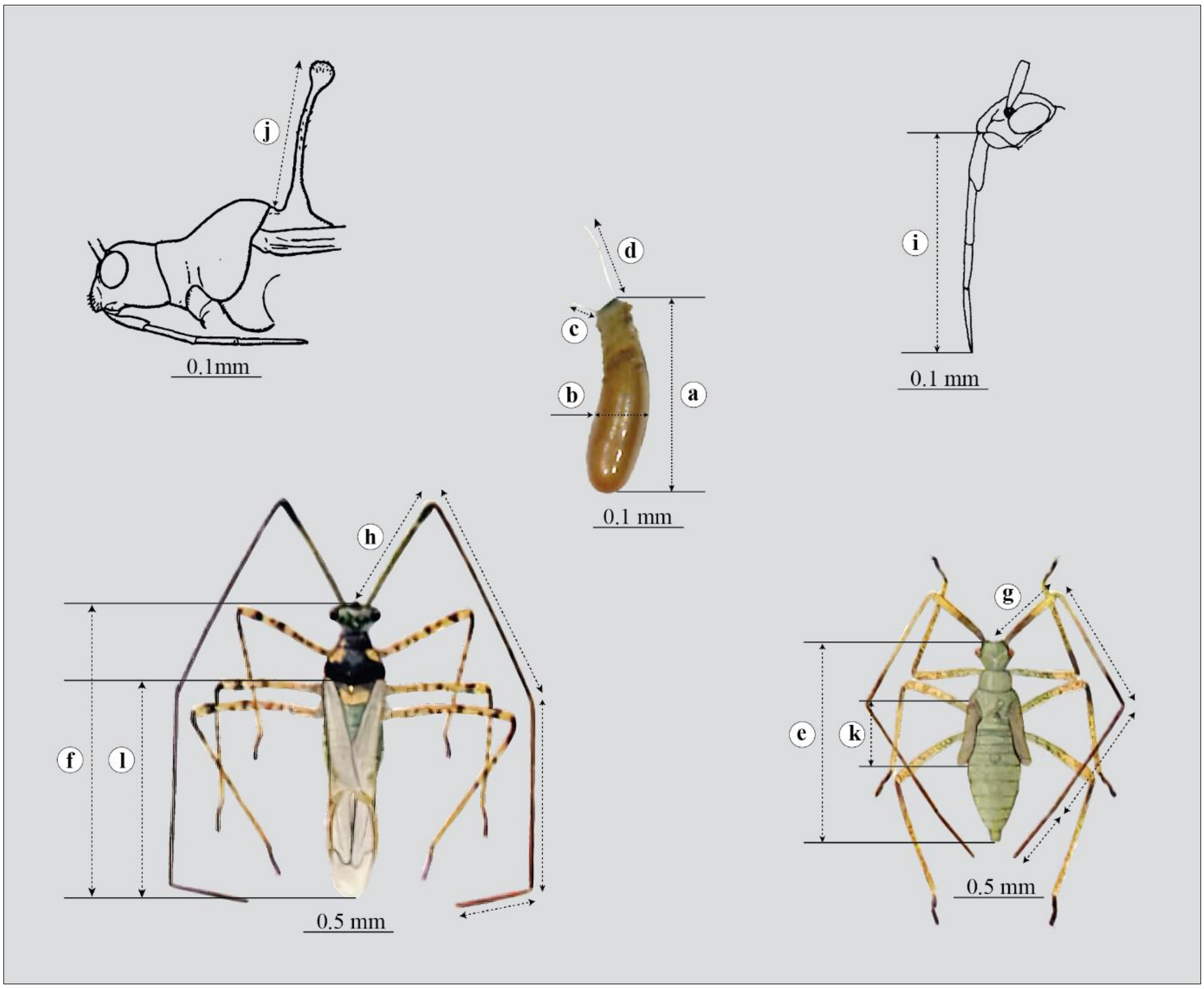

Figure 1. Measurement of egg and different body parts of *H. theivora*: (a, b) length and width of egg, (c, d) length of respiratory horns, (e) body length of nymphal stage, (f) body length of adults, (g, h) antennal length of nymph and adult, (i) stylet length, (j) length of scutellar horn, (k) wing pad of nymph, and (l) wing length of adult [Images except egg and stylet were taken and modified from Miller (1939, 1941) to indicate measurement]

Data Analysis

In the life cycle studies, the data recorded were added to calculate arithmetic mean and standard error (SE) using MS Excel. An independent two-sample T-test was performed to compare longevity between adult males and females. Data on the body length, antenna length, stylet length, scutellar horn length, and wing pad length of nymphal instars were analyzed by one-way ANOVA. Tukey's test was used to differentiate the means at a significance level of 0.05. The adult morphometric measurements, i.e., body length, antenna length, stylet length, scutellar horn length, and wing length between sexes, were analyzed using the independent two-sample T-test. The data were analyzed by SAS (version 9.4).

RESULT

Determination of the Life Cycle of *H. theivora*

Incubation period

The result revealed that the incubation period of *H. theivora* eggs ranged from 7 to 9 days. The mean value was 8.40±0.15 days (Table 2).

Table 2. Duration of different stages of the life cycle of *H. theivora* on tea under laboratory conditions

Life Stages	Duration (Day)	Mean±SE
Incubation period (days)	7-9	8.40±0.15
Nymphal duration (days)		
First instar	3-4	3.20±0.13
Second instar	2-4	2.90±0.28
Third instar	2-3	2.70±0.15
Fourth instar	2-3	2.80±0.13
Fifth instar	3-5	3.80±0.27
Total	12-19	15.20±0.96
Oviposition period	19-32	28.20±2.61
Fecundity	65-237	156.47±16.33
Adult longevity (days)		
Male	15-35	29.11±0.92
Female	20-45	37.23±1.65
Total life cycle (days)		
Male	34-63	52.71±4.33
Female	39-73	60.83±3.45
Sex ratio (Male: Female)		1:1.09

Nymphal period and instar determination

The nymphal period ranged from 12 to 19 days, averaging 15.20±0.96 days (Table 2). It was found that the first, second, third, fourth, and fifth instar nymphs had respective mean durations of 3.20±0.13, 2.90±0.28, 2.70±0.15, 2.80±0.13, and 3.8±0.27 days, respectively (Table 2).

The trend of daily changes in antenna length and body length of *H. theivora* nymph is shown in Figure 2. The length of the antenna increased between instars and varied with each instar. The length of the antenna increased significantly four times: from day three (2.50±0.03 mm) to four (3.17±0.04 mm), day six (3.33±0.02 mm) to seven (4.37±0.01 mm), day eight (4.55±0.02 mm) to nine (5.78±0.03 mm) and from day eleven (5.99±0.02 mm) to twelve (6.91±0.03 mm), with a corresponding increase of 0.67, 1.04, 1.23 and 0.92 mm. These increments produced steep gradients, indicating the moulting process had occurred during that date. Referring to the graph, *H. theivora* passed through five nymphal instars before emerging as an adult.

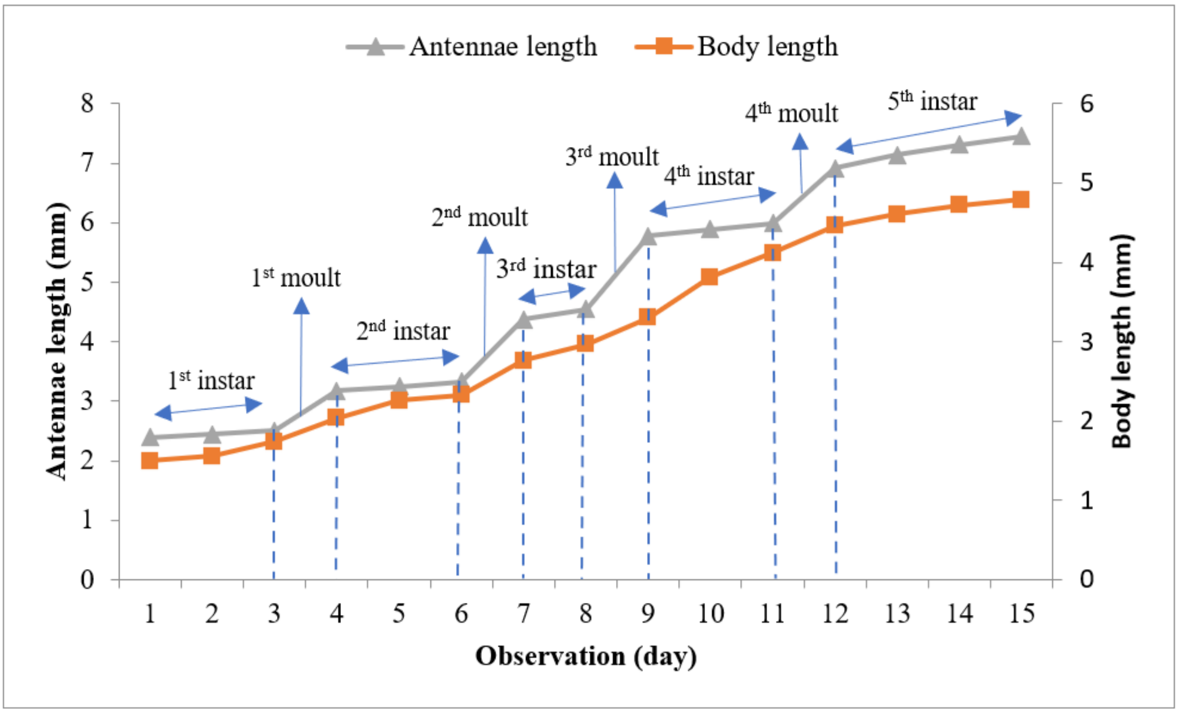

Figure 2. Determination of nymphal instars characterized by antennae length and body length

The trend of daily changes in body length was directly proportional to the length of the antenna, but it did not give a clear picture of differences between instars. Thus, the antenna length is more suitable as an indicator in determining nymphal instars than body length (Figure 2).

All nymphal instars had significantly variable antenna lengths (F = 2700.28; df = 4, 45; $P<0.05$). The average antenna length for the first, second, third, fourth, and fifth instars was 2.44±0.02 mm, 3.25±0.01 mm, 4.46±0.02 mm, 5.89±0.01 mm, and 7.20±0.02 mm, respectively. At the same time, there was a significant difference in body length among all nymphal instars (F = 1115.33; df = 4, 45; $P<0.05$), with values of 1.60±0.01 mm (first instar), 2.21±0.02 mm (second instar), 2.87±0.02 mm (third instar), 3.74±0.01 mm (fourth instar) and finally, 4.65±0.03 mm for fifth instar (Table 3).

Table 3. Measurement of body and antennal length of different nymphal instars

Nymphal Instars	**Mean±SE (mm)**	
	Body Length	**Antennal Length**
First instar	1.60±0.01e	2.44±0.02e
Second instar	2.21±0.02d	3.25±0.01d
Third instar	2.87±0.02c	4.46±0.02c
Fourth instar	3.74±0.01b	5.89±0.01b
Fifth instar	4.65±0.03a	7.20±0.02a

Means within column followed by different letters are significantly different by the Tukey's test at $P<0.05$.

Longevity of adult

The results showed significant longevity differences between males and females (t = 4.92, df = 48, *P<0.05*). Females survived longer than males. The mean longevity of adult females was observed as 37.23±1.65 days, ranging from 20 to 45 days, while the average longevity of males was 29.11±0.92 days with a range of 15 to 35 days (Table 2).

Fecundity

The present investigation observed that the fecundity ranged between 65-237 eggs per female, with a mean value of 156.4 ±16.33 eggs (Table 2).

Total life span and sex ratio

The overall lifespan of *H. theivora* from the incubation period until the death of an adult was recorded as 60.83±3.45 days for females and 52.71±4.33 days for males. The sex ratio of male to female was determined 1:1.09 (Table 2).

Morphological Description and Morphometric Measurements of *H. theivora* at Every Stage

Egg

Females deposited eggs singly or in batches of three to five, which were completely embedded in the epidermis tissue of tender shoots, leaf petioles, and the lower part of the midrib of leaves. A pair of unequal white-coloured respiratory horns were projected from the surface of plant tissue, indicating the presence of eggs (Figure 3). The newly laid eggs were white, cylindrical, slightly curved, and tapering at the tip (Figure 4a). Later, the eggs changed to light brown before hatching (Figure 4b). The mean length was measured at 1.134±0.02 mm, and the width was estimated at 0.283±0.01 mm (Table 4). The longer respiratory horn was 0.787±0.03 mm long, while the short was 0.264±0.02 mm.

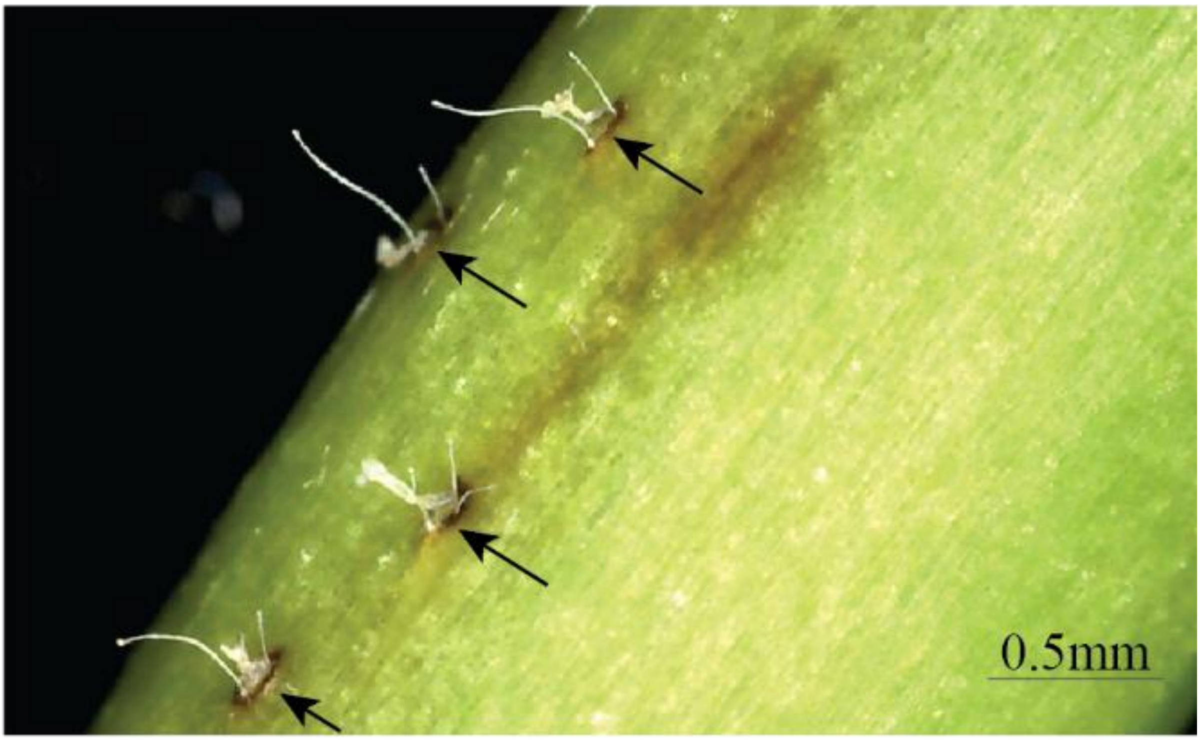

Figure 3. Eggs laid by a female adult in the epidermis of the stem showing filament outside

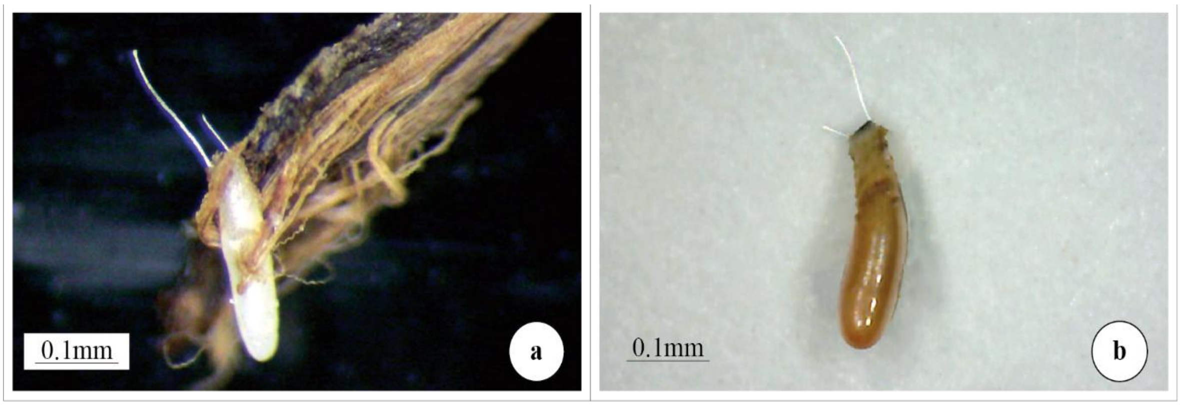

Figure 4. Eggs of *H. theivora* with respiratory horn (a) initial stage, and (b) later stage

Table 4. Measurement of eggs and the respiratory horn of *H. theivora*

Egg and Respiratory Horn	Average Measurement (mm±SE)
Egg	
Length	1.134±0.02
Width	0.283±0.01
Respiratory horn	
Long one	0.787±0.03
Short one	0.264±0.02

Nymph

The first instar nymph hatched out of the egg by opening the operculum at the periphery. After nymphal emergence, the embryonic membrane protruded from the empty eggshell, which remained buried in the plant tissue. Most eggs hatched between 8:00-10:00 am and 5:00-7:00 pm. Five nymphal instars occurred following egg hatching. The morphology of different nymphal instars was similar, with the main differences being body size, antenna length, and colour (Figures 5 a-e).

First instar

The freshly hatched nymphs were minute and light orange (Figure 5a). After a couple of hours, the head, antennae, legs, and abdomen turned light brown. Prominent and erect setae were present on the antennae, thoracic segments, abdomen, and legs. The eyes were pink, the antennae were longer than the body, and the rostrum reached half of the abdomen. The scutellar horn and wings were absent in this instar. The first instar nymph started feeding buds and tender leaves of the tea shoots immediately after emergence. The mean body length of this instar was 1.60±0.01 mm. Antennae were four segmented with a mean length of 2.44±0.02 mm (Table 5). The last abdominal segment was an elongated anal tube. Legs were elongated, yellowish-grey with fuscous bands. Duration of this stage ranged between 3-4 days with an average of 3.20±0.13 days (Table 2).

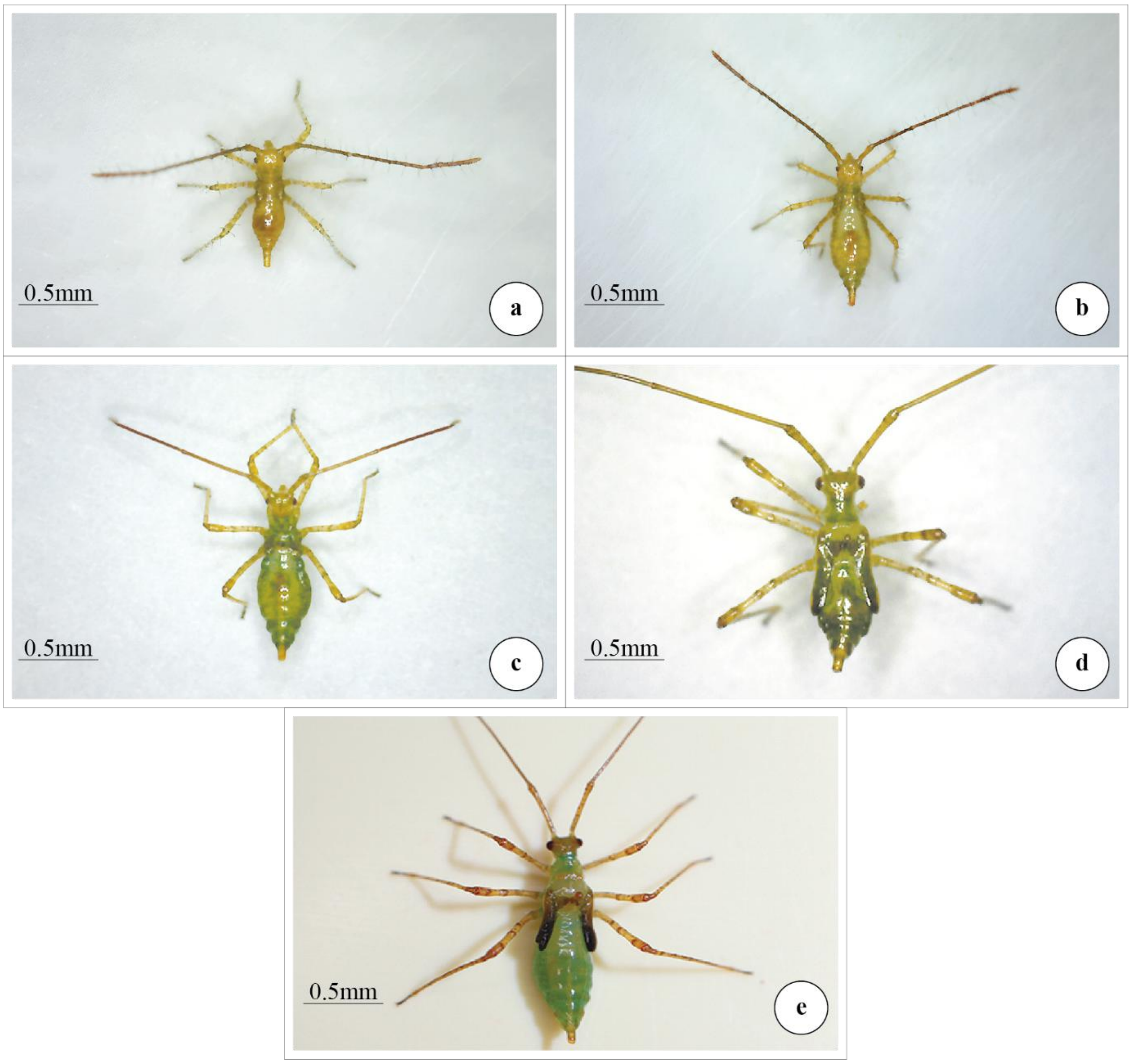

Figure 5. Nymphal instars of *H. theivora* (a) First instar, (b) Second instar, (c) Third instar, (d) Fourth instar and (e) Fifth instar

Table 5. Measurement of different body parts of nymphal instars of *H. theivora*

Body/Body Parts	**Average Measurement (mm±SE)**				
	First Instar	**Second Instar**	**Third Instar**	**Fourth Instar**	**Fifth Instar**
Body length	1.60±0.01e	2.21±0.02d	2.87±0.02c	3.74±0.01b	4.65±0.03a
Antennal length	2.44±0.02e	3.25±0.01d	4.46±0.02c	5.89±0.01b	7.20±0.02a
Stylet length	0.82±0.02e	1.23±0.01d	1.37±0.02c	1.52±0.01b	1.73±0.01a
Scutellar horn	-	0.19±0.01d	0.27±0.02c	0.39±0.01b	0.52±0.01a
Wing pad	-	-	0.28±0.01c	0.39±0.02b	0.53±0.01a

Means within the row followed by the different letters are significantly different by Tukey's test at *P<0.05*.

Second instar

The second instar's body was similar to the previous one but bigger than the first instar (Figure 5b). The head, first segment of antennae, legs, and abdomen were orange. Thorax was greenish. The scutellar horn without swollen knob was just visible in this instar. Wing buds were present but inconspicuous. The last abdominal segment was similar to the first instar. Legs were identical to the preceding; the tibia was covered with a few short setae. The average body length was 2.21±0.02 mm. Antennae were four segmented, and the average length was 3.25±0.01 mm (Table 5). Sexes were not distinct. Duration of the second instar varied between 2-4 days with an average of 2.90±0.28 days (Table 2).

Third instar

This instar's body was yellowish green (Figure 5c). Scutellar horn and wing buds were visible in this stage. The mean body length of the third instar nymph was 2.87±0.02 mm. Antennae were four segmented with a mean length of 4.46±0.02 mm (Table 5). Legs were yellowish-grey with more prominent black markings; sexes were not distinct. The mean longevity of the third instar was 2.70±0.15 days with a range of 2-3 days (Table 2).

Fourth instar

The body was greenish yellow (Figure 5d). The scutellar horn was pale yellow with a swollen knob. The wing buds became dark and extended over their abdomen. The mean body length of the fourth instar nymph was 3.74±0.01 mm. Antennae were four segmented with a mean length of 5.89±0.01 mm (Table 5). Legs were similar to the descriptions of the third instar except for the length. Sexes were not distinct. This instar lasted 2 to 3 days, averaging 2.80±0.13 days (Table 2).

Fifth instar

The fifth instar nymph was green in colour (Figure 5e). Thorax was reddish green. The abdomen was green. Wing buds covered half of the abdomen but did not overlap. Nymphs were 4.65±0.03 mm long. The antennae length of this instar was 7.20±0.02 mm (Table 5). The longevity of this instar was 3.80±0.27 days. This instar had the longest duration (Table 2).

Adult stage

Males and females of *H. theivora* were distinguishable by the size of the body (Figure 6 a-d). The newly emerged adults were yellowish, while the wings were grey. The characteristic colouration was developed within an hour. Antennae were four segmented with black. The base of the first antennal segment of both males and females had pale markings. The first segment of the antennae was thicker than the other three segments. The second segment was the longest, while the last was the shortest. The last portion of the 2nd antennal segment, 3rd and 4th antennal segments bore short stout setae. The head was black with a pale stripe laterally (Figure 7a). Eyes were oval and prominent. The pronotal collar was bicoloured in both sexes (Figure 7b). In the case of males, most of the posterior part of the pronotum was yellowish brown, whereas in females, it was broadly yellow.

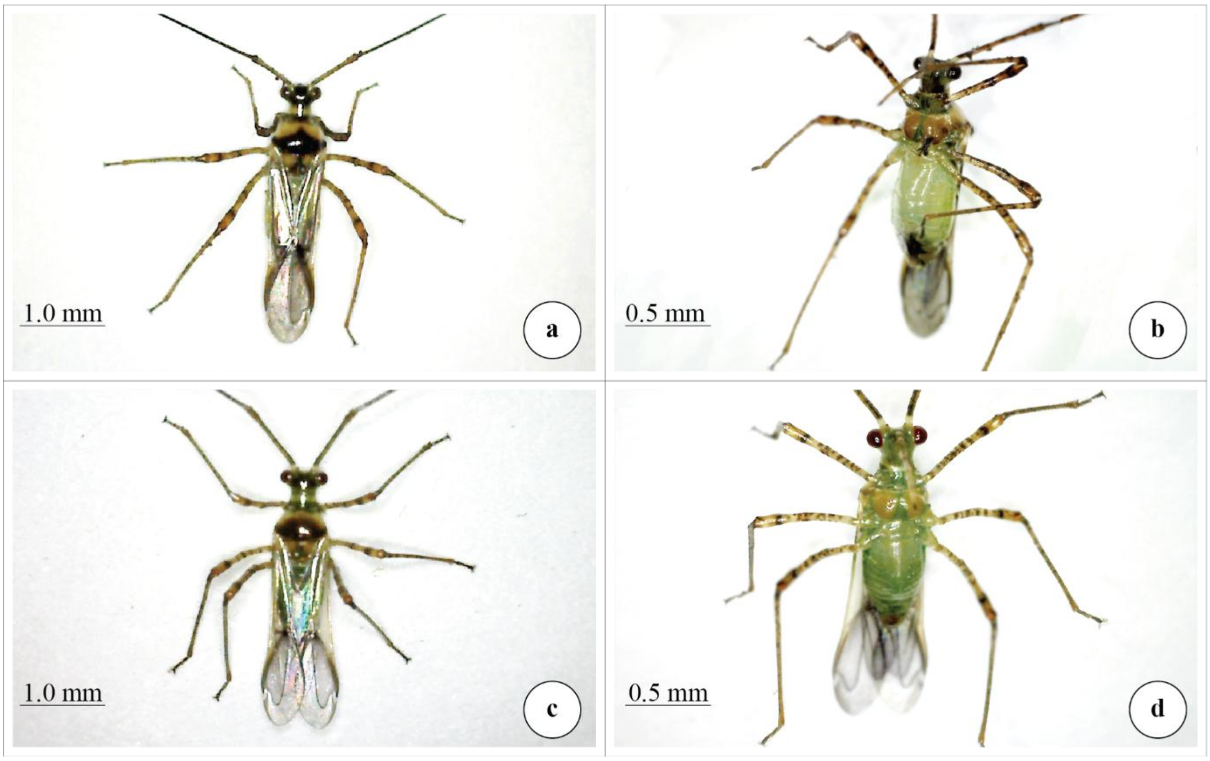

Figure 6. Adult female and male of *H. theivora* (a) dorsal and (b) ventral view of female, (c) dorsal and (d) ventral view of male

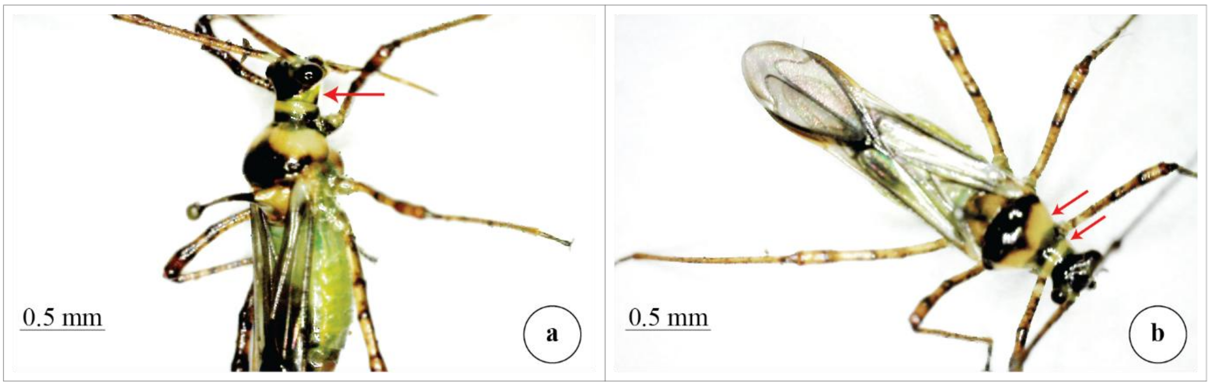

Figure 7. Head with lateral pale stripe (a), Bi-coloured pronotum (b)

The characteristic pin-like process called scutellar horn was raised from the scutellum and was yellowish brown. The scutellar horn was gently curved. The apex of the scutellar horn was swollen and funnel-shaped. The wings were blackish. The two pairs of wings overlap over the body when at rest. Hind wings were shorter, while the forewings covered the entire body. Abdominal sterna I-V was uniformly pale laterally. The abdomen was green for both female and male bugs. The female had a reddish-brown curved ovipositor. Legs were yellowish brown, with fuscous mottling and pale band basally.

The measurements of different body parts of the adult males and females of *H. theivora* are cited in Table 6. The body length of both sexes, of which 5.87±0.05 mm (male) and 6.89±0.19 mm (female), was statistically different (t = 9.08, df = 18, $P<0.05$). The antennal length of males (11.05±0.17) and females (11.94±0.28) was significantly different (t = 5.75, df = 18, *P<0.05*). On the other hand, the stylet length of both males (1.90±0.05 mm) and females (2.13±0.04 mm) was statistically different (t = 4.32, df = 18, *P<0.05*), while the length of the scutellar horn of male (1.19±0.05 mm) and female (1.31±0.08 mm) was significantly different (t = 2.51, df = 18, *P<0.05*). Forewing length of male (4.87±0.05 mm) was significantly (t = 11.62, df = 18, *P<0.05*) shorter than female (5.64±0.10 mm), and also hindwing of male (3.77±0.06 mm) was significantly (t = 12.42, df = 18, *P<0.05*) smaller than female (4.55±0.08 mm).

Table 6. Measurements of different body parts of the adult males and females of *H. theivora*

Body/Body Parts	**Average Measurement (mm±SE)**	
	Male	**Female**
Body length	5.87±0.05b	6.89±0.19a
Antenna	11.05±0.17b	11.94±0.28a
Stylet length	1.90±0.05b	2.13±0.04a
Scutellar horn	1.19±0.05b	1.31±0.08a
Wing		
Forewing	4.87±0.05b	5.64±0.10a
Hindwing	3.77±0.06b	4.55±0.08a

Means within the row followed by the different letters are significantly different at *P<0.05*.

DISCUSSION

Determination of the Life Cycle of *H. theivora*

The result of the incubation period is in accordance with the study of Roy et al. (2015) and Sudhakaran & Muraleedharan (2006), while Antony et al. (2018) reported 6.0 days for the incubation period in tea. The nymphal duration of this study was in line with Roy et al. (2015) and Sudhakaran & Muraleedharan (2006), who recorded that the nymphal development period varied from 11 to 19 days and 8.4 to 16.2 days, respectively. The average length of time for the first, second, third, fourth, and fifth instar nymphs differed from that recorded by Sudhakaran and Muraleedharan (2006), who documented that the first, second, third, fourth, and fifth nymphal instars lasted 3.8, 3.4, 2.8, 2.8, 3.0 days, respectively. In this study, the fifth instar nymph took a longer time to emerge as an adult. This result is in accordance with previous studies by Thube et al. (2019). A longer duration for the fifth instar nymph may be required to prepare to undergo the precious moult to emerge adult (Thube et al. 2019).

The use of antennae length measurement in the determination of nymphal instars in *Helopeltis* has been reported by previous studies (Roy et al. 2015; Serrana et al. 2021). The measurement values of this study differed from the data of Sudhakaran & Muraleedharan (2006). It might be due to the differences in experimental conditions such as temperature, relative humidity, and tea variety for their diet. Changes in the antenna length are due to the moulting process or ecdysis (Ghafoor 2011; Rostaee et al. 2021). As a result, *H. theivora* had five nymphal instars, which is in line with previous findings by Antony et al. (2018), and Roy et al. (2015) on tea, Rustam et al. (2014) on acasia, Srikumar & Bhat (2012) on cashew, Srikumar et al. (2016) on swamp weed, and Thube et al. (2019) on cocoa. On the other hand, four nymphal instars of *H. theivora* were also reported by Kalita et al. (2018) and Tea Research Association (2010) on red cherry pepper and tea, respectively.

The result is in accordance with the study of (Roy et al. 2015; Sudhakaran & Muraleedharan 2006) in the case of male longevity but differs in female longevity. They recorded that the average longevity of males was 28 days, whereas females lived for 48 days in tea. The fecundity of *H. theivora* in the present study was low compared to the reports by Sudhakaran & Muraleedharan (2006) from South India, who noted that as many as 325 eggs were laid by a female. However, the present finding recorded a maximum of 237 eggs, which is higher than the findings by Roy et al. (2009a) on tea. The sex ratio of male to female during one generation was 1:1.09 based on the number of emerged males and females, which is almost identical to the research by Srikumar (2013) and Sudhakaran and Muraleedharan (2006) who found that male to female ratio differ between 1:1.07 and 1:1.17 for cashew and tea, respectively. On the other hand, a higher sex ratio (male: female) of 1:1.65 was reported by Thube et al. (2019) on cocoa, and that of 1:1.43 was reported by Rustam et al. (2014) on acasia. Agro-climatic conditions, host plants, location, and elevation all affect the development of *H. theivora* (Roy et al. 2015). The life cycle of *H. theivora* varies in different countries. It was recorded for 21-35 days in Malaysia on cocoa (Bakar et al. 2017) and 20 days in South India (Antony et al. 2018). In comparison, it takes 23.5 days in India for cocoa (Thube et al. 2019) and 16-26 days in Indonesia for acasia (Rustam et al. 2014). Increases in temperature and humidity will decrease the life cycle duration and enhance fecundity (Rostaee et al. 2021; Roy et al. 2009a). Pazyuk & Reznik (2016) observed that the short-day length (10 hr) resulted in 1–2-day prolongation of nymphal development and a longer maturation time of *Macrolophus pygmaeus* (Hemiptera, Miridae) than the long-day length (16 hr). Hegazi et al. (2021) reported that day length may influence insects' basic physiological processes, such as metabolic rates and functions of the nervous and endocrine systems.

Morphological Description and Morphometric Measurements of *H. theivora* at Every Stage

The length and width of eggs measured in this study resemble that of Roy et al. (2015), who reported the range of egg length was 0.8-1.0 mm. The body length of different nymphal instars is almost similar to Roy et al. (2015) and Sudhakaran and Muraleedharan (2006). The female was larger than the males as females can produce eggs or offspring (Stillwell et al. 2014).

The colour description of the nymphal instars, except the second and third instars, is in line with Sudhakaran and Muraleedharan (2006), who stated that the first, second, third, fourth, and fifth instars were light orange, deep orange, reddish green, greenish yellow and green respectively. On the other hand, the colour description of the present study was different from Rustam et al. (2014), who described that the first instar was brownish, the second instar was brighter than the previous one, the third instar was reddish green, the fourth instar was light green, and finally, the fifth instar was dark green. Kalita et al. (2018) reported that the nymphs were orange in colour, which is also different from the present study.

The colour variation in the adult population of *H. theivora* is also reported by Roy et al. (2015) and Sarmah and Bandyopadhyay (2009). Based on head and pronotum, Roy et al. (2015) described three colour variants in the *H. theivora* population collected from Vietnam, South India, and Assam. Roy et al. (2009b) observed nine colour variants in the *H. theivora* population from the Sub-Himalayan Dooars tea plantation, three recorded in males and six in females. Sarmah & Bandyopadhyay (2009), on the other hand, described four types of variation in the colour of the female pronotum of *H. theivora* such as yellow, reddish-brown, yellowish-brown, and light brown from Assam tea plantation with the highest percentage of reddish-brown. In contrast, Kalita et al. (2018) reported that the pronotal region of male insects was black, and the female had a distinct orange colour. In the same and different cropping seasons, colour variation in adult tea mosquito bugs has been reported from the tea plantation by Roy et al. (2015). The colour variation of nymphal instars and adults is due to climatic conditions, host, and variety of tea plants (Kalita et al. 2018; Roy et al. 2015; Rustam et al. 2014). The same species' colour variation is sometimes considered a biotype (Rustam et al. 2014; Sarmah & Bandyopadhyay 2009).

CONCLUSION

Helopeltis theivora underwent egg, nymph, and adult stages in its life cycle. The eggs had an incubation period of 8.40±0.15 days. *Helopeltis theivora* population was identified to undergo five nymphal instars by referring to the graph of daily changes in antennal length. Thus, the length of the antennae is more suitable as an indicator in the determination of nymphal instars compared to body length. The nymphs of *H. theivora* were differentiated based on antennae length, body length, and colour variation. The average nymphal development period was recorded as 15.20±0.96 days. Males and females of *H. theivora* were distinguishable by their body size. The head was black with a pale stripe laterally. The pronotal collar was bicoloured in both sexes. In the case of males, most of the posterior part of the pronotum was yellowish brown, whereas in females, it was broadly yellow. The abdomen was green in both female and male bugs. Adult females and males had a mean lifespan of 37.23±1.65 days and 29.11±0.92 days, respectively. On average, each female produced 156.47±16.33 eggs with the sex ratio of male to female 1:1.09. The findings of the present study could be useful to formulate suitable management approaches for this pest. Since eggs are embedded within plant tissues, direct control is often ineffective. However, nymphs and adults, which actively feed on tender plant parts, are more accessible targets for pest management strategies. Thus, life cycle study of *H. theivora* allows for timely, targeted, and sustainable control measures that reduce economic losses, improve crop quality, and promote eco-friendly pest management practices of this pest in the tea plantation of Malaysia.

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AUTHORS DECLARATION

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Conflict of Interest
The authors declare no conflict of interest.

Ethics Declarations
This research does not include any ethical issues.

Data Availability Statement
It is a part of PhD study and the data are currently available in the thesis of Shovon Kumar Paul (2024).

Authors' Contributions
Shovon Kumar Paul (SKP) and Nur Azura Adam (NAA) originated this research and executed experiments; SKP, NAA, Syari Jamian (SJ), and Anis Syahirah Mokhtar (ASM) involved in data interpretation; SKP wrote the paper while NAA, SJ, and ASM provided the necessary corrections. The manuscript has been read and approved by all authors.

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