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Full Length Research Paper

Laboratory studies of the biology of Helopeltis schoutedeni Reuter (*Hemiptera: Miridae*), a major sucking pest of cashew (*Anacardium occidentale Linn.*)

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Cashew (Anacardium occidentale Linn.) has become a very important non-traditional tree crop in Ghana. Several insect pests, however, have been recorded on cashew and prominent among which is the cashew mosquito, Helopeltis schoutedeni Reuter (Hemiptera: Miridae). The biology of H. schoutedeni was studied in the laboratory at 24.9 to 33°C and 72.4 to 88.5% Relative humidity (R.H.). Females pre-dominated males in the ratio of 0.7:0.3. The mean pre-oviposition, oviposition and post-oviposition periods were 3.6 ± 0.1, 10.2 ± 0.6 and 6.6 ± 0.4 days, respectively. The life cycle from egg to adult emergence lasted 24.0 days. There were five nymphal stages and the mean durations ranged from 2.7 ± 0.10 days for the 3rd instar to 3.5 ± 0.10 days for the 4th instar. The incubation averaged 8.2 ± 0.05 days. The mean pre-copulation period was 2.6 ± 0.1 days. Peak copulation activity occurred between 1400 H and 1700 H. Copulation did not take place until both sexes were at least 2 days old. Males older than three days also did not mate, no matter the age of their companion.

Key words: Biology, Helopeltis schoutedeni, cashew, Ghana.

INTRODUCTION

In Ghana, cashew, Anacardium occidentale L. is a non-traditional crop, but it has become a crop of considerable economic importance. It is cultivated mostly in the drier parts of Guinea savanna, forest-savanna transition and coastal savanna zones, where the soils are considered unsuitable for most staple crops such as rice and maize (Anon, 2005).

Recent surveys in Ghana have shown that several old cashew plantations exist. The Government of Ghana, through Ministry of Food and Agriculture (MOFA) in collaboration with Cocoa Research Institute of Ghana (CRIG), has embarked on massive rehabilitation of the existing plantations, as well as establishing new ones, through the provision of technical support and farming incentives to farmers. The primary objectives are to increase cashew production and to export processed nuts. Government support has resulted in a significant expansion of hectarage under cashew cultivation from 18,000 to 52,000 ha and a corresponding rise in nut yield from 3,600 MT to 26,000 MT between 2000 and 2004 (Anon, 2005).

Several insect pests have been recorded on cashew in Ghana (Boakye, 1995; Yidana et al., 2004), prominent among which is the cashew mosquito, Helopeltis schoutedeni Reuter (Hemiptera: Miridae). The insect is prevalent in all the cashew growing areas in Ghana (Boakye, 1995; Yidana, et al., 2004). Both nymphs and adults feed on tender succulent shoots, inflorescences, immature nuts and the apples. Damage by *H. schoutedeni* is characterized by wilting and withering of the tender shoots and inflorescences (Anon, 2002; Boakye, 1995; Yidana et al., 2004). Earlier surveys conducted in 13 cashew growing communities in the Northern, Brong-Ahafo and Eastern regions of Ghana revealed *H. schoutedeni* to be most prevalent and destructive. It also recorded relatively larger numbers in all the localities than the other potential sap sucking pests found during the survey. Apart from the direct negative

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impact of the insect pests on cashew, the spread of diseases such as inflorescence blight caused by the fungi Gloesporium mangiferae and Phomopsis anacardii are known to be enhanced by Helopeltis spp. (Nambiar et al., 1990; Rao et al., 1993).

Despite the economic importance of H. schoutedeni, there is no comprehensive information on its biology in Ghana. The most recent literature on H. schoutedeni was produced by the Ministry of Agriculture and Food Security of Tanzania (Anon, 2002). This study, therefore, sought to determine the biology of the pest, the knowledge of which will be useful for the sustenance of its colony in the laboratory for further studies, and form the basis for developing effective control measures against it in the field.

MATERIALS AND METHODS

Raising and maintaining laboratory populations of Helopeltis schoutedeni

Fifty 4^{th} and 5^{th} instar nymphs were collected from cashew plantations at Somanya (longitude $0^{o}4$'E and latitude $6^{o}10$'N) in the Eastern Region of Ghana and taken to a laboratory at the Cocoa Research Institute of Ghana (CRIG) at Tafo, about 50 km from Somanya. They were kept in groups of five in hurricane lamp glass chimneys (top internal diameter of 12.5 cm, base internal diameter of 9.0 cm, and 18.0 cm high), placed upright in a 12 cm Petri dish lined with filter paper and the open top covered with nylon mesh to allow for aeration. The nymphs were supplied with fresh flush shoots of cashew daily and observed regularly till adult emergence.

Twelve hours after adult emergence, they were sexed relying on size difference, shape of abdomen and presence of ovipositor on the 6^{th} abdominal segment of the females. Five males and five females were then kept in transparent nylon mesh cages with wooden frame, measuring 60 x 45 x 45 cm, and fed with about 3-month-old potted cashew seedling which also served as oviposition substrate.

The seedlings were watered daily with 30 ml of water to avoid wilting. A new seedling was offered daily as the previous seedling was removed to examine for eggs. The presence of a pair of fine terminal egg filaments (small thread-like chorionic hairs) projecting from the surface of the plant tissue was indicative of the presence of eggs embedded in the bark. The seedling was labelled with oviposition date and number of eggs laid on it, and observed daily for hatching. Three days after oviposition, a fresh seedling was placed close to the one bearing the eggs such that a leaf of one touched that of the other. This enabled newly hatched nymphs to move onto the fresh seedling to feed. Any newly emerged nymph that remained on the old seedling was transferred to the fresh seedling by means of a fine camel hairbrush. Eleven days after oviposition, when all viable eggs were expected to have hatched, the old seedlings were discarded. The resultant adults and their progenies were used to conduct subsequent experiments. Temperature and humidity in the laboratory ranged from 24.9 to 33^{o}C and 72.4 to 88.5%, respectively.

Duration of the developmental stages of H. schoutedeni

Sixty-six eggs deposited in the epidermal tissues of flushing shoots of ten 3-month-old cashew seedlings were monitored at 24 h intervals until they hatched. The duration and the percentage of eggs that hatched were recorded. The trial was replicated three times. From 0 to 12 h after hatching, all the first instar nymphs (57 per replicate) were transferred singly into rearing chambers, using a camel hairbrush. The cut ends of the shoots were kept moist by placing them on water-soaked absorbent cotton wool in 20 ml vials. The nymphs were observed at 24 h intervals for the presence of cast skins, which indicated moulting. The cast skins were removed. The duration of the various instars and their percentage survivals were recorded.

After moulting into adults, the longevity of male and female bugs was determined. The durations of the pre-oviposition, oviposition and post- oviposition periods were recorded and the total number of eggs laid per female bug also recorded.

The morphology of the various developmental stages was described after sacrificing samples with ethyl acetate and immediately examining them with Alpha Stereo Expanded Pupil Microscope, manufactured by Mount Laboratories, UK. Fifty each of 1^{st} to 5^{th} instar nymphs and fifty each of 24 h old adult males and females were examined for the morphological characteristics. Five gravid females were also killed 24 hr after copulation, dissected and 60 eggs removed from the ovaries and preserved in 10% formaldehyde (Peterson, 1960). The eggs were fixed in a mixture of isopropyl alcohol, kerosene and glacial acetic acid in the ratio of 10:1:1 (v/v) respectively (Peterson, 1960). The size (length and breadth) of the eggs were measured using the stereomicroscope. The size of the nymphs and adults were also measured after fixing in a mixture of kerosene, 95% ethyl alcohol, glacial acetic acid and bioxane in the ratio of 10:1:1:1 (v/v) respectively (Peterson, 1960) . The body length was measured from the base of antenna to tip of abdomen excluding the ovipositor, while the body width was measured from the posterior section of the prothorax (Sundararaju and Babu, 2000).

Pre-oviposition, oviposition and post-oviposition periods of H. schoutedeni

The pre-oviposition, oviposition and post-oviposition periods of H. schoutedeni were determined, using 20 couples. Pairs of 12 h old adults were confined in glass rearing chambers and fed daily on tender shoots of cashew. Each of the 20 pairs was observed daily at 30 min intervals, from 0600 to 0600 h the next morning, to determine the periods of oviposition during a 24 h day. The trial was replicated three times.

Optimum age for copulating in H. schoutedeni

Pairs of laboratory-reared virgin male and female adults were kept for 24 h in glass rearing chambers in age combinations shown in Table 1, to determine the age at which H. schoutedeni copulates. Each age combination was replicated five times and observed at 30 min intervals for 24 h. Tender shoots of cashew were given as food. The couple was assumed to be copulating, if they faced opposite directions and the tips of their abdomen joined together (Plate 1).

Optimum time and duration of copulation in H. schoutedeni

Based on the results of optimum age for copulation, 20 pairs of 2-day-old male and 3-day-old female adults were placed in glass rearing chambers, with three replications, and observed hourly for 24 h. During the dark hours (1800 to 0500 H GMT), a torch, whose light intensity was reduced by covering with brown tissue paper (Khasimuddin, 1978) was used to monitor activity of the bugs. The time and duration of mating were recorded for each pair of the bugs and the means calculated.

Peak time of oviposition in H. schoutedeni

The time and peak of oviposition of *H. schoutedeni* were deter-

Table 1. Age combinations for the study of optimum time of copulation in H. schoutedeni

Sex	Age combination in days															
	1	1	1	1	2	2	2	2	3	3	3	3	4	4	4	4
	1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4

Plate 1. Adult male (arrowed) and female *H. schoutedeni* copulating.

determined using 30 pairs of 3-day-old male and female adults. Each pair was confined in a wooden/nylon mesh cage measuring, 60 x 45 x 45 cm, provided with a potted three-month-old cashew seedling to serve as feed and oviposition site. The seedlings were replaced with fresh ones at hourly intervals for 24 h and the previous seedlings carefully examined for eggs. The experiment was repeated three times.

Pattern of adult emergence in *H. schoutedeni*

Three hundred 5th instar nymphs of the same age were confined singly in petri dishes and kept under natural light regimes in the laboratory. From the 1st day after entering the 5th instar stage, observations were made at 24-hourly intervals to ascertain the numbers of male and female adults emerging per day. The trial was repeated once.

Time of feeding for adult *H. schoutedeni*

Sixty pairs (male and female) of one-day- old adult H. schoutedeni were confined in a glass-rearing chamber and provided with fresh flushing shoots of cashew as food source. Hourly observations for feeding were made for 24 h, beginning from 0600 H. Between the hours of 1800 and 0600 H, a subdued torchlight was used for the observation. A bug was deemed to be feeding if its stylet was inserted into the epidermal tissues of the flushing shoots. Similarly, 30 each of one-day-old of 1st to 5th instar nymphs of H. schoutedeni were individually kept in a glass-rearing chamber and observed for feeding. The experiment was repeated three times for each stage of the bug.

RESULTS

Eggs

The two unequal, creamy white, filaments arose from the anterior end of the egg, usually 0.4 - 0.5 mm long, with an average of 0.4 ± 0.05. Newly laid eggs of *H. schoutedeni* were pale white in colour but later changed to pinkish red. Eggs were ovo-elongate, slightly tapering apically and laterally compressed. H. schoutedeni mostly inserted its eggs into tender epidermal tissues of shoots, veins and midribs of leaves and leaf petioles of cashew. Eggs were laid singly or in small batches of 2 to 4. The eggs measured an average of 1.0 ± 0.05 mm long and 0.5 ± 0 mm wide (Table 2). The eggs had a mean developmental time of 8.2 ± 0.05 days (Table 3).

Nymphs

Measurements of the length and breadth of the various nymphal instars are presented in Table 2. There were five nymphal instars whose mean developmental periods are shown in Table 3. The average nymphal period from 1st to 5th was 15.6 days (range 13 - 18 days). The 1st instar nymph had light orange abdomen, head, antennae and legs. They possessed long legs with two-segmented tarsi. Legs, antennae and thoracic segments were covered with numerous setae, which arise from tubercles. The 1st nymphs were very active and moved about briskly. The average length and breadth the 1st nymphs were 1.5 ± 0.2 mm and 0.5 ± 0 mm, respectively (Table 2) with an average developmental period of 2.8 ± 0.03 days (Table 3).

The 2nd instar had deep orange abdomen, a prominent head, antennae and legs with a dull creamy scutellar horn, which was absent in the 1st nymph. Its mean size was 3.1 ± 0.2 mm long and 0.6 ± 0.05 mm wide (Table 2), with a mean developmental period of 3.5 ± 0.10 days (Table 3).

The 3rd instar nymphs had brownish orange head, ante-nnae and legs with an 8-segmented abdomen. The abdo-minal terga were dull creamy in colour. The wing rudi-ments were visible in the 3rd nymph. The scutellar horn was erect and tapering, with the apex swollen and funnel shaped.

Table 2. Size of laboratory bred H. schoutedeni.

Developmental stage	Body length (mm) (mean ± S.E)	Body width (mm) (mean ± S.E)
Egg	1.0 ± 0.05	0.5±0
1st instar	1.5 ± 0.2	0.5±0
2nd instar	3.1 ± 0.2	0.6 ± 0.05
3rd instar	5.6 ± 0.1	0.8 ± 0.04
4th instar	8.2 ± 0.4	1.0 ± 0.05
5th instar	10.2 ± 0.2	1.4 ± 0.05
Adult Male	11.3 ± 0.2	1.6 ± 0.1
Adult Female	12.2 ± 0.1	2.4 ± 0.1

Table 3. Life cycle of *H. schoutedeni* in the laboratory, CRIG, Tafo, Ghana; September 2004 to February, 2005.

Developing stages	Sample size	Percentage survival	% Surviving from previous stage	Duration in days (mean ± S.E)
Egg	198	100	-	8.2 ± 0.05
1st instar	171	86.4	86.4	2.8 ± 0.03
2nd instar	129	65.1	75.4	3.5 ± 0.10
3rd instar	120	60.6	93.0	2.7 ± 0.10
4th instar	14	57.6	95.0	3.5 ± 0.06
5th instar	114	57.6	100	3.3 ± 0.05
Adult	108	54.5	94.7	

a. b.

Plate 2. Adult *H. schoutedeni* reared on cashew flush (a = Male; b = Female)

The 4th instar had brownish orange head and legs. The legs, antennae and thoracic segments still had numerous setae. The wing pads were more prominent and the scutellar horn remained erect, tapering, with the apex swollen and funnel shaped.

The deep brown orange colour of the head and antennae were retained in the 5th instar nymph. The wing pads progressed beyond the scutellar horn, which was still erect, tapering with swollen apex and funnel shaped. The sizes of the various nymphal stages are indicated in Table 2.

Adult

On emergence, the adults were light orange, but turned brownish orange after about 30 min. The dorsum of the thorax became reddish in both sexes and the tergum of the abdomen turned dull white. A third tarsomere appeared in both sexes of the adults. The adult male was relatively smaller (Table 2 and Plate 2a) than the female and was shorter lived, with a mean longevity of 19.0 ± 2.5 days. The female (Plate 2b), had a mean longevity of 22.9 ± 0.9 days with average length and breadth of 12.2 ± 0.1 mm and 2.4 ± 0.1 mm, respectively. The ovipositor of the female was located on the ventral portion of the sixth abdominal segment. The scutellar horn in the adult was brownish orange. The antenna, as in the nymphs, was four-segmented, with the basal segment stouter and the terminal segment having numerous short hairs. The dorsum of the abdominal segments was deep orange in colour. The hemelytra overlapped as in a typical Hemiptera and covered the entire abdomen, with the distal end showing a triangular brownish black colouration. The femur had irregular deep brownish patches while the entire tibia was brownish black.

In this study, which was carried out in the laboratory, females predominated and the ratio of females to males was 0.7; 0.3. (approximately 2:1) (Table 4) . The entire duration of development of H. schoutedeni, from egg to adult on cashew took 24.0 days at the ambient tempera-

Table 4. Adult sex ratio, longevity and mean number of eggs of *H. schoutedeni* in the laboratory, CRIG, Tafo, Ghana; September 2004 to February, 2005.

Sex ratio		Longevity (days)		Mean number of eggs (n = 33)
Male (n = 33)	Female (n = 75)	Male (n = 33)	Female (n = 75)	
0.3	0.7	19.0 ± 2.5	22.9 ± 0.9	49.5 ± 3.1

Table 5. Duration of various stages of adult *H. schoutedeni* fed on cashew flush shoots

Adult stage	Duration (Mean ± S.E)	Range
Pre-copulation	2.6 ± 0.1 days	2 – 3 days
Copulation	2 h 48 min	1 – 4 h
Pre-oviposition	3.6 ± 0.1 days	3 – 4 days
Oviposition	10.2 ± 0.6 days	6 – 14 days
Post-oviposition	6.6 ± 0.4 days	4 – 7 days

ture range of 24.9 – 33^{o}C and relative humidity of 72.4 - 88.5%.

The durations of pre-copulation, copulation, pre-oviposition, oviposition and post-oviposition of male and female *H. schoutedeni* fed on tender shoots of cashew are presented in Table 5. The bugs were fully matured to copulate after 2 days but did not oviposit until about four days after copulation which could last nearly 3 h (Table 5).

Pattern of emergence in male and female *H. schoutedeni* is presented in Figure 1. Female *H. schoutedeni* bugs emerged 1 - 2 days earlier than the males. In the female bugs, emergence started two days after entering the 5th instar nymphal stage. Adults emergence reached a peak on the 3rd day. In the male, emergence began four days after entering the 5th instar nymphal stage. There was a sharp decline in numbers emerging the next day.

As shown in Figure 2, feeding started at between 1500 and 1600 H for the adult males and between 1700 and 1800 H for female adults throughout the period of observation. Percentage of male bugs feeding was higher than females. In both sexes the pattern of feeding appeared to be bimodal with a smaller peak occurring between 0300 and 0700 H. the major peaks of feeding were recorded between 1500 and 2100 H. Both males and females ceased to feed after 2100 H and resumed at 0300 H. The feeding patterns of the various nymphal stages were similar to the adults with a bimodal pattern except that percentage nymphs feeding increased with age of the nymphs.

Oviposition in *H. schoutedeni* occurred from 2100 to 2200 H but not during daylight hours (Figure 3), while copulation occur between 1000 H and 0200 H peaking between the day time period of 1400 to 1800 H (Figure 4)

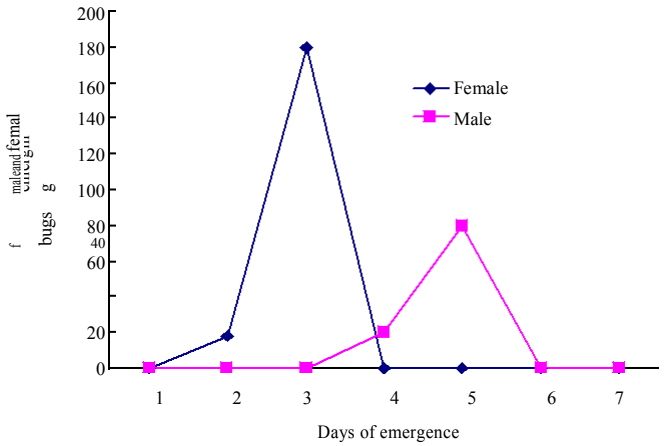

Figure 1. Pattern of emergence of adult male and female *H. schoutedeni* from 5th instar nymphs of the same age.

The age at which copulation occurs in H. schoutedeni is presented in Table 6. Copulation occurred more frequently between 2-day-old and 3-day-old adults and only rarely between 3-day-old males and 4-day-old females. Copulation did not take place until both sexes were at least 2 days old. Males older than three days also did not mate, no matter the age of their companion.

DISCUSSION

Helopeltis anacardii and *Helopeltis schoutedeni* have been reported in Tanzania as pests of cashew, with little emphasis on the life cycle of *H. anacardii*, but no information on the biology of the *H. schoutedeni* (Anon, 2002). The present study has shown that H. schoutedeni inserts its eggs in plant tissue singly or in small groups, usually with the filaments exposed. This is similar to the findings of Ambika and Abraham (1979) and Tan (1974). The eggs were inserted into epidermal tissues of the tender shoots, veins and midribs of leaves and leaf petioles of cashew. Pillai et al. (1976) investigating on various species of Helopeltis, showed that in a majority of them, the preferred oviposition sites depended on the host plant. Thus, *Helopeltis theivora* prefers cocoa pods (Miller, 1941) but will occasionally oviposit on young shoots. But on tea, this same species prefers new shoots to petioles and midribs of leaves (Das, 1984). Helopeltis

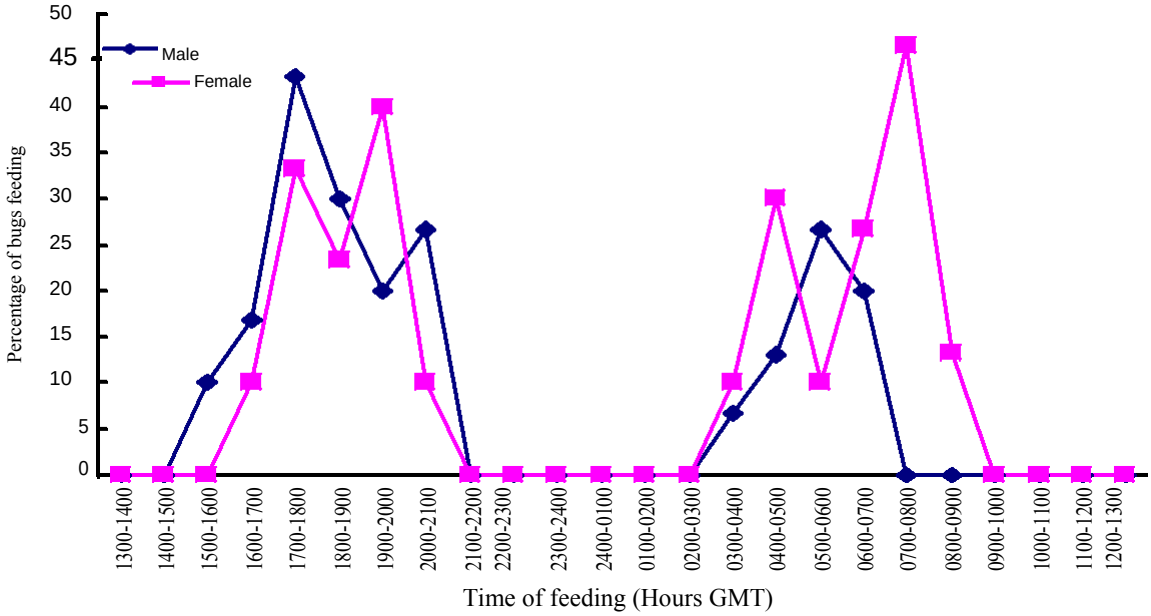

Figure 2. Feeding pattern of adult *H. schoutedeni* within a 24 h cycle.

Table 6. Copulation frequency for different age combinations of *H. schoutedeni*.

Age in days :	1:1	1:2	1:3	1:4	2:1	2:2	2:3	2:4	3:1	3:2	3:3	3:4	4:1	4:2	4:3	4:4
Percentage copulating	0	0	0	0	0	40	60	0	0	40	60	20	0	0	0	0

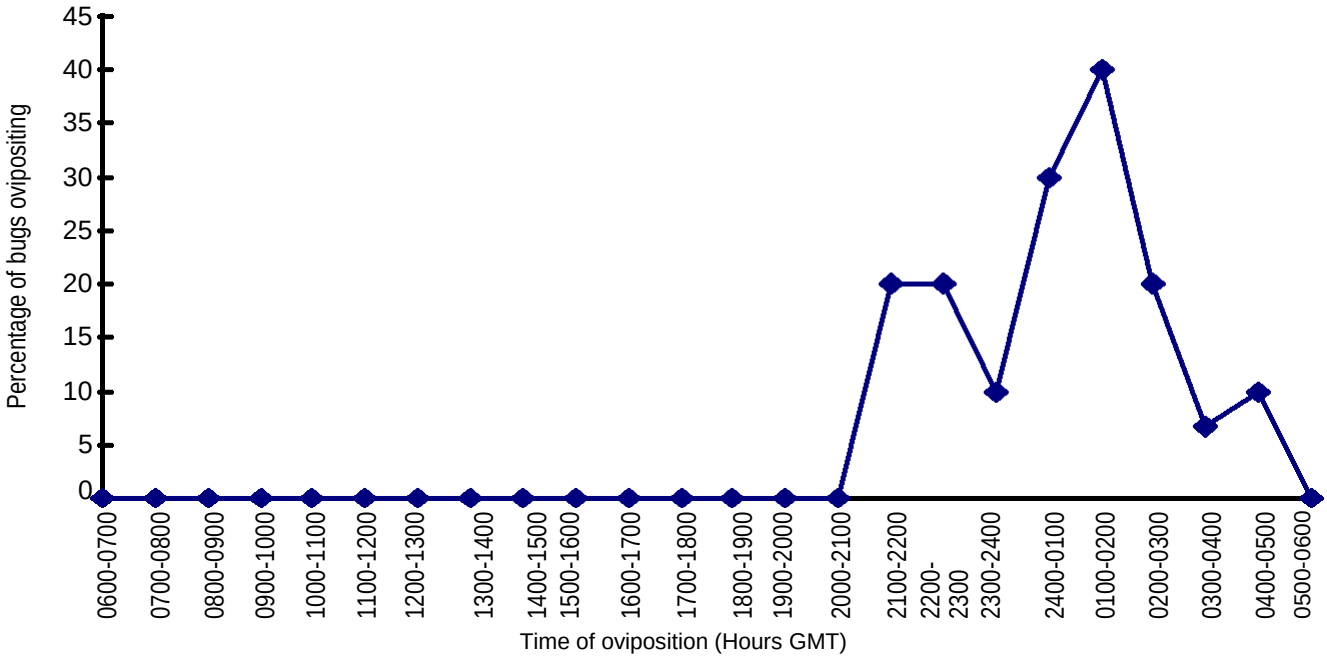

Figure 3. Pattern of oviposition in *H. schoutedeni* within a 24 h cycle.

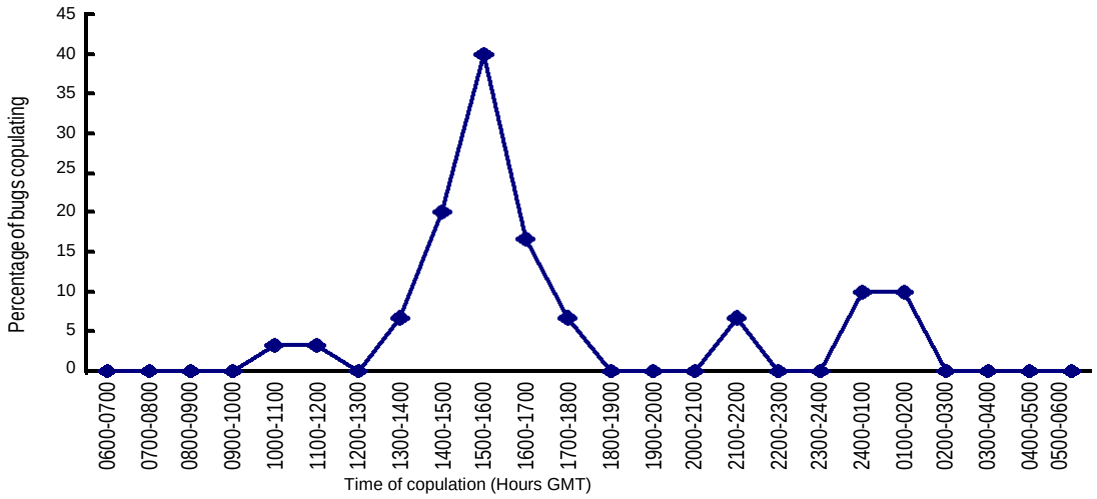

Figure 4. Percentage of bugs copulating at different periods of the day.

antonii lays its eggs primarily on the young shoots, inflorescence stalks and developing nuts of cashew, but will sometimes accept the petioles and ventral midribs of leaves (Ambika and Abraham, 1979).

The incubation period of the egg varies with locality and season, but it is generally in the range of 6 - 11 days (Stonedahl, 1991), although longer durations are observed occasionally, for example 20 - 27 days for H. theivora in northeast India and 13-16 days for winter populations of *H. bradyi* (Das, 1984) . H. schoutedeni exhibited a mean incubation period of 8.2 ± 0.05 days. The difference observed could be attributed to climatic factors and/or rearing conditions such as temperature and relative humidity prevailing at the location of the studies (Betrem, 1953).

The rate of nymphal development is affected by climatic conditions and quality of the food source (Betrem, 1953; Awang et al., 1988). In the present investigation, H. schoutedeni was raised on flushing shoots of cashew under ambient laboratory conditions and spent an average nymphal life span of 15.6 days (range 13 - 18 days). This finding compares with most reported nymphal life spans of *H. antonii* on cashew, which were in the range of 9 -19 days (Smith, 1973; Tan, 1974; Ambika and Abraham, 1979, Devasahayan, 1985). On the other hand, much longer periods have been reported for October-December populations of H. bradyi (27 - 43 days) and *H. cinchonae* (30 - 54 days) reared on tea in the Cameroon Highlands (Lever, 1949). December/January populations of H. theivora took 25 to 39 days on tea in north-east India (Das, 1984). The discovery of the suitable laboratory rearing conditions of the pest, H. schoutedeni (24.9 to 33^{o}C and 72.4 to 88.5%) makes it easier for future breeding and maintenance of populations of the insect for further scientific studies.

Adult longevity has been reported to also vary with the rearing conditions (Stonedahl, 1991). The mean longevity of male and female H. schoutedeni reared on flushing shoots of cashew was found to be 19.0 ± 2.5 days and 22.9 ± 0.9 days, respectively. Tan (1974) recorded a mean adult longevity of 30 days for *H. theivora* on cocoa pods in West Malaysia. The same species was reported by Awang et al. (1988) to have a mean longevity of only six days when raised on cocoa shoots.

It was observed in the present work that the period of adult emergence in female *H. schoutedeni* occurred earlier than that of the males. Similar finding were reported in other insects by Banerjee (1967). It is possible that females emerge ahead of the males because of the need for post-emergence sexual maturation (Engelmann, 1970), which might probably take a slightly longer time in the females than in the males.

Many insects are known to copulate during certain periods of the day and H. schoutedeni is no exception (Ingoffo et al., 1963). Peak copulation in H. schoutedeni bugs took place towards the dusk. In 2-day and 3-day-old bugs copulation occurred between 1300 and 1800 H. Most of the oviposition activity H. schoutedeni occurred between 2100 and 0600 H and the distribution was mono-modal. In the present study, oviposition always appeared to begin after feeding had stopped. When flight pattern of H. schoutedeni is examined in relation to other behavioural activities, peak flight activity coincided with the first peak of feeding activity. During this period the bugs may be searching for food or leaving the food to settle elsewhere.

Maximum age of copulation was recorded in 2- day-old and 3- day-old bugs of both sexes. Both male and female adults inability to copulate soon or a day after emergence may be attributed to the late sexual maturation after emergence (Campion and Outran, 1967). Both sexes may require a post-emergence maturation period in order

to become sexually matured. During this time, the process of spermatophore and pheromone productions, pheromone release and copulation as well as the development of ova, are initiated and completed (Shorey, et al., 1968).

Conclusion

The study has shown that H. schoutedeni can be reared in the laboratory at the ambient temperature and relative humidity range of 24.9 to 33^{o}C and 72.4 to 88.5%, respectively. It is, therefore, highly possible to raise and maintain large populations of the insect for further studies. The study showed that the life cycle of H. schoutedeni from egg to adult emergence lasted 24.0 days with five nymphal stages. The mean pre-copulation period was 2.6 ± 0.1 days. Peak copulation activity occurred during the daylight hours between 1400 H and 1800 H. Copulation took place mostly between males that were 2 to 3 days old and females that were 3 days old. Copulation, however, did not take place until both sexes were at least 2 days old. Males older than three days did not mate, no matter the age of their companion.

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