

Developmental Biology of the Oil Palm Leaf Miner *Coelaenomenodera elaeidis* Mlk. (Coleoptera: Chrysomelidae) on an Artificial Diet

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Authors' contributions

This work was carried out in collaboration between all authors. Author AT designed the study, wrote the protocol and monitored the laboratory work. Authors AC and AV anchored the field study and gathered the initial data. Authors AV and IB managed the literature searches, produced the initial draft and interpreted the data. All authors read and approved the final manuscript.

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ABSTRACT

The oil palm leaf miner, *Coelaenomenodera elaeidis* is one of the most destructive pests of the oil palm in Nigeria. It was reared on an artificial diet through all its developmental stages in the entomology laboratory of the Nigerian Institute for Oil Palm Research between July and September 2011 with the objective of observing the developmental duration and survivorship without predation. Rearing temperatures fluctuated between 26°C–34°C with a mean of 29.51°C and relative humidity between 52% - 92% with a mean of 77.06%. Development time of *C. elaeidis* was 91±3.51 days on artificial diet. Body length from 1st instar to adult ranged from 1.94 - 8.01 mm; weights from 1st instar to adult ranged from 0.112-0.184 g, while leaf area tunneled from 1st instar to adult ranged from 1.58-8.51 cm. Between temperatures of 26°C±1.37 and 34°C±1.73 and relative humidity of 53%– 92%, 91 days were required to complete *C. elaeidis* development. This study is important in providing the developmental biology of the oil palm leaf miner using artificial diet throughout its life cycle.

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1. INTRODUCTION

The oil palm leaf miner *Coelaenomenodera elaeidis* (Coleoptera: Chrysomelidae) is a key insect pest of the oil palm in Nigeria. It is endemic in almost all oil palm growing areas of West Africa and is regarded as the most devastating pest of the oil palm. The larvae are the main cause of damage, especially during swarming when several individuals can be found mining into the epidermis of the leaflets, leading to direct destruction or desiccation of the leaf or reduced photosynthetic surfaces [1]. In severely affected plantations, the lower canopies of most palms appear scorched, grey-brown, with desiccated rolled-in leaflets. Later the withered laminae shatter, leaving the leaflets' midribs only. In visibly heavy outbreaks, control measures become necessary. The best recorded outbreak of the beetle in the country was at Oyo State, Nigeria in 1966 [2]. Both the adult and larval forms of the leaf miner cause damage to the palm [3]. Accounts of the incidence, life cycle and damage of this pest have been reported [4] and [5]. The developmental periods (in days) are: eggs, 20; larvae, 44; pupae, 12; adult to egg laying 18; total 94 days (about 3 months). The adult lives on the under-surface of the leaf for 3-4 months after egg laying. There are thus 3 to 4 generations of this pest in a year [5].

Cultural control by pruning and heaping of all affected leaves during the rains has been reported to be very effective [6]. Ultracide 40 E.C. at 1.5 litres/hectare using the tecnomat for tall palms has also been reported [7]. Extracts of a naturally occurring marine moluscicide, Evisec, effective in controlling *C. elaeidis* have been identified [8]. Another control measure includes application of Methidathion 40 EC at 600g a.i. per hectare at pockets of population build up.

The mean temperature for Nigeria is 27°C. Over the last few decades, there has been a general increase in temperature throughout Nigeria [9]. Nigeria has a wide diversity of ecological conditions ranging from arid desert to swamp land. Concomitant with this diversity is a range of climate change and variability impacts that could affect insect pest distribution and abundance across the oil palm belt. An understanding of both life cycles and seasonal cycles for a pest species is required for proper management [10].

Artificial diets have been used to rear numerous species of dipterans, lepidopterans and coleopterans have been successfully reared under controlled laboratory conditions [11].

This study seeks to rear the developmental stages of *C. elaeidis* with artificial diet.

2. MATERIALS AND METHODS

2.1 Study Site

The study site is located at the main station of the Nigerian Institute for Oil Palm Research (NIFOR). It lies 29 km North-West of Benin-City on the coordinates of latitude 6° 33' and longitude 50° 37'.

2.2 Weather Data

Temperature was measured with the maximum and minimum thermometer and relative humidity was recorded with a hygrometer in the NIFOR entomology laboratory. Temperature and relative humidity readings were recorded at 12h00 and 16h00 respectively throughout the developmental period of the *C. elaeidis*.

2.3 Field Collection

Sampling was conducted between 07h00 and 11h00 from study site. The site lies on the coordinates of latitude 6° 33' N and longitude 5° 37' E. The size is 2.95Ha consisting of 443 mature palms at 9m triangular spacing located at the main station of the Nigerian Institute for Oil Palm Research (NIFOR) near Benin, Edo State, Nigeria. The palms whose crown canopy had not formed a continuous layer and sunlight could still penetrate to the ground, were planted in the year 2000. Methods for insect collection were sweep netting and direct handpicking. The boundary row was skipped because of possible border effects. Systematic sampling was done at the base, middle and crown levels of trees. Adult *C. elaeidis* were collected from the field and introduced into 60 cm x 60 cm x 60 cm screen cages in the laboratory.

2.4 Laboratory Studies

The Leaf miner was cultured in the laboratory using artificial diets through out their

developmental phases. The cages were 60 x 60 x 60 cm metal frames and wire netting of 2 mm mesh to let in air and light for development. Cage stands were put in water to prevent ants from entering the cages. Three replicates each of rearing cages and containers were monitored in the laboratory. Temperature and humidity readings were collected daily through out the different life stages of the *C. elaeidis* (eggs, larvae, pupae and adult) kept in culture vials and cages. A Wild Heerbrugg M 3B Binocular Microscope equipped with a standard ocular micrometer and a Samsung S760, 7.2 Mega pixels were utilized. The Sartorius CP 22025 weighing balance was used to weigh the different life stages of the *C. elaeidis*.

Pruned, damaged and infested leaves were cut open and studied for presence of different life stages of *C. elaeidis*.

2.5 Artificial Diet

Fresh oil palm leaves were cut from the field, washed and oven dried at 70°C for seven days in the gallenkamp size 1 incubator. When dried, it was manually pounded in a wooden mortar to a powdery substrate, weighed (5 gm) and added to diet formulation. Pollen grains were collected from oil palm inflorescence, weighed and also added to the diet. Weights were measured using a Mettler P1210 weighing balance. Other components of the diet for *C. elaeidis* [12] were made up of: Water (500 ml), Agar (15 gm), Ascorbic acid (3 gm), Menthol paraben (1 gm), Sugar (10 gm), Soy flour (10 gm), Wheat flour (28 gm), Potassium hydroxide (128.54 gm), Acetic acid (5 ml), Vegetable oil (2 ml), Palm leaflet powder (20 gm), Pollen grain (2.5 gm), Formaldehyde (2 ml), Vitamin and salt mix (5 ml), Vitamin and mineral capsule (1). An electric blender was used to blend the feed and left to cool for one hour. Surface of the feed was lacerated. It was later stored in transparent plastic containers covered with serviette paper and lid. These cans were kept in plastic trays containing water to prevent ants from entering the containers.

2.6 Larval Rearing

Fresh leaflets were collected from the field and taken to the laboratory. The basal parts of the leaves were inserted in plastic containers with four holes for aeration containing water to maintain turgidity. The rearing cages and containers were kept in the laboratory where

daily temperature and relative humidity records were collected. Prior to infestation, the glass vials were sterilized in a steamer and oven dried. The diet was poured in while hot. The glass vials were half filled and *C. elaeidis* larvae were put in the glass vial at the rate of one per glass vial. Infestation took place after the diet had cooled and its surface scarified to ease larval penetration. All infestation was done under a laminar flow station which ensured sterilized air. Cotton plugs were used to cover the vials to prevent humidity build-up and prevent larval escape.

3. RESULTS AND DISCUSSION

The oil palm plays a dominant role in supporting rural livelihoods and economic growth over most of Southern Nigeria [13].

3.1 Air Temperature

Each specie of insect has a range of temperature within which it can survive. This range is referred to as the tolerable zone [14]. Most insects have an upper temperature tolerance between 40 and 50°C, and no known insect can survive temperatures in excess of 63°C [15].

As expected, temperature variation in the morning followed the air minimal while that in the afternoon followed the air maxima. Mean temperature for 12h00 was 27.47°C and 31.54°C at 16h00. The observed temporal fluctuation can be attributed to daily weather condition. There was no statistically significant difference between temperatures at 12h00 and 16h00. During the experimental period, average minimum temperature values were 26°C±1.37 and maximum temperature values were 34°C±1.73.

3.2 Relative Humidity

Humidity is the invisible water content of the air. Mean relative humidity was 76.66% at 12h00 and 77.44% at 16h00. There was no statistically significant difference between the relative humidity at 12h00 and 16h00. The month of August had the lowest value of 53% while the months of July and September recorded maximum values of 92%.

3.3 Development of the *C. elaeidis*

Table 1 shows summary statistics of body length (mm), weight (g) and leaf damage (cm) of the leaf miner during their developmental period. The

body length from 1st instar to adult ranged from 1.94 ± 0.184 to 6.12 ± 0.162 (M); 8.01 ± 0.074 (F) respectively. The weights from 1st instar to adult ranged from 0.112 ± 0.006 to 0.184 ± 0.0069 respectively. The leaf damage from 1st instar to adult ranged from 1.58 ± 0.078 to 8.51 ± 0.074 respectively. In this study, the length of leaflet damage was indicative of leaf miner developmental stage which provides information for its timely control.

Fresh oil palms were added to the diet to improve its quality. The oil palm (*Elaeis guineensis*) leaves has 8% higher total polyphenols content than green tea extract and contains epigallocatechin, catechin, epicatechin, epigallocatechin gallate, epicatechin gallate amongst their various flavonoids glucosides [16].

Development of the *C. elaeidis* was completed within a total period of 91 days (Table 2).

3.4 Description of the Developmental Stages of *C. elaeidis*

3.4.1 Embryonic development

The *C. elaeidis* egg is creamy white and embryonic development like in most insects proceeds within the egg after it has been laid or deposited. It lays its eggs singly. The egg is ovoid and surrounded by a protective chorion, which gives it a characteristic shape and surface appearance. The hatching process begins immediately on completion of embryonic development, thereby taking advantage of favorable environmental conditions. Plate 1 shows eggs of *C. elaeidis* on leaflet.

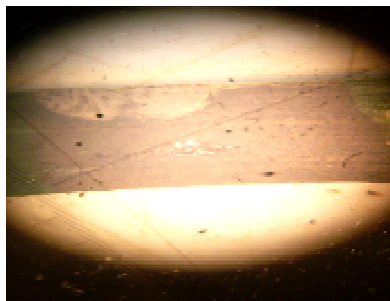


Plate 1. Eggs of *C. elaeidis* on mined leaflet groove (X100)

3.4.2 Larval stages

The larval stage was characterized by a rapid feeding habit. Leaf miner damage is most critical

at this phase. Increase in size was most pronounced with resultant molting. They are light brown with increasing darkening pigmentation until the 4th instar stage. They lack compound eyes, possess reduced antennae and lack external evidence of wing formation. However, they possess a well developed head and mouthparts adapted for biting. They feed well on artificial diet in the laboratory. However, they were found to thrive in the mined crevices of damaged and infested oil palm leaves. Plate 2 show leaflets indicating egg laying and hatching.



Plate 2. Leaflets removed from rearing cage indicating egg laying and hatching in the leaflets (X100)

In the laboratory, larvae prefer to stay directly in the diet. They usually do the following: they could burrow directly into the feed; they could have their mouth parts in the feed and their abdominal section extended outside the feed; and they could also burrow to the bottom of the rearing cage along lacerated parts. The larvae do not usually move far from their initial feeding position and tend to stay apart from one another.

3.4.2.1 First instar larva

The first instar larva is creamy yellow (Plate 3).



Plate 3. First instar larva (X100)

3.4.2.2 Second instar larva

The second instar larva is creamy yellow with the beginning of darkening at the abdomen (Plate 4).



Plate 4. Second instar larva (X100)

3.4.2.3 Third instar larva

The third instar larva is light brown (Plate 5).



Plate 5. Third instar larva (X100)

3.4.2.4 Fourth instar larva

The fourth instar larva is light brown with a better developed head region (Plate 6).



Plate 6. Fourth instar larva (X100)

3.4.3 Pupa stage

The pupa is dark brown and covered by a thin, soft cuticle. There is no feeding and it appears inactive. The female pupa was observed to be bigger (Plate 7).



Plate 7. Male and Female pupa of *C. elaeidis* (X100) (Female is bigger: right arrow)

3.4.4 Adult stage

Adults emerge from the pupa stage by pulling itself out from their old skins. It was observed that emerging pupa to adult move away from light source. Observation showed that the pupation process from pupa to adult is from the head and wings, and extending to other parts (Plates 8, 9 and 10).



Plate 8. Pupa emerging to adult (X100)

The adults are reddish brown and the new adult is soft and initially immobile until after a short period. The female adult was also observed to be bigger with a swollen abdomen and a thicker antenna. Plate 11 shows the adult.

Table 1. Summary statistics of body length, weight and leaf damage of the leaf miner

Parameters	Body length (mm)	Weight (g)	Leaf damage (cm)
1 st Instar	1.94±0.184 (2.2 – 1.6)	0.112±0.006 (1.2 – 0.1)	1.58±0.078 (1.5 – 1.7)
2 nd Instar	6.35±0.58 (7.0 – 5.0)	0.137±0.007 (0.15 0.13)	2.85±0.337 (2.5 – 3.5)
3 rd Instar	8.65±0.412 (8.0 – 9.0)	0.146±0.0052 (0.15 – 0.14)	3.35±0.242 (3.0 – 3.5)
4 th Instar	8.64±0.664 (9.0 – 11.5)	0.16±0.0067 (0.15 – 0.17)	6.0±0.624 (5.0 – 7.0)
Pupa	8.64±0.664 (8.0 – 10.0)	0.17±0.0074 (0.18 – 0.16)	7.81±0.30 (7.0 – 8.0)
Adult	6.12±0.162 (6.5 – 6.0) (M) 8.01±0.074 (8.1 – 7.9) (F)	0.184±0.0069 (0.17 – 0.19)	8.51±0.074 (8.4 – 8.6)



Plate 9. Pupa prior to adult emergence (X100) (ventral surface)



Plate 10. Pupa prior to adult emergence (X100) (dorsal surface)

Their flights are of short duration and their darting movement makes it difficult to follow their course with the eye. They were usually less active with approaching higher temperatures in the afternoon. They were usually observed feeding next to each other. The adult *C. elaeidis* head is made up of heavy, muscular and circular reddish cranium with mouth parts adapted for

biting off leaf tissue and scraping leaf surface. A pair of compound eyes is present. The paired antenna length measured 0.6–0.9 mm. It's blackish in color with horizontal striations. The thorax is made up of rigid, hardened plates. The abdomen consists of 8–10 segments. Three pairs of legs are present measuring 1.0–1.3 mm. It has two pairs of wings present with a membranous system of veins running through out. Its length measures 1.8–2.2 mm.



Plate 11. Adult (X100)

Table 2. Duration (days) of developmental stages of *C. elaeidis*

Life stages	Duration of stage (days)
Egg	15
1 st Instar	16
2 nd Instar	13
3 rd Instar	10
4 th Instar	7
Pupa	13
Adult	17
Total	91

Adults are basically phytophagous feeding on oil palm leaves. They feed slowly, scraping and biting the leaf surface. They were observed to be feeding early in the morning at lower temperatures usually preferring the upper fronds. They tend to feed next to one another. Under the microscope, adult female was observed to have a swollen abdomen, thick antenna and ovipositor.

4. CONCLUSION

Artificial diets have been developed and proposed for the maintenance, and continuous rearing of economically important insects [15,17]. A proper understanding of the life cycle of the *C. elaeidis* is required for its adequate management. The use of ground fresh oil palm leaflets added a natural flavour and enhanced its value for the insects. The use of artificial diet is suitable for mass rearing of the *C. elaeidis*. The addition of ground fresh oil palm leaflets added a natural flavour and enhanced its value. 91±3.51 days were required to complete *C. elaeidis* development on artificial diet. Biological control involves intentional natural enemy manipulation to obtain reduction in pest status. The future scope going forward with this study is the use of artificial diet to rear parasitoids for control of the oil palm leaf miner.

This study is important in providing the developmental biology of the oil palm leaf miner using artificial diet throughout its life cycle.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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