



Research Article

STUDY OF DEVELOPMENT CYCLE OF *RECILIA MICA* KRAMER (HEMIPTERA: CICADELLIDAE), VECTOR OF BLAST DISEASE IN OIL PALM NURSERIES

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ABSTRACT

Recilia mica Kramer is the main vector of blast disease in oil palm nurseries in Côte d'Ivoire. This disease can destroy more than 50% of the plants. Work on this insect has been limited to the damage caused by the disease it transmits and to research on control methods. In order to achieve integrated pest management in the current context of sustainable agriculture, a knowledge of the biological cycle of this pest is necessary. The present work proposes to study the life cycle of *R. mica*. A rearing of this insect was carried out under semi-controlled conditions in cages on six refuge plants of this species, namely *Digitaria horizontalis*, *Pueraria phaseoloides*, *Panicum maximum*, *Eleusine indica*, *Cynodon dactylon* and *Sporobolus pyramidalis*. Thus, the average length of the development cycle from egg to adult is 40.45 ± 1.73 days. The average incubation time is 9.97 ± 0.04 days. The duration of the larval stage is 25 to 40 days. The results of this study will contribute to the development of an integrated control strategy against blast, a disease transmitted by *R. mica*, in oil palm nurseries.

Keywords: Life cycle, *Recilia mica*, Blast, Oil palm, Refuge plants.

INTRODUCTION

Recilia mica Kramer (Homoptera, Cicadellidae, Deltocephalinae) is the vector of blast disease in oil palm (*Elaeis guineensis*) nurseries. It manifests itself as a brown lesion at the base of the arrow, which becomes moist and then rots with a foul-smelling odour (Bachy, 1958). Seedlings in the 5 to 8 leaf stage are most susceptible to this disease. Blast can kill more than 50% of the plants if no precautions are taken. To this end, it is able of destroying the most beautiful nurseries in only one or two months. Adults of *R. mica* acquire the pathogen responsible for blast during sap ingestion when feeding (Julien *and al.*, 2015). After ingestion, the phytoplasma in the body of the leafhopper, enters the digestive tract. It crosses the intestinal barrier and then reaches the haemolymph, which ensures its migration to the salivary glands where it multiplies (Julien *and al.*, 2015). During a meal on an oil palm plant, the insect transmits the pathogen by injecting contaminated saliva (Saguez *and al.*, 2015). For the control

of this disease, two control methods have been proposed, i.e. cultural and chemical control. Chemical control was provided by aldicarb and then carbofuran. Despite their effectiveness, these products were withdrawn from the market because of their high toxicity. In a context of sustainable agriculture, the search for ecologically sound control methods is essential. This requires a good knowledge of the biology of this species. Therefore, the aim of this work is to study the life cycle of *R. mica*.

MATERIALS AND METHODS

Study site

Study was carried out at the Station of the National Center of Agronomic Research of La Mé (5° 26' Latitude North and 3° 50' Longitude West) from September to December 2019. The climatic characteristics at this period were 27.16°C as average temperature, with a relative humidity of 83.26%.

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Animal material

The animal material consisted mainly of the different stages of development of *Recilia mica*.

Vegetal material

The vegetal material consisted of oil palm plants *Elaeis guineensis* (≤ 8 months) from the *Tenera* C1001 variety, which are more sensitive to blast and refuge plants of *Recilia mica*.

Rearing of *R. mica*

A rearing of *R. mica* was carried out in semi-controlled conditions (in cages). For this purpose, cages consisting of a metal frame measuring 45 cm x 30 cm on the ground and 70 cm high were made. These cages were lined with a rigid white nylon mesh net 600 microns mesh. A small door of 30-cm in size was installed to make access easier. Each cage contained one refuge plant of *R. mica*, namely *Digitaria horizontalis*, *Pueraria phaseoloides*, *Panicum maximum*, *Eleusine indica*, *Cynodon dactylon*, *Sporobolus pyramidalis* (Anougba *et al.* 2020). Adults of *R. mica* were captured between 7-10 am and 4-6 pm in herbaceous vegetation using an 18x100 mm test tube closed with a cotton plug and/or a vacuum cleaner and mowing technique to be released in the different cages. In total 50 individuals of *R. mica* of which 25 males and 25 females were released into each cage. The study was repeated 3 times on each plant species. A total of 10 newly emerged adults (5 males and 5 females) obtained from the laboratory colony were transferred onto *Eleusine indica* to determine the pre-oviposition period. *Eleusine indica* was selected because it was readily available at the experimental site.

Determination of the different development stages of *R. mica*

From the day after the insects were introduced into the cages, a daily check was carried out. It consisted of observing the leaves of the refuge plants to look for eggs and larvae until the emergence of new adults.

Eggs and average incubation time

The eggs observed were described, marked and monitored until the first larva was observed. The date of hatching was recorded. The incubation period (D_i) is the period from egg laying (P) to egg hatching (O). It was calculated according to the following formula:

$$D_i = \frac{\sum t_j d_j}{\sum d_j} \quad t_j = O - P \text{ (Doh, 2018)}$$

With t_j : time from egg laying to egg hatching and d_j : number of eggs hatched.

Duration of life of the different larval stages

At hatch, the newly emerged early stages were reared three by three in cages containing the plants for feeding until adult emergence. Each larval stage was monitored daily until molting. This was confirmed by the presence of exuviae at successive stages. The molt dates were noted

and made it possible to calculate the lifespan of each stage of development. The average time from one larval stage (D_{lx}) to the next is the time between a moult corresponding to a given larval stage and the next moult. It has been calculated according to the following formula:

$$D_{lx} = \frac{\sum s_i n_i}{\sum n_i} \quad s_i: \text{Time between one moult and the next; } n_i: \text{number of x-stage larvae.}$$

Therefore, the average survival rate of the larval stages was calculated as follows (Doh, 2018):

$$\text{Average larval survival rate} = \frac{\sum a_i b_i}{\sum b_i} \times 100$$

a_i = (number of larvae of the next instar) / (number of larvae of the previous instar) and b_i : number of females.

Duration of the pre-oviposition period

The average duration of pre-oviposition (D_p), expressed in days, is the time between adult emergence (T_e) and first oviposition (P_p). It is obtained according to the following formula: $D_p = \frac{\sum t_i j_i}{\sum j_i}$ and $t_i = P_p - T_e$ t_i : Time taken from emergence to first egg laying; j_i : Number of females.

Duration of the development cycle

The average length of the development cycle (D_c) is the period between egg laying and adult emergence (period between the date of observation of the 1st egg and the date of observation of the 1st immature adult).

$$D_c = \frac{\sum p_i q_i}{q_i}$$

$p_i = D_i + D_{l1} + D_{l2} + D_{l3} + D_{l4} + D_{l5}$; q_i : adults effective (Doh, 2018)

Average number of eggs laid per female and adult lifespan

From the time of laying, the number of eggs laid per female was assessed on each plant species. All eggs observed on each plant were counted and the average number of eggs laid by each female was calculated according to the following formula:

$$\text{Average number of eggs} = \frac{(\text{Total number of eggs})}{(\text{Total number of females})}$$

Lifespan (D_v) of adults is the time elapsed between the date of emergence (D_e) of the imago and its death (D_m). $D_v = (\sum k_i l_i) / l_i$; $k_i = D_m - D_e$ and l_i : number of adults

Egg hatch rate

The number of eggs hatched on each plant was recorded and the average rate of eggs hatched was calculated according to the following formula:

$$\text{Average hatching rate} = \frac{(\sum T_e f_i)}{(\sum f_i)} \times 100$$

with

T_e = (number of eggs hatched) / (number of eggs laid) and f_i : number of females (Doh, 2018)

Sex-ratio

The sex ratio is the ratio of the number of males and females within a sexually reproduced species for a generation or in the offspring of an individual. It has been calculated for the descendants of each female according to the formula:

$$\text{Sex-ratio} = (\text{number of males}) / (\text{number of females})$$

Data Analysis

The mean durations of each stage of development, as well as the respective standard deviations, were calculated using Statistica 7.1 software. In case of significant differences

(threshold of 0.05), the Newman-Keuls test was used to identify the different homogeneous groups.

RESULTS AND DISCUSSION

Females of *R. mica* lay eggs under the epidermis and/or in the veins on the underside of young leaves using an ovipositor which distinguishes them from males (Figure 1). The eggs (about 1 mm) develop in the epidermis of the leaves and hatch a few days after oviposition to give successively five (05) larval instars (Figure 2). The incubation period of the eggs lasts from 9 to 13 days with an average of 9.97 ± 0.04 days (Table 1).

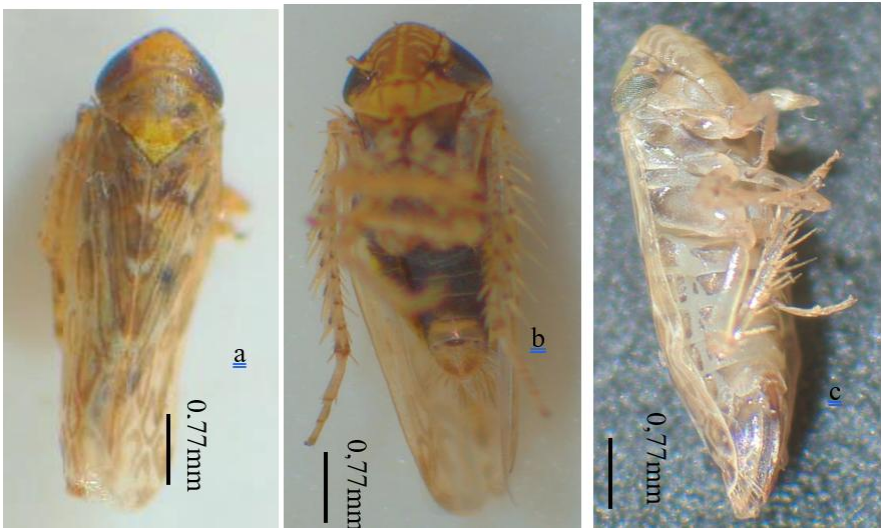


Figure 1. *Recilia mica*

a: Dorsal view b: Ventral view of male c: Ventral view of female

Table 1. Life cycle of *Recilia mica*.

Development stage	Minimum duration (Days)	Maximum duration (Days)	Mean ± Standard deviation (Days)
Pre-oviposition	3	8	4,66 ± 2,88
Egg	9	13	9,97 ± 0,04
1 st larva instar	3	7	4,40 ± 0,13
2 nd larva instar	3	5	3,84 ± 0,14
3 rd larva instar	3	5	4,1 ± 0,09
4 th larva instar	3	5	3,91 ± 0,07
5 th larva instar	4	5	4,41 ± 0,14
Adult lifespan	1	14	5,16 ± 0,31
Total	29	52	40,45 ± 1,73

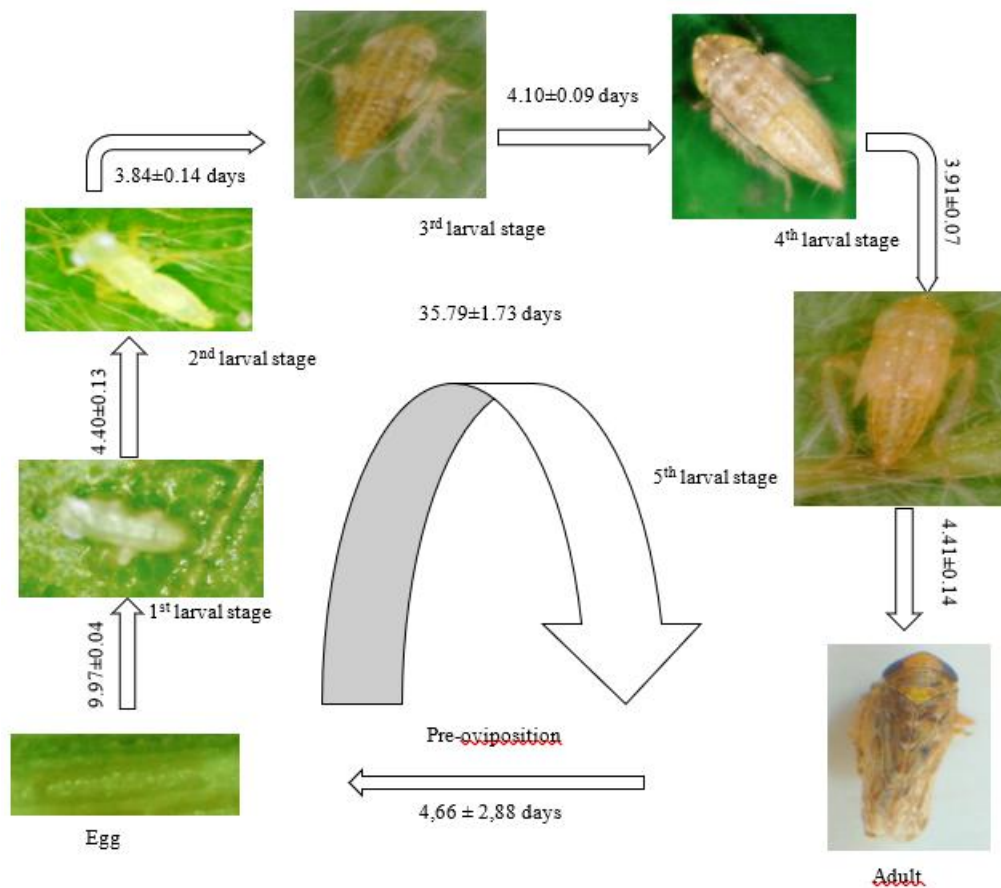


Figure 2. Development cycle of *Recilia mica* Kramer.

Table 2. Number of adults on plants and sex ratio.

Plant species		Adult males	Adult females	Sex-ratio
<i>Pueraria phaseoloides</i>	8	3	5	0,6
<i>Eleusine indica</i>	23	6	17	0,35
<i>Digitaria horizontalis</i>	15	6	9	0,67
<i>Cynodon dactylon</i>	8	3	5	0,6
<i>Sporobolus pyramidalis</i>	3	1	2	0,5

After hatching, the first (L1) and second (L2) instar larvae are white translucent. They measure 0.8 mm and 1.2 mm respectively and have very little mobility. The mean development time of the first larval instar was 4.40 ± 0.13 days and that of the second instar was 3.84 ± 0.14 days (Figure 2, Table 1). The third larval instar is a faint ivory-yellow color and 1.5 mm long. At this stage, the mean development time was 4.1 ± 0.09 days. The 4th Larval instar is 1.8 mm long and pale ivory yellow in color. The average development time was 3.91 ± 0.07 days. The fifth larval stage or pupal stage is dark ivory yellow in color. It

measured 2 to 2.2 mm in length with an average development time of 4.41 ± 0.14 days (Table 1). The total duration of the larval stage is 25 to 40 days with an average of 30.63 ± 0.61 days. The first eggs of *R. mica* were laid between three (03) to eight (8) days, with an average of 4.66 ± 2.88 days, after adults were paired (Figure 2, Table 1). Results showed that daily oviposition varied between 1 and 3 eggs on the plants. Analysis of variance revealed two distinct groups ($P < 0.001$) regarding the average number of eggs laid. Thus, the highest average numbers of eggs were laid on *Eleusine indica* with 1.33 ± 0.17 eggs, *Digitaria*

horizontalis with 1.13 ± 0.2 eggs, *Cynodon dactylon* with 1.10 ± 0.23 eggs and *Pueraria phaseoloides* (1 ± 0.07 eggs) (Figure 3). *R. mica* used these plants as a substrate for egg-laying. The average number of eggs laid was lowest on *Sporobolus pyramidalis* and *Panicum maximum*, with respectively 0.33 ± 0.16 eggs and 0.27 ± 0.03 eggs laid (Figure 3). The fertility rate (or hatching rate) of eggs was very high on *Eleusine indica* with $80.25 \pm 1.45\%$, high on *Pueraria phaseoloides* and *Digitaria horizontalis*. The fertility rate was $64.23 \pm 13.67\%$ and $69.96 \pm 7.79\%$,

respectively. The mean fertility rate was observed on *Cynodon dactylon* ($49.41 \pm 6.78\%$). However, it was very low on *Sporobolus pyramidalis* ($11.11 \pm 19.24\%$) and *Panicum maximum* where no egg was found ($00 \pm 00\%$) (Figure 4). The sex ratio of adults of *R. mica* obtained on plants during our study was in favor of females. However, the highest sex ratio (0.67) was observed on *Digitaria horizontalis* and the lowest (0.35) on *Eleusine indica* (Table 2).

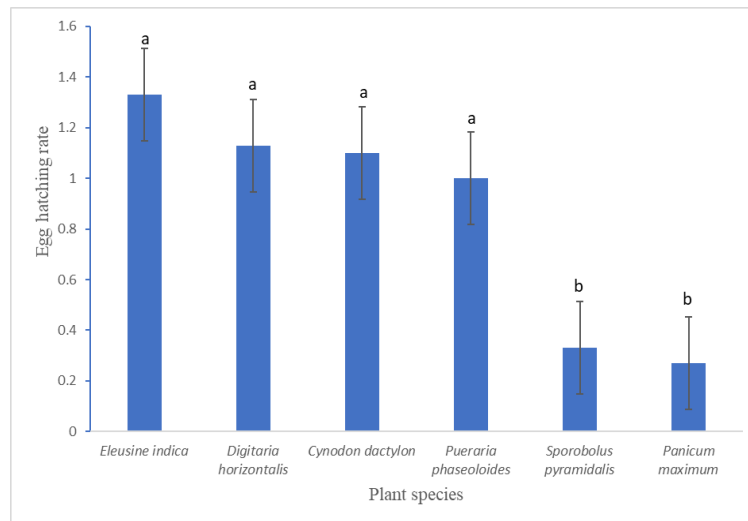


Figure 3. Average number of eggs laid per female on plants .

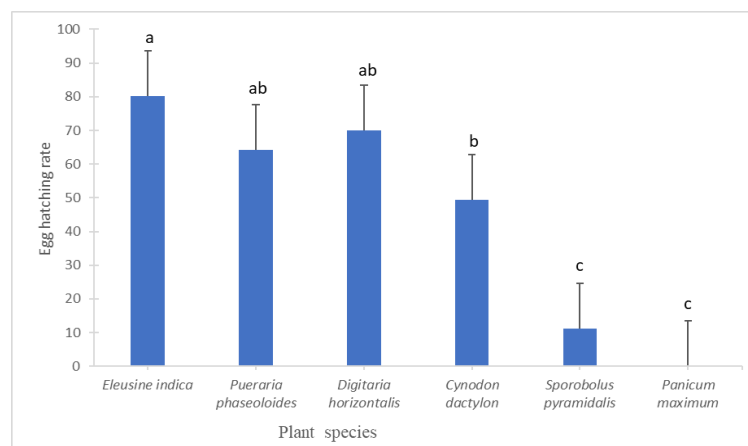


Figure 4. Egg hatching rate on plants.

Study of life cycle of *Recilia mica* Kramer showed three main stages of development, namely egg, larval stage and adult. Observations have shown that it is a heterometabolic type of development, with no immobile stage observed between the larvae and the adult, contrary to what occurs in holometabolic insects (Leraut and Mermet, 2003). Generally, in heterometabolic insects, and particularly in leafhoppers, the larvae look like the adults, have the external contours of wings, and grow with each molt (Engel and Grimaldi, 2004). In fact, from hatching to adult emergence, the various larval stages observed were separated by molts. Incubation period of *R. mica* eggs

varied between 9 and 13 days after laying. This incubation period is longer than that observed in *Recilia dorsalis* a closely related species, which is 7-9 days (Dennis, 2008). It also differs from that of the cotton leafhopper *Empoasca* spp. which hatch 6-10 days after egg-laying (Dennis, 2008). This incubation time of *R. mica* eggs can be related to temperature, which is believed to play an important role in the hatching of the eggs. According to Baillod *et al.*, 1993, the embryonic development of leafhopper eggs depends on temperature, but this should be below 32°C . Average temperature recorded during our study was 27.16°C .

R. mica has five (05) larval instars as in most species of the Cicadellidae family such as *Recilia dorsalis*, *Scaphoideus titanus*, *Empoasca vitis*, *Metcalfa pruinosa* and *Empoasca fabae* (Dennis, 2008; Voynaud *et al.*, 2019). These five larval instars are also observed in species of the Cercoidae family such as *Pseudotheraptus devastans* (Tano *et al.*, 2011; Doh, 2018), *Pseudotheraptus wayi* (Egonyu *et al.*, 2014). Total development time of the five larval stages of *R. mica* ranges from 25 to 40 days. The long duration of the larval stages may be due to the anatomical, morphological and physiological changes that these larvae undergo before reaching adulthood (Doh, 2018). This duration is longer than that of *R. dorsalis* which is 16-18 days and *Empoasca spp.* which ranges from 14-18 days (Dennis, 2008). Total life cycle length (from egg to adult death) averaged 40.45 ± 1.73 days. These results differ from those of (Philippe, 1991) who showed that the life cycle of *R. mica* averaged 35 days. This period is similar to that of *Scaphoideus titanus*, which ranges from 35 to 55 days (Chuche and Thiery, 2014; Grauby, 2018). The sex ratio of *Recilia mica* was female-biased, with females being more numerous than males in the laboratory progeny. A female-biased sex ratio may represent an adaptive reproductive strategy that maximizes population growth. It may also result from differential survival of the sexes during immature development or from favorable rearing conditions, including the quality of the host plant, which may enhance female survival and development.

CONCLUSION

The development cycle of *R. mica* is characterized by five larval stages. The average development duration from egg to adult is 40.45 days, with an incubation period of 9.97 days and a pupal development period of 25 to 40 days. Adults live about 5.16 days. *R. mica* performs its development cycle on plants *Eleusine indica*, *Digitaria horizontalis*, *Pueraria phaseoloides*, *Cynodon dactylon*, *Sporobolus pyramidalis*. These plants constitute the reservoirs of this species in the surroundings of the nursery. The results of this study will contribute to the development of an integrated control strategy against blast, a disease transmitted by *R. mica*, in oil palm nurseries.

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CONFLICT OF INTERESTS

The authors declare no conflict of interest

ETHICS APPROVAL

Not applicable

FUNDING

This study received no specific funding from public, commercial, or not-for-profit funding agencies.

AI TOOL DECLARATION

The authors declare that no AI and related tools are used to write the scientific content of this manuscript.

DATA AVAILABILITY

Data will be available on request

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