Biocidal effect of \textit{Crataeva religiosa} extracts against \textit{Caryedon serratus} (Olivier, 1789) pest of peanut stocks and seeds in Senegal

Khady Fall, Ablaye Faye *, Toffène Diome and Mbacké Sembène

Department of Animal Biology, Laboratory of Entomology and Acarology, Faculty of Science and Technology, Cheikh Anta Diop, University of Dakar, Senegal.

Abstract

This laboratory study was conducted to evaluate the biocidal effect of \textit{Crataeva religiosa} (Capparidaceae) on eggs and adults of \textit{Caryedon serratus}. An aqueous formulation obtained by maceration at 50 degrees was used in three concentrations 0.2kg/L, 0.1kg/L, 0.06kg/L. The results reveal that the extracts of \textit{C. religiosa} showed a rather remarkable ovicidal activity which varies from 41.66% in C1 and C2 to 38.88% in C3. A very important nymphal activity of 88.88% was also noted with the highest dose. The monitoring of surviving eggs showed a lengthening or a shortening of the different development durations and an imbalance of the sex ratio in favor of females. The adulticidal activity showed mortality spread over time of all doses. The extracts of \textit{C. religiosa} showed a fairly high adulticidal activity of 53.33% in C1, 40% in C2 and 46.66% in C3. A reduction in the fecundity of females treated with all doses was also noted and a balance in the sex ratio of survivors from treated adults.

Keywords: \textit{Caryedon serratus}; Peanut; \textit{Crataeva religiosa}; Biocide

1. Introduction

The peanut currently occupies a predominant place in the economic system of Senegal where its culture covers more than half of the cultivable surfaces. With its scientific name \textit{Arachis hypogaea} \textit{L.} it is the richest seed legume in terms of protein (25%) and lipids (50%) and constitutes a very important nutritional contribution for the local populations. Its cultivation is one of the best and cheapest solutions to fight against protein deficiency in Africa [1]. This legume remains the country’s main export crop and brings in about 80 million CFA francs each year, which represents 40% of the country’s total exports [2]. Unfortunately, it is attacked by several insects, the most dreaded of which is \textit{Caryedon serratus} (Olivier), which is responsible for quantitative losses of up to 83% after a storage period of 4 months [3]. The holes left by the larvae of \textit{C. serratus} facilitate the attack of other insects that decrease the nutritional quality of the peanut and promote the development of a mold, \textit{Aspergillus flavus}, that produces aflatoxin, which is highly carcinogenic. All these losses, which occur at all stages from harvest to consumption, not only harm farmers but also cost the national economy considerably [4].

The chemical pesticides widely used to control stock pests present potential risks to the health of the population and the environment, and their very high-cost forces farmers to resort to traditional control techniques again and revives the interest of specialists in orienting their thinking towards the use of biocidal substances of plant origin [1, 5].

In this study, which is part of the development of protection strategies for peanuts in Senegal, we tested the insecticidal effect of \textit{C. religiosa} extracts on the survival of eggs and adults of \textit{C. serratus} under controlled conditions.
2. Material and methods

2.1 Harvesting and conservation of plant material

The peanut used for mass rearing was purchased at Tilène market (Dakar). These peanut seeds were brought to the laboratory of Entomology and Acarology of the Faculty of Science and Technology of Cheikh Anta Diop University in Dakar where they were put in bags and kept in the freezer for 96 hours at 4°C to eliminate any hidden infestation. The seeds were then brought to room temperature and placed in glass jars 16 cm high and 8 cm in diameter, hermetically sealed to prevent further infestation. The leaves of *C. religiosa* are harvested in the vicinity of the Department of Animal Biology of the Faculty of Science and Technology of the Cheikh Anta Diop University of Dakar. The harvest is done early in the morning before sunrise. After harvesting, the leaves are freshly ground and used for aqueous extractions by maceration for biological tests.

2.2 Mass rearing

The original strain of *C. serratus* was obtained from peanut pods collected in the locality of Keur Baka (14° 09’ N-16° 04’ W) 22 km south of the Kaolack region. They are collected and kept in the laboratory in plastic bags at room temperature for at least two months. Cocoons formed outdoors are isolated in Petri dishes. Adults emerging from these cocoons are reared in the laboratory. Mass rearing is carried out in cylindrical glass jars (about 16 cm in diameter and 8 cm high), the lids of which are perforated and covered with muslin cloth to allow the insects to breathe. In each jar, peanut seeds are introduced until the base is completely hidden with enough male and female insects, a zigzag folded paper that allows the insects to move easily inside the jar and cotton soaked in distilled water. The jars are then left at room temperature. After 48 hours, the seeds having received oviposition, were deposited in glass Petri dishes where the egg will continue its development cycle until the emergence of the adult. Adult emergence was recorded and monitored every two days to maintain the cohort and avoid mixed batches of generations. Biological tests were performed on adults (adulticidal effect) and eggs (ovicidal effect) of *C. serratus* from this farm.

2.3 Preparation and conservation of the test solution

The method used is a maceration whose solvent is water. For *C. religiosa* 1 kg of fresh crushed leaves are extracted in 5 liters of tap water which is the solvent used at 50 degrees. The solution obtained is left at room temperature for a week and then filtered through a household sieve reinforced with muslin. The aqueous extracts are stored in glass bottles. These are placed in the refrigerator and used as needed. Three different concentrations are obtained: C1 = 1 kg /5L = 0.2 kg /L is the concentration of the initial solution from which two other concentrations are obtained by dilution; C2 = C1/2 = 0.1 kg/L and C3 = C1/3 = 0.06 kg/L.

2.4 Ovicidal test

Forty-eight-hour-old females *C. serratus* from mass rearing were placed in pairs to lay eggs on healthy peanut seeds. 24 hours after contact, the adults were removed from the seeds and the seeds were observed with a monocular magnifying glass to see eggs deposited on them. If a seed receives more than one egg, only one is left and the others were peeled off with fine tweezers so that there is no intraspecific larval competition.

In each Petri dish, 12 seeds containing one egg each are then sprinkled, with a 1 ml micropipette of each of the three concentrations, and the dish is lightly shaken so as to impregnate the seeds uniformly. For each concentration, three replicates and a blank control were made. For the blank control, the seeds were not treated. The next day, the seeds were placed in rectangular plastic boxes. Each box had 3 rows of 4 wells (logis) numbered by letters and numbers in index from 1 to 12. For each dose, three boxes were filled. The set of boxes was placed on the laboratory bench and monitored every day. The experiment was conducted at room temperature between 29°C and 35°C and 47-92% relative humidity. This study device allows to follow the eggs individually. For each seed, the date of oviposition corresponding to the day before the beginning of the experiment is mentioned. The same applies to the dates of hatching, cocoon formation and emergence of the surviving adults. It is then easy to calculate some biological parameters such as the mortality percentages of eggs, larvae, and the total mortality rate.

-Percentage of embryonic mortality

\[
\% \text{Embryonic Mortality} = \frac{\text{Number of unhatched eggs}}{\text{Total number of eggs}} \times 100
\]

The percentage of larval mortality
% Larval Mortality = \frac{\text{Number of dead larvae}}{\text{Total number of larvae}} \times 100

Total mortality percentage

Total Mortality = \frac{\text{Number of unhatched eggs} + \text{Number of dead larvae}}{\text{Total number of eggs}} \times 100

These mortalities were then corrected by Abott’s formula, which gives the corrected values of mortality in percentage according to the mortalities of the treated samples and the white control.

\[ M_C = \frac{M_T - M_{T0}}{100 - M_{T0}} \times 100 \]

Where

- \( M_C \): corrected mortality
- \( M_T \): mortality of treated insects
- \( M_{T0} \): mortality of untreated insects

### 2.4.1 Monitoring of "rescued" eggs

The study of the parameters of the developmental cycle conducted on the rescued eggs of *C. serratus* focused on:

- The duration of oviposition-hatching which represents the embryonic stage of development.
- The duration hatching-weaving of the cocoon or larval development which takes place essentially inside the seed.
- The duration of weaving-emergence or pupal stage.
- The duration of oviposition-emergence or total development phase covers the time between oviposition and emergence of the adult.
- The oviposition-death of the adult or total life span covers the time between oviposition and death of the adult.

### 2.4.2 Reproductive activity of "rescued" adults

The monitoring of these "rescued" adults of *C. serratus* is carried out to evaluate the possible effect of the extracts of these plants tested on a certain number of their biological parameters, such as:

- The sex ratio of the "rescued" adults
- The fecundity, the fecundity of these females and the life span of the "rescued" adults

The sex ratio, which corresponds to the ratio between the numbers of male individuals having emerged on the number of female individuals, is determined for each test product. Sexing of the emerging adults is done by observing the last abdominal segment which is curved in the male and elongated in the female. Mating between males and females was then performed. Each couple is placed alone in a numbered Petri dish with oviposition substrate. The oviposition of the "surviving" females of *C. serratus* was followed on healthy peanut seeds; for the importance of oviposition, the number of eggs laid on the walls of the jars and on the seeds by each female was counted every day under a binocular magnifying glass. Thus, infested seeds are replaced by perfectly healthy ones. It should be noted, however, that the conditions of no water and no food are applied to these emerging young adults. The experiment takes place at room temperature in the rearing room. The monitoring of the pairs is interrupted with their death, thus allowing the calculation of the total life span of the adults of *C. serratus*.

### 2.5 Adulticidal tests

The treated adults come from mass rearing in the laboratory in glass jars; they are at most 72 hours old. In each Petri dish, 10g of peanut seeds are placed. The seeds are then infested with 10 adults of *C. serratus* (5 males and 5 females). For each concentration, one milliliter (1ml) was sprayed on the peanut seeds in each box. This was then lightly shaken for 2-3 minutes to ensure distribution of the solution on the substrate. For each given concentration, three replicates and two controls (a white control and a solvent control) were performed. In the blank control, adults had no contact with the solutions and in the solvent control, one milliliter (1ml) of tap water was sprayed onto the peanut seeds. The insects were exposed to the aqueous extracts for one week. Dead bruchids were counted every 24 hours and laid eggs were also counted. The proportion of dead adults (number of dead/total number x 100) was calculated for each concentration of the solution tested. The results obtained were corrected using the formula of Abbott.

The number of eggs laid (fecundity) and the sex ratio of the offspring were also evaluated.
2.6 Statistical analysis

Repeat mean calculations and graphs were performed in Excel 2013. The Statistical analyses of the variables were performed with the R software. The normality of the data was checked by the Shapiro-Wilk normality test. Most variables did not follow the normal distribution. Thus, we used the non-parametric test and the most suitable was the Kruskal-Wallis test. It allowed us to compare the averages of the different doses used in order to know whether or not there were significant differences at the 5% threshold. Once the difference is significant, a multiple comparison between the doses will be made using the perwise test.

3. Results

3.1 Ovicidal effects of C. religiosa extracts

The results obtained by processing C. religiosa on C. serratus eggs are presented in Table 1.

Table 1 Ovicidal effect of C. religiosa extracts: mean (standard deviation)

<table>
<thead>
<tr>
<th>Solution</th>
<th>Concentrations</th>
<th>Embryonic Mortality</th>
<th>Larval Mortality</th>
<th>Pupal Mortality</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>C1</td>
<td>41.66± 14.43</td>
<td>68.33± 10.10</td>
<td>88.88± 19.25</td>
</tr>
<tr>
<td></td>
<td>C2</td>
<td>41.66± 22.04</td>
<td>80.55± 17.35</td>
<td>11.11± 19.24</td>
</tr>
<tr>
<td></td>
<td>C3</td>
<td>38.88± 12.73</td>
<td>58.99± 11.12</td>
<td>41.66± 38.19</td>
</tr>
<tr>
<td></td>
<td>TB</td>
<td>16.66± 8.33</td>
<td>88.88± 19.25</td>
<td>00± 00</td>
</tr>
<tr>
<td></td>
<td>TS</td>
<td>36.10± 26.78</td>
<td>40.55± 22.74</td>
<td>33.33± 57.73</td>
</tr>
</tbody>
</table>

Values are means followed by standard deviation. On a vertical line, means followed by the same superscript letter(s) are not significantly different from each other (p > 0.05).

The mortality rate of eggs treated with C. religiosa extracts varied from 38.88% to 41.66%. Thus, there is a significant difference between the highest dose C1 and the white control (p > 0.05). 41.66% of the eggs did not hatch with the highest doses; this mortality rate decreases as the dose is decreased; it is 38.88% with C3.

The larvicidal results show that the mortality rate obtained with the white control (88.88%) is significantly higher than those obtained with the lowest dose C3 (58.99%). The highest doses C1 and C2 gave mortality rates of 68.33% and 80.55% respectively.

Pupal efficiency is very high with the highest dose C1 (88.88%), which is significantly higher than the dose C2 (11.11%) and the white control where no pupal mortality is noted.

3.1.1 Monitoring of "surviving" eggs

Table 2 Effect of C. religiosa extracts on the mean durations (± standard deviation) of the different developmental phases of rescued eggs

<table>
<thead>
<tr>
<th>Average duration (Days)</th>
<th>Concentrations</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>C1</td>
</tr>
<tr>
<td>Laying/ hatching</td>
<td>8.71± 1.31</td>
</tr>
<tr>
<td>Hatching/cocon weaving</td>
<td>44± 4.12</td>
</tr>
<tr>
<td>Weaving/emergence</td>
<td>23± 00</td>
</tr>
<tr>
<td>Spawning/emergence</td>
<td>79± 00</td>
</tr>
<tr>
<td>Lifetime (P/M)</td>
<td>85± 00</td>
</tr>
</tbody>
</table>

Durations are expressed in days; values are means followed by standard deviation. On a horizontal line, followed by the same superscript letter(s) are not significantly different from each other (p > 0.05).
To determine the biocidal efficacy of *C. religiosa* extracts on the insect *C. serratus*, a follow-up of the treated eggs was conducted in the laboratory. The study of the parameters of the development cycle carried out on the “rescued” eggs concerned the average duration of oviposition/hatching, hatching/cocoon weaving, cocoon weaving/emergence, oviposition/emergence, and the life span of the “rescued” adult. The different results obtained from the experiments were listed in a table.

The results obtained from the biocidal action of *C. religiosa* on the average duration of the different developmental phases of the “rescued” eggs of the insect *C. serratus* are presented in Table 2.

The average oviposition/hatching time induced by *C. religiosa* is $8.75 \pm 0.37$ days with a minimum duration of $8.4 \pm 1.31$ days in C3 and a maximum duration of $9.14 \pm 1.27$ days in C2. The average duration of larval development was about $44.44 \pm 0.38$ days. All doses gave almost the same hatching/cocooning times. The average duration of weaving/emergence was $27.08 \pm 3.54$ days with a minimum duration of 23 days in C1 and a maximum duration of $29.25 \pm 8.77$ days in C3. Regarding the oviposition/emergence parameter, the average duration induced by the three doses tested is $82 \pm 2.64$ with a minimum duration of 79 days with C1 and a maximum of $84 \pm 11$ days in C2. The highest dose C3 induced a complete development time of about $83 \pm 9.14$ days.

As for the average lifespan, an increase in adult lifespan is noted; it is of the order of $100.05 \pm 13.4$ days, with a minimum duration of $85 \pm 0$ days in C1 and a maximum of $110.67 \pm 21.19$ days in C2 ($75.5 \pm 3.53$ days for the control).

### 3.1.2 Reproductive activity of “rescued” adults

Eggs previously treated with *C. religiosa* extracts and which have reached their adult stage are called “rescued” adults. We had planned to form pairs and then follow them individually to evaluate the effect of the biocides on the reproductive activity of the females, but our results do not allow us to form pairs. The study of the reproductive parameters carried out on the “surviving” populations of *C. serratus* only concerned the determination of the sexual ratio. The percentages of offspring from eggs previously tested with *C. religiosa* extracts and divided into male and female individuals allowed us to determine the sex ratio of the survivors. The results obtained are presented in the following table.

**Table 3** Effects of *C. religiosa* extracts on the sex ratio of male and female offspring from treated eggs

<table>
<thead>
<tr>
<th>Solution</th>
<th>Concentrations</th>
<th>Rescued individuals</th>
<th>Individuals Males</th>
<th>Individuals Females</th>
<th>Sex-ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. religiosa</td>
<td>C1</td>
<td>$2.77^a \pm 4.81$</td>
<td>0.00</td>
<td>$100^a \pm 1$</td>
<td>0.00</td>
</tr>
<tr>
<td></td>
<td>C2</td>
<td>$8.33^a \pm 8.33$</td>
<td>$67^a \pm 0.58$</td>
<td>$33^a \pm 0.58$</td>
<td>$2.03^a \pm 0.58$</td>
</tr>
<tr>
<td></td>
<td>C3</td>
<td>$13.89^a \pm 9.62$</td>
<td>$19.88^a \pm 0.58$</td>
<td>$80.12^a \pm 0.58$</td>
<td>$0.25^a \pm 0.29$</td>
</tr>
<tr>
<td></td>
<td>TB</td>
<td>$8.33^a \pm 14.43$</td>
<td>$33^a \pm 0.58$</td>
<td>$67^a \pm 1.15$</td>
<td>$0.49^a \pm 0.29$</td>
</tr>
<tr>
<td></td>
<td>TS</td>
<td>$27.78^a \pm 29.26$</td>
<td>$60.06^a \pm 2.64$</td>
<td>$39.94^a \pm 1.15$</td>
<td>$1.50^a \pm 1.32$</td>
</tr>
</tbody>
</table>

The values are averages followed by the standard deviation. On a vertical line, means followed by the same superscript letter(s) are not significantly different from each other (p less than 0.05).

These results show that the rates of rescued individuals are significantly the same (p > 0.05). For the survivors, the average of the percentages obtained in C1, C2 and C3 is $8.33 \pm 5.56$ for the control $8.33 \pm 14.43$. The highest dose C1 caused the lowest emergence rate ($2.77\%$) and was lower in absolute value than the control. In males, the results obtained show a total absence of males in C1. The highest number of males was obtained with the intermediate dose which was equal to $67\%$. In females, the results obtained with *C. religiosa* show significant differences. The lowest dose C3 induces a rate of female offspring significantly higher than those obtained in C2, TB and TS.

By analyzing the ratio between males and females, we can see that the highest sex ratio is obtained with the C2 dose which is $2.03 \pm 0.58$. The C3 dose gives a sex ratio equal to $0.25 \pm 0.29$.

### 3.2 Adulticidal effects

The results obtained from the adulticidal activity of *C. religiosa* extracts are presented in the form of histograms.

Figure 1 allows to compare the effects between concentrations. Plots followed by the same alphabetical letter are not significantly different at p ≥ 0.05.
For the *C. religiosa* extracts, the analysis according to the concentrations shows no significant difference between them (p > 0.05).

The average adult mortality rates obtained at these three concentrations were 53.33% for C1, 40% for C2 and 46.66 for C3. The untreated adults and the solvent control had mortalities of 33.33% and 30% respectively.

### Corrected mortalities

We can say that all doses gave mortalities lower than 50%. The highest dose C1 gave the highest mortalities 29.99%. The other doses C2 and C3 gave 10% and 19.99% mortality respectively. No significant difference was noted between these three doses.

**Table 4** Percentage of corrected mortality of *C. serratus* adults induced by *C. religiosa* extracts

<table>
<thead>
<tr>
<th>Solutions</th>
<th>Concentrations</th>
<th>Corrected mortalities</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>C. religiosa</em></td>
<td>C1</td>
<td>29.99 ± 36.87</td>
</tr>
<tr>
<td></td>
<td>C2</td>
<td>10 ± 12.5</td>
</tr>
<tr>
<td></td>
<td>C3</td>
<td>19.99 ± 19.09</td>
</tr>
</tbody>
</table>

Values are means followed by standard deviation. On a vertical line, means followed by the same superscript letter(s) are not significantly different from each other (p less than 0.05)

### Evolution of adult mortality as a function of applied dose and exposure duration

This treatment was spread over a 10-day interval beyond which no dead individuals were collected.

When we evaluate the level of mortality of adults of *C. serratus* according to the duration of exposure, we can note that the mortality rates vary according to the concentration and the duration of the treatment. Therefore, with the highest dose, the highest mortality was noted on the first day of treatment with 20%. The intermediate dose C2 gave between 1 and 3 days from the beginning of treatment 16.67% mortality and no mortality from day 4 to day 7 of treatment. The lowest dose C3 gave the highest mortality on day 7 with 10% mortality (Figure 2).
Figure 2 Mortality of adults treated with *C. religiosa* extracts as a function of dose and duration of exposure

### 3.2.3 Effect of *C. religiosa* extracts on the fecundity of treated females

The results obtained on the fecundity of *C. serratus* females treated with *C. religiosa* are presented in Figure 3 below. The diagram followed by the same alphabetical letter are not significantly different at \( p \geq 0.05 \).

![Figure 3: Effect of *C. religiosa* extracts on fecundity of treated females](image)

Evaluated after 10 days of treatment with *C. religiosa* extracts, the average fecundity of *C. serratus* females varied with the applied dose. Compared to the samples, we can say that the extracts of *C. religiosa* reduce the fecundity of the treated females, in absolute value. The most important reduction in fecundity was noted with the C2 dose with an average fecundity of 2.33 eggs per female. Doses C1 and C3 caused average fecundity with 3.47 and 3.87 eggs respectively. However, the statistical analysis does not show any significant difference between the average number of eggs laid by the females treated with the different concentrations and the samples.

### 3.2.4 Rescued" adults

Adults previously treated with *C. religiosa* and whose eggs have reached the adult stage are called "rescued" adults. The study of the reproductive parameters carried out on the "rescued" populations of *C. serratus* focused on the determination of the sexual ratio.
The process followed for the insecticide tests on eggs was to first examine the efficiency of the extracts on the eggs and to evaluate the impact of the treatment on the development of those that survived as well as on the sex ratio of their offspring.

The analysis of the results shows a rather remarkable ovicidal activity. Compared to the white control, we can note a reduction in the hatching of *C. serratus* eggs with the application of all doses between 38.88% and 41.66%. Statistically, we noticed an equal efficiency of the effect of all doses at p < 0.05. Thus, the highest doses induced the same percentage of ovicidal mortality (41.66%).

Unlike eggs, the larvicidal effect observed in the white control (88.88) was significantly higher than those obtained with the lowest dose C3 (58.99). No significant difference was noted between the highest doses (C1; C2) and the white control.

Regarding nymphal mortality, the highest dose proved to be more effective with 88.88% mortality, which is significantly higher than those obtained with the C2 dose and the blank control. This reveals the persistence of its biological activity during the treatment leading to a disruption or stopping of the development of the insect at the pupal stage. The lowest doses C2 and C3 gave mortality rates of 11.11% and 41.66% respectively. These results are in line with those of Faye [7] who showed an ovicidal effect ranging from 33.3 to 45% with fresh grinded leaves of *Senna occidentalis* on *Callosobruchus maculatus*. The same author obtained with the aqueous extracts of *C. religiosa* an ovicidal efficiency between 50.02 and 57.70%. Thiaw et al., [7] showed a larval mortality of 13.96 ± 4.85% with methanolic extracts of *S. occidentalis*, with the hexane and acetate fraction a mortality of 4.17 and 5.63 ± 2.52% respectively on *C. serratus*.

Our results also show that *C. religiosa* extracts affect the average durations of the different developmental stages of treated *C. serratus* eggs. Its action varies according to the doses applied. The average duration of egg laying/hatching induced by *C. religiosa* was 8.75 ± 0.37 days with a minimum duration of 8.4 ± 1.31 days in C3 and a maximum duration of 9.14 ± 1.27 days in C2. The white control caused an embryonic development time of 8.02 ± 0.14 days. The average duration of hatching/coconut weaving was almost equal with all doses 44 days in C1 and 44.66 days in C2 and C3. A slight extension of larval development time was noted between the three doses tested and the control (41 days). Ndiaye [8] reports that the incubation period is 6-8 days, and the development time from egg to the resulting adult is 45-47 days. Delobel [9] reports that, under the usual conditions in Senegal, the egg hatches after about a week and the neonate larva punctures the pod, passes through the pericarp, pierces the seed coat and enters the seed, which it consumes. He also specifies that the larval development lasts a little more than one month; at the end of this time, the larva weaves a cocoon from which an adult will emerge 15 days later. This same duration of pupation was noted by Robert [10] at 35°C. The work of Ndiaye [3] and Delobel & Tran [11] indicate a development of the larvae between 40 and 58 days depending on the temperature and relative humidity conditions. Gueye [12] reveals, in his studies, a larval stage duration of about 45 days on average at 35°C. In contrast, our results revealed a pupal development that varied from 23 ± 00 to 29.25 ± 8.77 days depending on the dose applied. These results confirm those of Thiaw [1] who showed a duration of pupal development that varied 21.33 and 33.43 days with the extract and the methanolic fraction of *Calatropis procera* and *Senna occidentalis* on the same insect. From oviposition to adult emergence, the treated eggs show a duration that varies from 79 ± 00 to 84 ± 11 days. From oviposition to adult death, they showed a total life span ranging from 85 to 110.67 ± 21.19 days. Like most insects, the characteristics of larval development and adult performance are primarily temperature dependent. Many authors have described in various insects a positive correlation between temperature

### Table 5: Effects of *C. religiosa* extracts on the sex ratio of male and female offspring from treated adults

<table>
<thead>
<tr>
<th>Solution</th>
<th>Concentrations</th>
<th>Rescued individuals</th>
<th>Individuals Males</th>
<th>Individuals Females</th>
<th>Sex-ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>C. religiosa</em></td>
<td>C1</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
</tr>
<tr>
<td></td>
<td>C2</td>
<td>0.33± ±0.58</td>
<td>100 ±58</td>
<td>0.00</td>
<td>0.00</td>
</tr>
<tr>
<td></td>
<td>C3</td>
<td>1± ± 1.73</td>
<td>33.33±±0.58</td>
<td>66.67±1.15</td>
<td>0.49±0.29</td>
</tr>
<tr>
<td></td>
<td>TB</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
</tr>
<tr>
<td></td>
<td>TS</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
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</tr>
</tbody>
</table>

Values are means followed by standard deviation. On a horizontal line, means followed by the same superscript letter(s) are not significantly different from each other (p less than 0.05)

### 4. Discussion

The process followed for the insecticide tests on eggs was to first examine the efficiency of the extracts on the eggs and to evaluate the impact of the treatment on the development of those that survived as well as on the sex ratio of their offspring.

The analysis of the results shows a rather remarkable ovicidal activity. Compared to the white control, we can note a reduction in the hatching of *C. serratus* eggs with the application of all doses between 38.88% and 41.66%. Statistically, we noticed an equal efficiency of the effect of all doses at p < 0.05. Thus, the highest doses induced the same percentage of ovicidal mortality (41.66%).

Unlike eggs, the larvicidal effect observed in the white control (88.88) was significantly higher than those obtained with the lowest dose C3 (58.99). No significant difference was noted between the highest doses (C1; C2) and the white control.

Regarding nymphal mortality, the highest dose proved to be more effective with 88.88% mortality, which is significantly higher than those obtained with the C2 dose and the blank control. This reveals the persistence of its biological activity during the treatment leading to a disruption or stopping of the development of the insect at the pupal stage. The lowest doses C2 and C3 gave mortality rates of 11.11% and 41.66% respectively. These results are in line with those of Faye [6] who showed an ovicidal effect ranging from 33.3 to 45% with fresh grinded leaves of *Senna occidentalis* on *Callosobruchus maculatus*. The same author obtained with the aqueous extracts of *C. religiosa* an ovicidal efficiency between 50.02 and 57.70%. Thiaw et al., [7] showed a larval mortality of 13.96 ± 4.85% with methanolic extracts of *S. occidentalis*, with the hexane and acetate fraction a mortality of 4.17 and 5.63 ± 2.52% respectively on *C. serratus*.

Our results also show that *C. religiosa* extracts affect the average durations of the different developmental stages of treated *C. serratus* eggs. Its action varies according to the doses applied. The average duration of egg laying/hatching induced by *C. religiosa* was 8.75 ± 0.37 days with a minimum duration of 8.4 ± 1.31 days in C3 and a maximum duration of 9.14 ± 1.27 days in C2. The white control caused an embryonic development time of 8.02 ± 0.14 days. The average duration of hatching/coconut weaving was almost equal with all doses 44 days in C1 and 44.66 days in C2 and C3. A slight extension of larval development time was noted between the three doses tested and the control (41 days). Ndiaye [8] reports that the incubation period is 6-8 days, and the development time from egg to the resulting adult is 45-47 days. Delobel [9] reports that, under the usual conditions in Senegal, the egg hatches after about a week and the neonate larva punctures the pod, passes through the pericarp, pierces the seed coat and enters the seed, which it consumes. He also specifies that the larval development lasts a little more than one month; at the end of this time, the larva weaves a cocoon from which an adult will emerge 15 days later. This same duration of pupation was noted by Robert [10] at 35°C. The work of Ndiaye [3] and Delobel & Tran [11] indicate a development of the larvae between 40 and 58 days depending on the temperature and relative humidity conditions. Gueye [12] reveals, in his studies, a larval stage duration of about 45 days on average at 35°C. In contrast, our results revealed a pupal development that varied from 23 ± 00 to 29.25 ± 8.77 days depending on the dose applied. These results confirm those of Thiaw [1] who showed a duration of pupal development that varied 21.33 and 33.43 days with the extract and the methanolic fraction of *Calatropis procera* and *Senna occidentalis* on the same insect. From oviposition to adult emergence, the treated eggs show a duration that varies from 79 ± 00 to 84 ± 11 days. From oviposition to adult death, they showed a total life span ranging from 85 to 110.67 ± 21.19 days. Like most insects, the characteristics of larval development and adult performance are primarily temperature dependent. Many authors have described in various insects a positive correlation between temperature
and developmental rate or between temperature and performance [13, 14, 15]. In C. serratus, in particular, low temperatures (20°C) induce an extension of developmental time (183 days on average) [5].

The average emergence rate of adults from C. serratus eggs previously treated with C. religiosa extracts was 8.33 ± 5.56 with a maximum of 13.89 ± 9.62 in C3 and a minimum of 2.77 ± 4.81 in C1. This could be explained by the persistence of the biocide action of the product, thus reducing the emergence of adults. This work is not in line with the results of Thiaw [1] which showed higher emergence rates ranging from 30.55 to 52.78% with crude extracts and extract fractions of C. procera.

The effect of C. religiosa extracts on the sex ratio is in favor of females for doses C1 and C3 which would increase the risks of population increase. For the C2 dose, the effect is in favor of males, leading to a competition between males. The opposite effect was observed in the work of Gningue et al. [16], where the sex ratio was in favor of males for doses C1 and C2; females for dose C3 with the application of C. religiosa extracts.

Studies on the evolution of the adulticidal activity of C. religiosa extracts revealed mortality rates that varied according to the concentration and duration of the treatment. It showed an adulticidal efficacy between 46.66 and 53.33% with the application of all doses. The highest dose C1 caused, in 48 h, 30% mortality; the other concentrations were less effective. The work of Faye [6] showed 87.25% efficacy in C1 (0.2g/cm3) and C2 (0.13g/cm3) and 74.87% in C3 (0.1g/cm3) with the aqueous extract of C. religiosa leaf powder on C. maculatus adults at day 10.

The females fecundity evolution treated with the different concentrations of C. religiosa showed in absolute value a reduction of the fecundity, compared to the white control, with the application of all the doses. The greatest reduction in fecundity was noted with dose C2 with an average fecundity of 2.33 eggs per female; 7.73 for the control. Doses C1 and C3 caused average fecundities with 17.33 and 19.33 eggs respectively. These results are at variance with the work of Thiaw et al. [7] who reveal that biocidal extracts of S. occidentalis showed percentages of reduction in fecundity of females from treated eggs ranging from 27.29% (methanol extract) to 67.2% (ethyl acetate fraction) on the same insect. The work of Gueye [17] also showed a reduction in fecundity with the extracts of Lantana camara. This reduction could be explained by a low longevity of females due to the biocide effect of the product. Indeed, during its application on adults, we noticed that a few minutes after the treatment, the insects were dead or paralyzed. Therefore, the females did not have time to lay eggs or they are unable to lay eggs because of paralysis.

The offspring of the tested adults (the survivors) were obtained only with the low doses with very low numbers. These results confirm the results of Doumma et al. [18] who studied the action of ground leaves of Boscia senegalensis on C. maculatus. They are also in agreement with the results of Mazibur and Gerhard, [19] and Ketoh et al. [20]. Regarding the sex ratio, the results obtained show that it is in favor of females for the C3 dose. From these results, we can conclude that the predominance of females would increase the risks of population increase in stored seeds, hence the importance of damage in storage areas. The work of Gningue et al. [16], shows a sex ratio in favor of males with the application of C. religiosa extracts on C. serratus.

5. Conclusion

The purpose of this study was to examine the efficiency of C. religiosa extracts on the survival of C. serratus in the egg and adult stages.

The results obtained revealed that the extracts of C. religiosa had both ovicidal and adulticidal effects on peanut bruchid, which varied according to the concentration. The high concentration C1 induces a more marked adulticidal effect than the other concentrations C2 and C3. On the other hand, at the egg level, both two highest doses showed the same ovicidal effects. In addition to those ovicidal effects, the biocidal activity of the C. religiosa extracts on the physiology and behavior of the insect is also expressed by a lengthening or a shortening of the duration of the developmental stages of the treated eggs and an imbalance in the sex ratio of the surviving adults in favor of the females.

Monitoring of treated adults revealed a reduction in fecundity of these females and a balance in the sex ratio of the survivors.

It is not sufficient to limit ourselves to laboratory tests, therefore, confirmation of these results in real storage situations is essential as well as a series of experiments to better situate the optimal application doses and to test the germination of processed seeds.
Compliance with ethical standards

Acknowledgments

The authors thank all those who participated in the success of this research article.

Disclosure of conflict of interest

No conflict of interest on this research work.

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