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# Evaluation of the Insecticidal Activity of Powders of *Ageratum conyzoide* L. Leaves and Roots of *Securidaca longepedunculata* Fresen on the Germination Rate of Cowpea Infected with *Callosobruchus maculatus*

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**Abstract:** Cowpea is a vegetable that contributes to food security in Guinea. Despite its importance, more than 30% of its production is lost between harvesting, storage and consumption. The main cowpea pest is *Callosobruchus maculatus*. The commonly used control strategy is essentially based on chemicals whose use is toxic, expensive and restrictive. In the search for alternatives to chemical control, this work was undertaken to evaluate the insecticidal potential of *Ageratum conyzoid* leaf powders and *Securidaca longepedunculata* roots. Powder toxicity and insecticidal efficacy tests were performed separately on groups of 20 *Callosobruchus maculatus* using 3 doses of *Ageratum conyzoid* leaf powders and *Securidaca longepedunculata* roots in jars each containing 100 g cowpea. The mortality of *Callosobruchus maculatus* and the impact of powders were noted respectively for 96 h and 4 months after infestation. The germination capacity of the treated seeds was assessed at the start of the study. Doses of 1 and 8 g of *Ageratum conyzoides* leaf powders and *Securidaca longepedunculata* roots were applied respectively to every 100 g of cowpea and led to 100% mortality of *Callosobruchus maculatus*. At these same doses, more than half of the treated seeds were attacked during the 4 months of storage. Germination tests have shown that powders of *Ageratum conyzoides* leaves and roots of *Securidaca longepedunculata* have no negative effect on the germination power of cowpea seeds. Therefore, they could be considered as excellent bio-insecticides that socio-professional strata (farmers and warehouse workers) can use in the fight against cowpea insects intended for storage.

**Key words:** Ethnobotany, insecticidal activity, cowpea, *Ageratum conyzoides*, *Securidaca longepedunculata*, *Callosobruchus maculatus.*

## 1. Introduction

Legumes and cereals are the most traded products of international markets and are the basis of the diet of most countries in the world, especially developing countries and underdeveloped countries such as Guinea where their diet is based mainly on legumes as a source of protein for example: cowpea with all its varieties, the different seeds of lentils, peas, chicks and others, and on rice, maize as a source of carbohydrate.

Every year, nearly 20,000 species of insects threaten world production and destroy a large part of cereals and food legumes and cause significant damage; they degrade the nutritional and organoleptic quality of the stored product and lead to the total loss of the product, of which more than 30% of production is lost between harvest, storage for consumption [1].

To increase the production of legumes, reduce the pressure of all kinds of pests, and to limit its damage, farmers, peasants and traders in markets use different control methods but the most used is chemical control

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[2] including the use of pesticides on a large scale.

Unfortunately studies have shown that these products used are dangerous on the environment and harmful to human health, these dangers have led the WHO (World Health Organization) to ban the use of certain chemical insecticides. Food crops are economically important for developing countries [3]. It is therefore necessary to seek effective methods of controlling insect pest populations in order to limit losses due to *Callosobruchus maculatus*, which are among the main pests of cowpea seeds.

Among insect pests, the cowpea buff *Callosobruchus maculatus* is of unknown origin and its affinity is subtropical and tropical. The search for effective insect pest control methods is beneficial for human health and the environment, many studies are done and others are developed to give many possibilities in the plant kingdom to isolate substances that have a role as insecticides [2].

The main objective of our study is to evaluate the insecticidal activity of two plants on cowpea bruche adults; *Callosobruchus maculatus*, which we assume as a possibility of reducing biodegradability by choosing this theme “Ethnobotanical investigation, monographic and evaluation of the insecticidal activity of two plants sold in the different markets of Conakry”.

## 2. Materials and Methods

### *2.1 Presentation of the Study Zone*

The study was carried out in Conakry, which is the capital of the Republic of Guinea and located on the Atlantic Ocean in the peninsula of Kaloum, its surroundings is extend inwards by the Kakoulima massif, which culminates 1,007 m. The city is between 9°35′ and 9°40′ latitude north and between 13°37′ and 13°42′ longitude west, altitude 16 m. The climate of the city is tropical, with a dry season from December to April, and a rainy season due to the African monsoon that extends approximately from Mid-May to mid-November, rainfall totals 3,775 mm per year: they are therefore very abundant. Average temperature of the coldest month (August) is 26.0 ℃ that of the hottest month (April) is 28.7 ℃. Its area is 450 $km^2$ with an estimated population of 2,317,376 inhabitants in 2017. Its vegetation consists of mangroves, palm trees and coconut trees. It is a cosmopolitan city where many people live side-by-side ethnic groups of Guinea [4] (Fig. 1).

### *2.2 Study Framework*

The organic chemistry laboratory of the Gamal Abdel Nasser University of Conakry, the laboratory of the national quality control office of Matoto Conakry, served as frameworks for studies.

### *2.3 Experimental Equipment (Cowpea)*

The cowpea used for the test was purchased at the Matoto market (Conakry) on 30 January 2020. Prior to testing, sorting was done to remove bad grains and foreign matter. Before using them in the different test steps, they were then frozen for 2 days at -7 ℃. After freezing the cowpea grains were dried in the sun for 1 h, then sterilized in the oven for 15 min, to remove traces of eggs and larvae (Fig. 2)

### *2.4 Assessment of Cowpea Seed Infestation in the Laboratory*

#### 2.4.1 *Callosobruchus maculatus* (Cowpea Weevil)

The bruchids used in this study were collected from a batch of 8 kg cowpea seeds purchased in shops in Madina (Conakry), using a mouth vacuum cleaner. The insects were placed in 10 pairs in jars of 1 L of volume and containing 100 g of cowpea bought at the Madina market, and not treated with insecticides. The openings of these jars have been carefully covered with muslin cloth to allow aeration and prevent the exit of insects and any other external contamination. This phase lasted 30 days to allow the emergence of new adults. After this operation, the cowpea seeds contained in the various jars were sieved; Adult bruchids were removed and rearing continued with egg-infested cowpeas. This second phase lasted 23 days and the insects obtained, aged 3 days, were used to

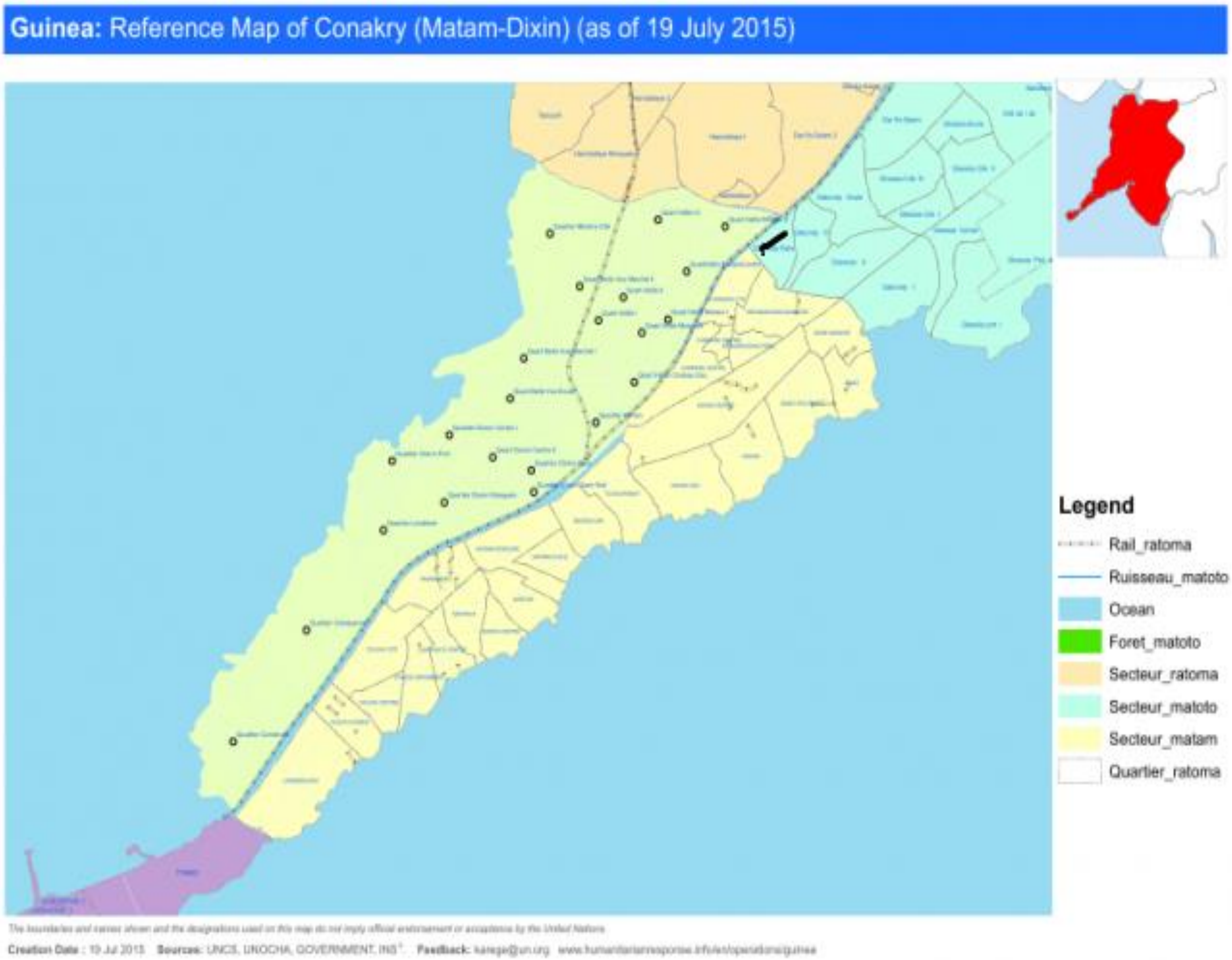


**Fig. 1 Map of Conakry.**

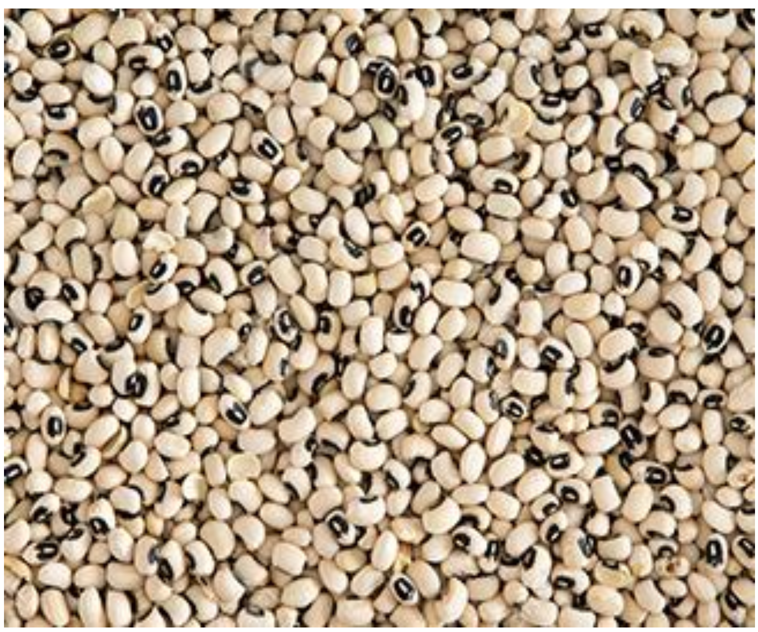

**Fig. 2 Uninfested cowpea seeds from the Matoto market (Conakry), photo Mr. Diogo Bangoura (30/01/2020).**

infest the cowpea and submit to the various treatments. All these breeding operations were carried out at the organic chemistry laboratory of the Department of Chemistry of Gamal Abdel Nasser University in Conakry at an average temperature of 32 ± 3 ℃ and an average relative humidity of 51% ± 7%.

2.4.2 Experimental Device

In total we used 24 jars. They were arranged in 3 batches of 8 jars and 3 repetitions for each bio-insecticide. The first two batches were intended for the study of the insecticidal effect of powders of *Ageratum conyzoides* leaves and roots of *Securidaca longipedunculata* and the last batch for the evaluation of the damage caused by *Callosobruchus maculatus* for 120 days. The jars were kept in a room well ventilated to laboratory conditions (Fig. 3).

2.4.3 Cowpea Infestation

According to the AGRAR-2013 method (African Research on agriculture, Food, and nutrition) 100 g of cowpea seeds were weighed and then introduced into jars previously marked $T_0$ (control), Fac1, Fac2, Fac3 (powder of the leaves of *Ageratum conyzoides*) and Fsl1, Fsl2, Fsl3 (powder of the roots of *Securidaca longipedunculata*). Thus the powder of leaves and roots were weighed at masses respectively, 1 g, 4 g and 8 g for each, added to 100 g of cowpea homogenized in the jars. Control jars ($T_0$) were not treated [5]. After weighing the powder of the different plants was mixed with the seeds and the whole was well shaken in order to homogenize and introduce into each jar 10 pairs of adult bruchids. These jars were then covered with muslin cloth and sealed with the elastic fronds to prevent any external contamination. It was found that the contents of the jars were infested on the same day (Fig. 4).

### 2.5 Data Collection

After 4 days, the contents of the jars were observed to determine the toxicity of the powder of the leaves *Ageratum conyzoides* and roots *Securidaca longipedunculata* was evaluated according to the doses administered. For this, the jars were sieved to extract the adult weevils and then keep the grains infested. At the same time, the dead individuals were counted and for this it was considered dead, an individual who did not move after several touches of the legs and antennae using a syringe or needle. It was also found that after 5 days the presence of laying traces and 10 days that of the larvae were detected (Fig. 5).

**Fig. 3 Experimental design 1 (Bangoura M.D., 2020).**

**Fig. 4 Experimental design 2 (Bangoura M.D., 2020).**

**Fig. 5 Experimental design 3 (Bangoura M.D., 2020).**

*2.6 Determination of Evaluated Parameters*

2.6.1 Cowpea Moisture

Constant weight drying method cowpea grains. The result is given by Eq. (1) [6]:

$$\mathrm{H}\ (\%) = \frac{P_2 - P_1}{Pe} \times 100 \qquad (1)$$

where: $H$ (%) = Humidity; $P_2$ = Weight of the capsule + sample before drying; $P_1$ = Weight of the capsule + sample after drying; $P_e$ = Weight of the test portion.

2.6.2 Toxicity Calculation

A few days after the introduction of adult weevils and powders into different jars, mortalities in the treated boxes (*Mo*) were expressed according to the W. S Abbott formula. Adjusted mortalities (MC) [7], take into account natural mortalities observed in control boxes (*Mt*).

According to the Eq. (2):

$$Mc = \frac{Mo - Mt}{100 - Mt} \times 100 \qquad (2)$$

where: *Mc* is the corrected mortality; *Mo* mortality in treated dishes and *Mt* mortality in control boxes.

2.6.3 Damage Assessment

The two criteria for assessing damage are: the percentage of attack and that of weight loss.

(1) Attack percentage **A (%)**

It consists of separating a batch of 100 or better of 1,000 healthy grains and that of the attacked grains and is determined by Eq. (3):

$$A(\%) = \frac{Na}{Ns + Na} \times 100 \qquad (3)$$

where: *Ns* is the number of healthy grains and *Na* is the number of grains attacked.

(2) The percentage of weight loss *P* (%)

It is calculated by the Eq. (4):

$$P(\%) = \frac{PsNa - PaNs}{Ps(Na + Ns)} \times 100 \qquad (4)$$

where: *Ps* represents the weight of healthy grains; *Pa* represents the weight of grains attacked.

Alternatively, the weight of treated seeds after the last emergence of *C. maculatus* individuals and control lots can be used using a precision scale. This method is applied by the Eq. (5):

$$P\ (\%) = [(\text{Initial weight - Final weight}) / \text{Initial weight}] \times 100 \tag{5}$$

2.6.4 Insecticidal Efficacy of the Two Plants *Ageratum conyzoides* and *Securidaca longipedunculata* against *Callosobruchus maculatus* (Cowpea Weevil)

Subsequently, the impact of bioinsecticide powders on damage caused by *Callosobruchus maculatus* was measured after 120 days.

2.6.5 Germination Test

After contact treatments, the seeds were subjected to the germination test which consists of taking 20 seeds at random from each treated sample and then introducing to the inside of the cotton soaked in water in petri dishes without a lid. After six days, sprouted seeds were counted for each sample (Fig. 6). For the test, we made two repetitions made on 100 g of cowpea each.

The germination rate was evaluated before the experiment, and by counting sprouted grains against seeded grains according to Eq. (6) [5].

$$G\ \ (\%) = \frac{NGG}{NGE} \times 100 \tag{6}$$

where: G (%) is the germination percentage, NGG is the number of sprouted grains and NGE is the number of seeds seeded.

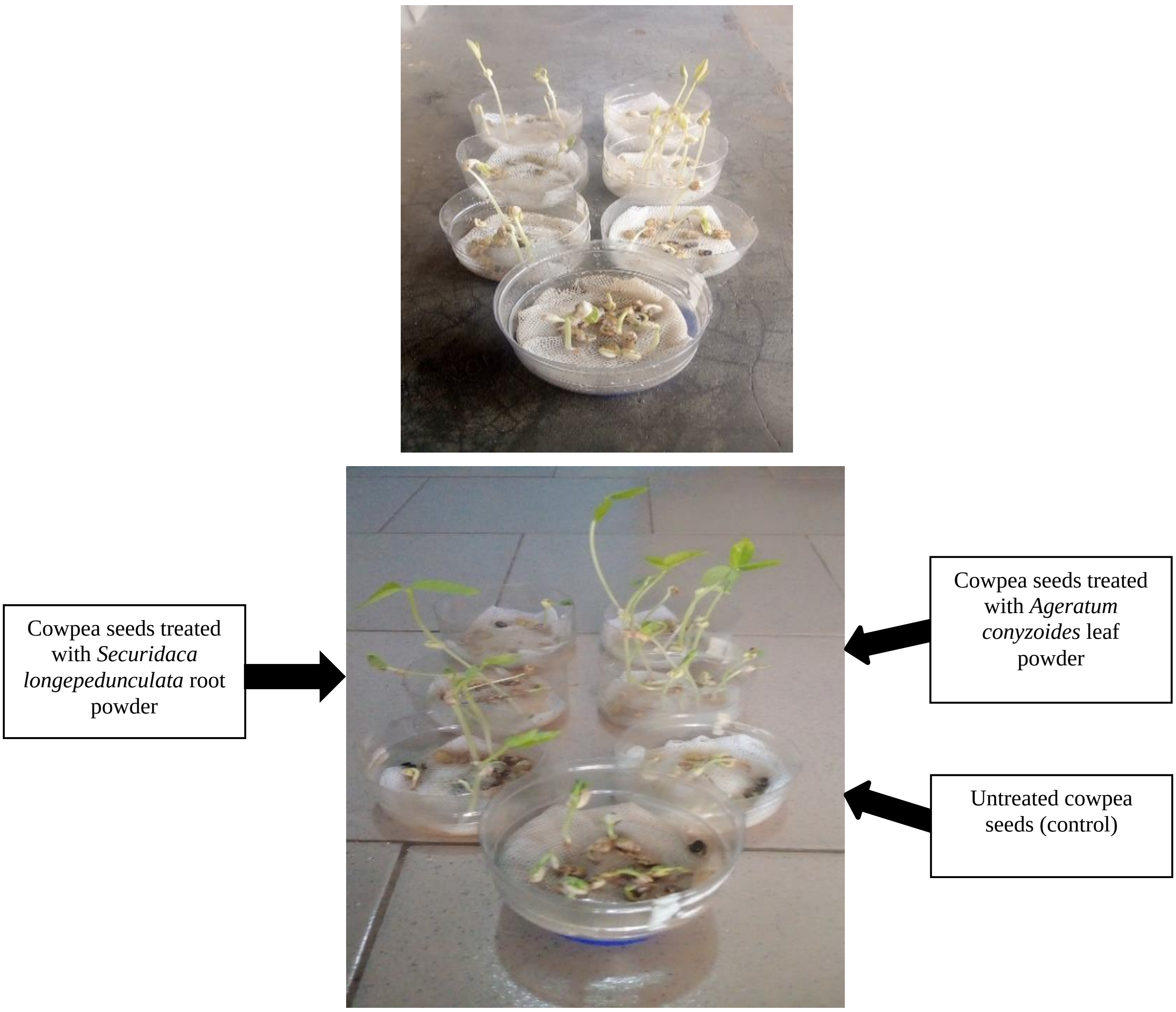


**Fig. 6 Experimental device of the germination test of seeds treated with the powder of leaves of *Ageratum conyzoides* and roots of *Securidaca longepedunculata* (Photo Mr. Diogo Bangoura 01/03/2020).**

*2.7 Data Analytics*

The data obtained were submitted to Excel 2013 and the averages of the different treatments were compared using Student's test at the 5% level.

## 3. Results and Interpretations

*3.1 Determination of Humidity*

According to the literature the humidity of cowpea in general varies from 13%-15%, in our case the humidity is 14.80. So the moisture of the cowpea that was used for the test is within the standards [8].

The average moisture content of cowpea grains after drying was 14.60%, which is the limit of appreciation of drugs 15% [5]. These values show that cowpea grains purchased from the market to carry out this work had a high moisture content and are presented in Table 1.

*3.2 Evaluation of the Toxicity of Powder of Ageratum conyzoides Leaves and Roots of Securidaca longipedunculata*

During the evaluation of the insecticidal effect of powder of *Ageratum conyzoides* leaves and *Securidaca longipedunculata* leaves, all doses (1 g; 4 g; 8 g) used resulted in mortality of *Callosobruchus maculatus*. The different mortality rates observed depend on the dose administered. Thus, mortality rates of 75 ± 0.82; 85 ± 0.8 and 100% were observed after 5 days of exposure of *Callosobruchus maculatus* to *Securidaca longipedunculata* root powder at the same dose per 100 g cowpea each respectively. As for the powder of the leaves of *Ageratum conyzoides* although the results obtained show no significant difference ($p < 0.05$) with those recorded with the powder of the roots of *Securidaca longipedunculata*, a slight decrease in the mortality rate was observed. In general, depending on the doses and the number of dead individuals, the powder of the leaves of *Securidaca longipedunculata* appears more toxic vis- à-vis *Callosobruchus maculatus* than the powder of the leaves of *Ageratum conyzoides*. A significant difference ($p < 0.05$) was recorded between the mean mortality obtained in jars treated with *Securidaca longipedunculata* leaf powder and *Ageratum conyzoides* leaf powder and that of control jars [6] (Table 2 and Fig. 7).

The root powder of *Securidaca longepedunculata* is the most toxic, because mortality undergoes an average increase of 5% from the lowest dose of 1 g (Fig. 7).

**Table 1 Cowpea grain moisture results.**

| Designation | $P_2$ | $P_e$ | $P_1$ | H% | $H_M$ |
|---|---|---|---|---|---|
| | 27.83 | 5.00 | 27.10 | 14.60 | |
| Cowpeas | 28.93 | 5.00 | 28.20 | 14.40 | 14.60 |
| | 25.84 | 5.00 | 25.10 | 14.80 | |

**Table 2 Toxicity assessment of *Ageratum conyzoides* leaf powder and *Securidaca longipedunculata* roots.**

| Treatment | Doses (g/100 g cowpea) | Average mortality rate (%) (standard deviation) |
|---|---|---|
| $Fac_1$ | 1 | 60 ± 0.83 |
| $Fac_2$ | 4 | 80 ± 0.81 |
| $Fac_3$ | 8 | 95 ± 0.81 |
| T0 | 0 | 1.5 |
| Rsl | 1 | 75 ± 0.82 |
| Rsl | 4 | 85 ± 0.8 |
| Rsl | 8 | 100 ± 0.81 |

Fac: Leaves of *Ageratum conyzoides*; Rsl: *Securidaca longepedunculata* root.

In this table, the means do not show a significant difference according to Student's test at the 5% threshold. Fac: powder treatment of *Ageratum conyzoides* leaves; Rsl: powder treatment of the roots of *Securidaca longepedunculata*; T0: witness.

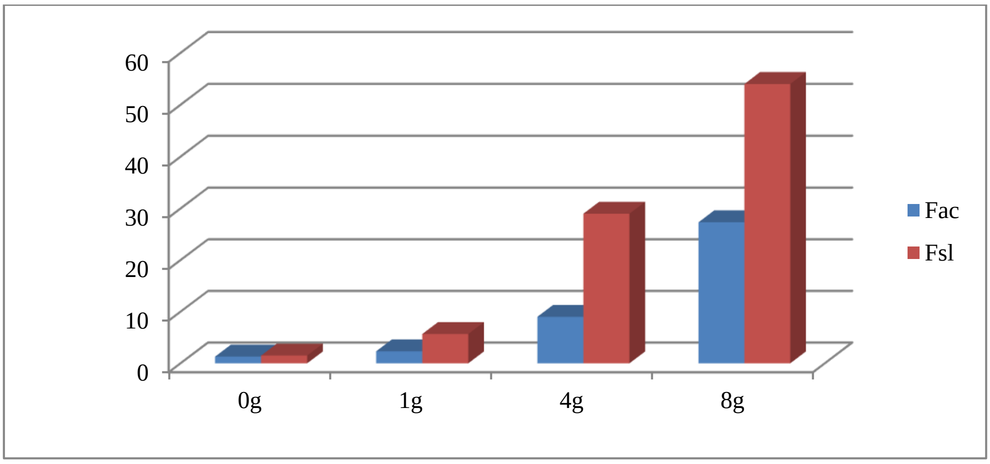


**Fig. 7 Toxicity of powders of *Ageratum conyzoides* leaves and roots of *Securidaca longipedunculata* at different doses.**

**Table 3 Larvicidal effect of powders of *Ageratum conyzoides* leaves and roots of *Securidaca longipedunculata*.**

| Treatment | Doses (g/100 g cowpea) | Average number of larvae |
|---|---|---|
| Fac | 1 | 9.32 |
| Fac | 4 | 5.66 |
| Fac | 8 | 2.66 |
| T0 | 0 | 16.32 |
| Rsl | 1 | 6.33 |
| Rsl | 4 | 4.66 |
| Rsl | 8 | 1.33 |

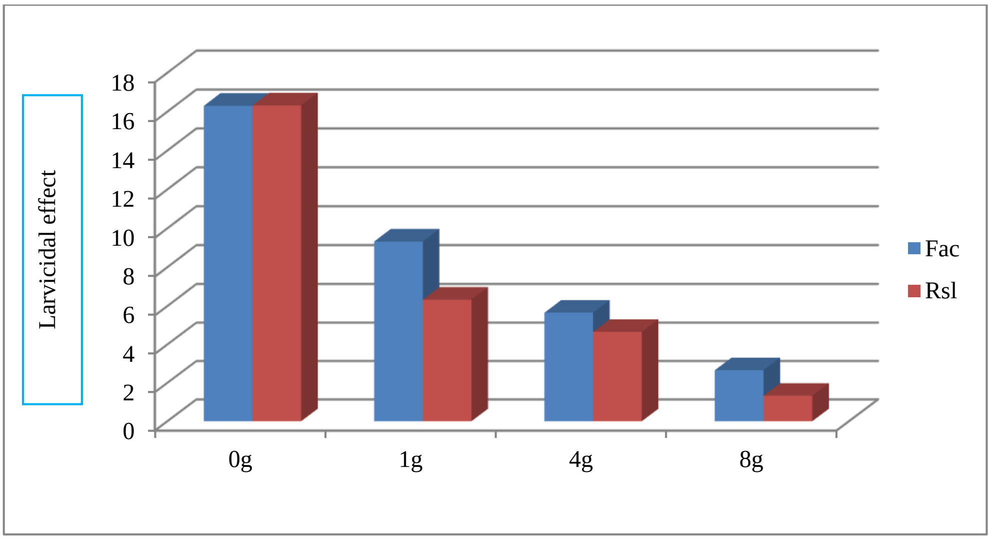


**Fig. 8 Larvicidal effect of the powder of the leaves of *Ageratum conyzoides* and the roots of *Securidaca longipedunculata* according to the different doses.**

The 8 g dose of Rsl significantly reduced the number of larvae of *Callosobruchus maculatus* weevils on cowpea grain. The number of larvae that emerged decreased between 6.33; 4.66 and 1.33 compared to Fac whose growth reduction is 9.32; 5.66 and 2.66 versus 16.32 (Witness).

The results of Table 3 show that the larvae undergo slight decreases with the use of *Ageratum conyzoides* powder, while they undergo marked decreases with the root powder of *Securidaca longepedunculata* which allowed recording a number of larvae of 6.33 from the 1 g dose (Fig. 8).

### *3.3 Impact of Powders from Ageratum conyzoides Leaves and Roots of Securidaca longipedunculata on Damage Caused by Callosobruchus maculatus*

At the end of the test, the results obtained show that in the control jars 9.33 ± 0.09 cowpea grains have a percentage weight loss (Table 4). However, in jars treated with *Ageratum conyzoides* leaves powder, the percentage weight loss was 8.66 ± 0.14; 4.66 ± 0.09 and 2.33 ± 0.14, at a dose of 1; 4 and 8 g per 100 g cowpea respectively. For powder treatments of the roots of *Securidaca longipedunculata*, it was 5.66 ± 0.23; 2.66 ± 0.18 and 1.33 ± 0.14 at the same dose. *Callosobruchus maculatus* damage is found to be concentrated in the control jars, showing a significant difference ($p < 0.05$) compared to the powder-treated jars of *Ageratum conyzoides* leaves and *Securidaca longipedunculata* root powder (Table 4).

The loss in weight of the seeds, is proportional to the number of unsold emerged, in fact, it is higher in the control samples with an average of 9.33 ± 0.09, then it undergoes decreases to 8.66 ± 0.14; 5.66 ± 0.23; 4.66 ± 0.09; 2.66 ± 0.18; 2.33 ± 0.14 and 1.33 ± 0.14 for powders of *Ageratum conyzoides* leaves and roots of *Securidaca longepedunculata*. According to Table 4, Figs. 9 and 10, the number of damaged seeds is less than 65% for the dose of 1 g and the weight loss at this same dose is at least 5% for *Securidaca longepedunculata* root powder, while the number of damaged seeds (greater than 70%) and weight loss of 1 g of *Ageratum conyzoides* are considerable compared to that of *Securidaca longepedunculata*.

### *3.4 Impact of Powders from Ageratum conyzoides Leaves and Roots of Securidaca longipedunculata on Germination*

The germination test performed at the beginning of the trial gave an average germination rate of 83%. When cowpea grains are in direct contact with Fac and Rsl leaf powder, the germination rate varies depending on the dose administered. Thus in jars treated with powder leaves of Fac at the dose of 1; 4; and 8 g per 100 g of cowpea each, the germination rate of cowpea grains is 25%, 40% and 71% respectively. In jars treated with Rsl root powder dose of 1; 4 and 8 g per 100 g cowpea each, the germination rate is 38%, 60%, 85% respectively. On the other hand, in control jars (T0) where insects are in direct contact with the grains, the germination rate is 11% (Table 5). In addition, some research programs on the germination power of grains claim that the decline in seed germination can probably be related to multiple causes, including heterogeneity, color, size, weight and chemical compositions [7].

Fig. 11 shows that the germination rate is greater than 80% from the dose 8 g, for *Securidaca longepedunculata* and it is greater than 60% for the same dose of *Ageratum conyzoides*. The powder extracted from *Securidaca longepedunculata* is the most favorable to fortify the germination of cowpea seeds since it has recorded averages above 80%.

**Table 4 Impact of powders from *Ageratum conyzoides* leaves and roots from *Securidaca longipedunculata* on damage caused by *Callosobruchus maculatus.***

| Treatment | Doses (g/100 g cowpea) | Damaged seeds in (%)/(standard deviation) | Percentage weight loss/(standard deviation) |
|---|---|---|---|
| Fac | 1 | 71.32 ± 1.24 | 8.66 ± 0.14 |
| Fac | 4 | 44 ± 0.81 | 4.66 ± 0.09 |
| Fac | 8 | 24.32 ± 1.24 | 2.33 ± 0.14 |
| To | 0 | 92 ± 0.81 | 9.33 ± 0.09 |
| Rsl | 1 | 61.32 ± 2.05 | 5.66 ± 0.23 |
| Rsl | 4 | 40 ± 1.63 | 2.66 ± 0.18 |
| Rsl | 8 | 10.33 ± 1.24 | 1.33 ± 0.14 |

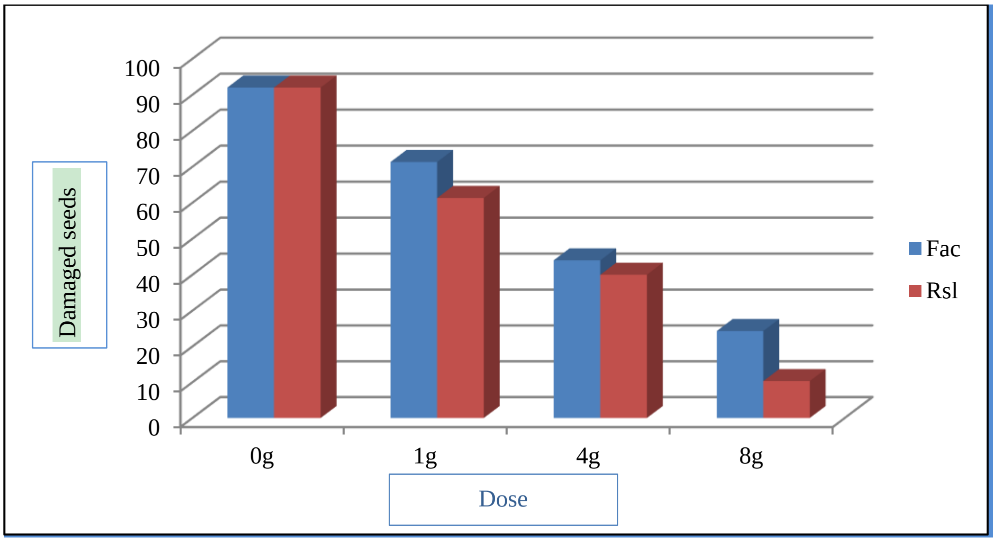


**Fig. 9 Impact of *Ageratum conyzoides* leaf powder and *Securidaca longipedunculata* roots on damage caused by *Callosobruchus maculatus* (damaged seeds).**

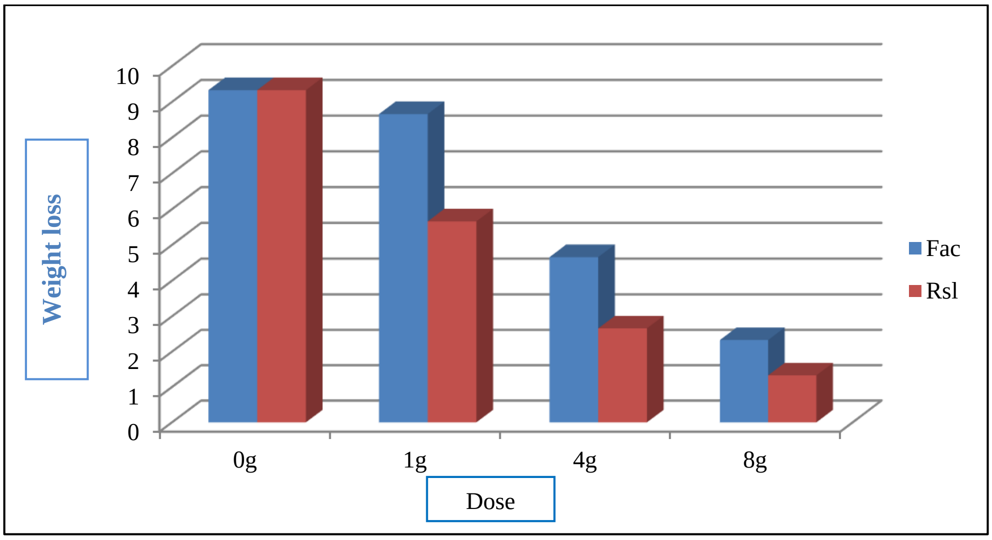


**Fig. 10 Impact of Ageratum conyzoides leaf powder and Securidaca longipedunculata roots on Callosobruchus maculatus damage (weight loss).**

**Table 5 Impact of *Ageratum conyzoides* leaf powder and *Securidaca longipedunculata* root powder on germination.**

| Treatment | Doses (g/100 g cowpea) | Germination percentage |
|---|---|---|
| Fac | 1 | 25 |
| Fac | 4 | 40 |
| Fac | 8 | 71 |
| To | 0 | 11 |
| Rsl | 1 | 38 |
| Rsl | 4 | 60 |
| Rsl | 8 | 85 |

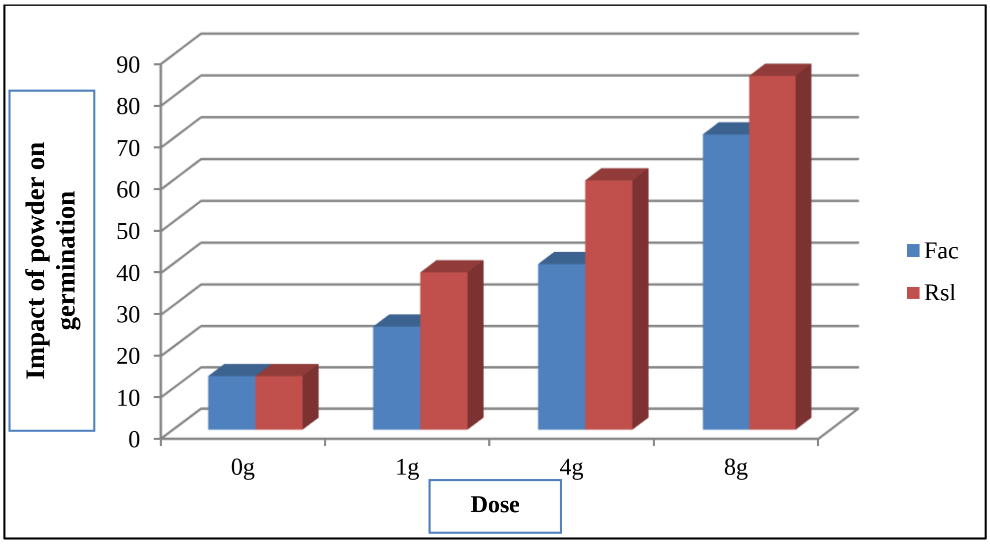


**Fig. 11 Impact of powders of *Ageratum conyzoides* leaves and roots of *Securidaca longipedunculata* on germination.**

## 4. Discussion

Numerous scientific studies published in the literature have highlighted the repellent effect of plant powders against stock insects. The leaf and root powders of the plants we tested had a contact toxic effect on *Callobosobruchus maculatus* adults. According to the results obtained, the longevity of *Callosobruchus maculatus* adults is inversely proportional to the dose of these plant powders administered. It was observed that over a period of 5 days, the dose of 8 g/100 g of *Securidaca longipedunculata* root powder administered resulted in 100% mortality of *Callosobruchus maculatus*. However, with the same dose of *Ageratum conyzoides* leaf powder, 95% mortality of the same insect was recorded. Thus, it appears that increasing the dose significantly reduces the lifespan of *Callosobruchus maculatus*. This result corroborates those found by other authors [[9, 11, 12]. The highest germination rate of treated cowpea grains was recorded in both cases with the 8 g/100 g dose, which can be explained by a low emergence of *Callosobruchus maculatus* as most of the seeds kept their albumen intact. This resulted in a germination rate of 85% for *Securidaca longipedunculata* and 71% for *Ageratum conyzoides* during 5 days of observation. These results are similar to those found by other authors [9-12]. This can be explained by the fact that *Securidaca longipedunculata* and *Ageratum conyzoides* have repellent and toxic actions on *Callosobruchus maculatus*. The actions of these two plants, *Securidaca longipedunculata* and *Ageratum conyzoides*, have inhibiting effects on the growth and development of the insect pest of cowpea grains intended for storage [1].

## 5. Conclusion

These samples of *Ageratum conyzoides* leaves and *Securidaca longipedunculata* root purchased in the different markets of Conakry were subjected to the evaluation of the insecticidal activity of their powder in the fight against *Callosobruchus maculatus* and their effect showed significant efficacy. This was observed according to the doses administered which vary from 1 to 8 g of the powder of each plant and the mortality of the bruchids was evaluated at 100% during 96 h of exposure of *Callosobruchus maculatus* to these bio-insecticides. In addition, with the same doses administered, the rate of damaged cowpea seeds was relatively zero after 4 months of storage. Thus, it is clear that powders of *Ageratum conyzoides* leaves and roots of *Securidaca longipedunculata* inhibit the development of bruchids and significantly reduce the emergence of adults of *Callosobruchus maculatus*.

Germination tests have shown that powders of *Ageratum conyzoides* leaves and roots of *Securidaca longipedunculata* have no negative effect on the germination power of cowpea grains. Therefore, they could be considered as excellent bio-insecticides that socio-professional strata (farmers, warehouse workers) can use in the fight against cowpea insects intended for storage.

## Conflicts of Interest

We, the authors of this work, declare that there is no conflict of interest regarding its publication.

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