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RESEARCH PAPER

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Insecticidal efficacy of aqueous plant extracts against lepidopteran larvae infesting maize in the Sudano-Guinean savannah zone of Northern Cameroon

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Abstract

Insect infestation constitutes a major constraint to maize production in the tropics and subtropics. To control pest insects, chemical insecticides are widely applied. This has become challenging as insects have developed resistance to these chemicals, in addition to the rising toxicity to humans and the negative environmental impacts. This situation has prompted the need for the development of safer, more effective and sustainable plant-based pest management products. Therefore, the insecticidal potentials of aqueous extracts from the dried pulverized leaves of Calotropis procera (C.p.), Callistemon rigidus (C.r.) and Plectranthus glandulosus (P.g.) were assessed against lepidopteran larvae infesting maize plants for the 2018 and 2019 cropping seasons at Ngaoundere in the Sudano-Guinean Savannah zone of northern Cameroon. Treatments included Cypercot® (Cy) as reference insecticide (positive control), phyto-insecticides (C.p., C.r., P.g.) and no insecticide (Co) (negative control). The different products were applied early in the morning (between 6:00 h and 8:00 h) 50 days after sowing, with distinct manual gauge sprayers and, repeated every two weeks until flowering. Four dominant species of lepidopteran insects (Spodoptera frugiperda, Eldana saccharina, Busseola fusca and Sesamia calamitis) were considered in the field experiment. All tested phyto-insecticides significantly reduced the larval densities of Spodoptera frugiperda (28.88%), E. saccharina (61.6%) and B. fusca (22.85%). In addition, these phyto-insecticides substantially diminished the damage caused on leaves (30%), stems (25.86%) and cobs (24.7%) and, somewhat increased maize yield performance across the two cropping seasons. The beneficial effects of the tested phyto-insecticide products increase over time. Based on our results, these phyto-insecticides could be considered as potential natural insecticides for the management of lepidopteran larvae infesting maize plants in the field.

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Introduction

Agriculture is the most important business in Africa, with about 60% of people earning a living from it (Abate *et al.*, 2000; FAO, 2011). In Cameroon, maize is one of the main cultivated cereals and also the most important staple food for many people (MINADER, 2009). However, maize production is often affected by a variety of diseases, weeds, cattle and pests (DRADER, 2013). The pests include insects, birds and rodents with insects being the most important (Ntambo *et al.*, 2015). Insect pests of maize include leaf hoppers, grass hoppers, army worms, aphids and weevils as well as moths (Lepidopteran larvae), which is the dominant group devastating the maize crop (Ndemah *et al.*, 2001; Ntambo *et al.*, 2015).

The currently widely used chemical control methods for the control of the lepidopteran larvae attacking cereals in Africa have become problematic (Charles et al., 2015). Chemical insecticides based on synthesized pyrethrinoïd (Cypermethrin like Deltamethrin, Lambda-Cyhalothrin, Betacypermethrin and Alpha-Cypermethrin like Cyfluthrin) used to control pests can cause several problems, such as environmental contamination, increase pest control costs and the death of the natural enemies of these pests. More so, they negatively affect the health of humans (ICIPE, 2000; Clive, 2003). In addition, the use of plantbased extracts in crop protection is being investigated in different parts of the world (Elizabeth et al., 2021). Botanical insecticides are increasingly becoming attractive alternatives to synthetic insecticides for pest control (Cox, 2002, Elizabeth et al., 2021). They are assumed to be more biodegradable and specific and thus pose less problems to the environment and humans (Addis, 2016). Unlike synthetic insecticides, botanical insecticides are less likely to result in the development of resistance in the insect pest (Elizabeth et al., 2021). Calotropis procera, Callistemon rigidus and Plectranthus glandulosus species were revealed as plants with insecticidal potentials. The larvicidal activity of Calotropis procera extracts was demonstrated by Verma et al. (1989), Jahan et al. (1991), Abbassi et al. (2003) and Ahmed et al. (2006). The bioactivity of Callistemon rigidus and P. glandulosus, were shown against stored product insect pests (Nukenine et al., 2010, 2011; Goudoum *et al.*, 2012, 2013; Danga *et al.*, 2014). It is therefore imperative to develop and promote plant-based insecticides to prevent the destruction of maize plants, as alternatives to environmentally devastating synthetic chemicals.

In view of improving maize productivity in the Guinean savannah agro-ecological zone of Cameroon, this study aimed at circumventing detrimental effects of lepidopteran larvae on maize plants through the promotion of the use of phyto-insecticides. Specifically, the field efficacy of aqueous extracts from the pulverized leaves of *Calotropis procera, Callistemon rigidus* and *P. glandulosus* against lepidopteran larvae infesting maize plants were assessed, as well as their influence on plant damage and crop yields.

Materials and methods

Field experiments were carried out during two cropping seasons from July to December of 2018 and 2019 in the Sudano-Guinean agro-ecological zone of Cameroon (Ngaoundere, Adamawa, Northern part), located at Dang-Malo, Ngaoundere III subdivision, with the geographic coordinates: $07^{\circ}27'09.0$ " N; $13^{\circ}32'57.3$ " E; altitude 1096 ± 1 masl. The Shaba maize variety, collected from the Institute for Agricultural Research and Development (IRAD) at Wakwa, Ngaoundere was used.

Experimental design and treatments

The field trial was laid out in a completely randomized block design with four replications. The blocks were 44 x 4 m² in size, with plot dimensions 8 x 4 m² separated 1 m from each other. According to PNAFM (2012), two maize grains were sowing hole within and between row spacing were 0.80 m and 0.25 m respectively. Maize plants were reduced to one plant at 21 days after sowing (DAS). Treatments were: the synthetic insecticide Cypercot® (Cy), botanical extracts from Calotropis procera (C.p.), Callistemon rigidus (C.r.), Plectranthus glandulosus (P.g.) and control plot (Co) without treatments as negative control. In each plot, maize plants were treated with fertilizers as follows: NPK 20-10-10 (650g/plot) and urea (160g/plot) at 32 DAS, and by urea (650g/plot) only at flowering.

Formulation of insecticide products and their application

Aqueous extracts of the pulverized dried leaves of from C.p., C.r. and P.g. were used in this work. The process of extracting bio-insecticides was carried out using the methods of AGROBIO 47 (2012) and Raveloson (2015) modified as follows: the concentrate was obtained by macerating 25 g of powder of the leaves from the different plants in one litter (1L) of water. The product was filtered twice (with 0.7 mm mesh sieve and with Terylene linen), after thirty-four hours in maceration. The concentrate resulting from the maceration was then diluted to 10% with water, for a working concentration of 2.5g/L.

The synthetic insecticide solution based on cypermethrin (Cypercot[®]) was obtained following the manufacturer's specifications (diluting 12 mL of Cypercot[®] in 10 L of water). The resulting insecticides were sprayed on maize plants, using Act Line- 2 L branded manual sprayers between different products, early in the morning 6:00 h and 8:00 h, 50 DAS, and repeated every two weeks until flowering. A separate sprayer was used for each insecticide.

Sampling and data collection

Because of the fragility of maize plants and their particular morphology, sampling methods were random for plant removal and stem dissection (Dabiré-Binso, 1980). This allowed the precise determination of the larval populations of the lepidopteran insects. The sampling was done once every two weeks until harvest.

Sampling began one week after the first spray; each plot was divided into three parts: two external parts (2nd to 11th and 22nd to 31st plant in each row) and one central part (12th to 21st plant in each row) as indicated by Gomez and Gomez (1984). In each external part, four maize plants were randomly selected and observed for the presence of different lepidopteran species and damage symptoms on the different plant parts (leaves, stems and cobs). Then, each plant was uprooted and dissected to determine the larval number in the stems, the actual number of internodes attacked and the gallery lengths tunnelled by the larvae.Each species of larvae was identified with the help keys of Moyal and Tran (1989) and, the field guide of crop pests by Alejandro (1988). Color photographs accompanied by a description of the damage symptoms, the larval morphology, the life cycle of the different species of pests as well as the plant they attacked and the phenological stage of the latter helped in the identification.

Determination of the population density and dynamics of lepidopteran larvae

The average larval population of lepidopterans in different treatments and their variations during sampling periods were assessed at 57 DAS, 71 DAS, 85 DAS, 99 DAS and 113 DAS.

Assessment of damaged plant parts

A total of 40 plants randomly taken per plot at 2 weeks interval (8 plants per sampling date) were carefully examined and the different damage symptoms on each plant part were noted. The damage to the different plant parts (leaves, stems or cobs) were assessed and the extent of damage to each part (percentage of leaves, stems and cobs damage) was made noted during the last sampling (5th sampling).

Stem damage

Two main stem damage parameters were recorded: damage on stems (internodes attacked) and damage in stems (galleries length).

> Percentage of internodes attacked

All internodes attacked per plant were counted, and the percentage in each treatment as well as their variation were determined by using the following formulae:

$$M_{Ia} = \frac{Nt}{No} *\%Ea$$
 and $\%Ea = \frac{n}{Ni} * 100$

%M_{Ia}: mean percent of internodes attacked, n: number of internodes attacked Nt: total number per plot, Ni: total number of internodes No: number of plants observed and, observed per plant %Ea: percentage of internodes attacked per plant

➤ Gallery Length (cm)

Following each sampling, plants from different plots were dissected to observe the galleries (gallery depth). Each plant was dissected, from the terminal bud to the last underground node. Using a graduated ruler, different galleries were measured along each plant and summed.

Crop yield

Yield was assessed by weighing the dried seeds for all stored cobs from each plot. Yield was recorded in gram (g) for different treatments (g/treatment) and estimated in kilograms per hectare (Kg/Ha). The different yields were obtained by weighing the dried seeds using an electronic scale (SF-400 electronic kitchen scale; max: 7Kg and d: 1g).

Statistical analysis

The statistical analysis was carried out using IBM SPSS statistics 20.0 software. ANOVA was used to compare larval densities and damaged plants between treatments. The Turkey test was used to separate different treatments, and the student t test for comparisons between two means. The various graphs were done using Excel (Microsoft Office 2013 software).

Results

Insecticidal efficacy of aqueous plant extracts on the larval population density of lepidoptera

In this ecological experiment to control lepidopteran larvae attacking maize plants, a diversity of pest species was determined. According to their consistency, plant parts damages and the larval density during the sampling period, four different larval species were found to be dominant: *Spodoptera frugiperda, Eldana saccharina, Busseola fusca* and *Sesamia calamitis.* The average larval densities of lepidopteran per maize plant in different insecticide treatments is presented in fig. 1.

Insecticidal efficacy of aqueous plant extracts on the larval density of Spodoptera frugiperda

Phyto-insecticide treatments have effectively reduced the larval density of *Spodoptera frugiperda* during the 2018 and 2019 cropping seasons. In 2018, insecticide treatments did not significantly reduce (P= 0.25) the larval density compared to the negative control. In addition, phyto-insecticides induced lower larval densities than the Cypercot. Among phytoinsecticide treatments, *P. glandulosus* extract had the most pronounced effect on larvae density with 44% reduction. Despite a 20% reduction in the larval population of *Spodoptera frugiperda*, *Calotropis procera* extract was the least effective phyto-insecticide. In 2019, all phyto-insecticide treatments applied to maize plants significantly reduced (P = 0.05) the larval density of *Spodoptera frugiperda* compared to negative control and Cypercot. Of all phyto-insecticide treatments, *Callistemon rigidus* extract was most effective with 77.78% reduction in larval density compared to *Calotropis procera* (40.75%) and *P. glandulosus* (66.67%). In general, the larval density of *Spodoptera frugiperda* was lower during the 2019 cropping season compared to 2018.

Insecticidal efficacy of aqueous plant extracts on the larval density of Eldana saccharina

Insecticide treatments did not significantly reduce (P = 0.57) the larval density of *E. saccharina* in 2018 compared to negative control. The synthetic insecticide Cypercot contributed to the total reduction of the larval population of this species. With 70.59% reduction of larval density, P. glandulosus extract was the most effective phyto-insecticide. Callistemon rigidus extract was the least effective phyto-insecticide with a 29.42% reduction in larval density of E. saccharina. In 2019, all insecticide treatments significantly reduced (P = 0.005) the larval density of E. saccharina (61.6%) compared to negative control. Cypercot completely reduced the its density by 81.82%, Callistemon rigidus extract was the best phytoinsecticide while, P. glandulosus extract was the least. The larval density of E. saccharina was lower during the 2018 cropping season compared to 2019.

Insecticidal efficacy of aqueous plant extracts on the larval density of Busseola fusca

In relation to the larval density of *B. fusca* per maize plant, all insecticide treatments did not significantly reduce larval density in 2018 compared to negative control (P = 0.20). The Cypercot further reduced the larval density of *B. fusca* by 83.18%. *Callistemon rigidus* treatment however, had a higher larval density than the negative control proceeded by *Calotropis procera* treatment. *P. glandulosus* extract was the most effective phyto-insecticide with a 10.52% reduction in the larvae density than others. In 2019, insecticide treatments significantly reduced the larval density of *B. fusca* with 22.85% reduction compared to negative control. The synthetic insecticide contributed to the total reduction in the larval density of this species. *Calotropis procera* extract was the most effective phyto-insecticide with 40% reduction in larval density compared to *P. glandulosus and Callistemon rigidus* extracts, which had higher larval densities same y as the negative control, respectively. The larval density of *B. fusca* was lower in 2019 cropping season than in 2018.

Insecticidal efficacy of aqueous plant extracts on the larval density of Sesamia calamitis

The different insecticide treatments did not significantly reduce (P = 0.11) the larval density of *Sesamia calamitis* in 2018 compared to negative control. Among phyto-insecticide treatments, *Callistemon rigidus* extract had a more pronounced effect on the larval population of *Sesamia calamitis* than others.



Fig. 1. Variation of larval density of dominant species as affected by insecticide treatments in 2018 and 2019.

Co: Control; C.p.: *Calotropis procera*; C.r.: *Callistemon rigidus*; P.g.: *Plectransthus glandulosis*; Cy: Cypercot.

Bars affected with the same letters are not significantly different to 5% (Tukey's test).

It reduced the larval density by almost 54%. Cypercot completely reduced its density by 68.29%. The other phyto-insecticides *Calotropis procera* and *P. glandulosus* had higher larval densities than the negative control. In 2019, similar observations to those of 2018 were noted.

The synthetic insecticide helped in further reducing (80%) larval density of *Sesamia calamitis*. All phytoinsecticides performed less effectively than the negative control with *Callistemon rigidus* extract being the least effective phyto-insecticide on these larvae. The larval density of *Sesamia calamitis* was lower during the 2018 cropping season compared to 2019.

Insecticidal efficacy of aqueous plant extracts on the larval population dynamics of lepidoptera

The variations of larval population densities of lepidoptera were more or less influenced by the different insecticides treatments (Figs. 2-5).

Insecticidal effects of plant extracts on the larval population dynamics of Spodoptera frugiperda

The larval population dynamics of Spodoptera frugiperda were most influenced by different insecticides during sampling periods. Fig. 2 shows different variations in the average larval density per maize plant in different insecticide treatments in 2018 and 2019. The larval population of Spodoptera frugiperda was the most affected in 2018 by phytoinsecticides compared to the synthetic insecticide that recorded the highest density, except for the first sampling. Negative control showed a continuous increase during the sampling period compared to bioinsecticide treatments with stable densities. In 2019, the larval population of Spodoptera frugiperda was also positively influenced by the different phytoinsecticides compared to negative control and synthetic insecticide that recorded the highest larval densities.

Insecticidal effects of plant extracts on the larval population dynamic of Eldana saccharina

Fig. 3 shows variations in the average larval density of *E. saccharina* per maize plant in different insecticide treatments during the sampling period of 2018 and 2019 maize cropping. It is distinguished by the fact that the larval densities of *E. saccharina* vary more or less in all treatments in 2018. Insecticide treatments did not have too much effect on the larval population of *E. saccharina* with the exception of *P. glandulosus* extract, the latter was the most effective phytoinsecticide. In 2019, a fluctuation in the larval population of *E. saccharina* in various treatments is

observed during the sampling period that got stabilized in the *Calotropis procera* and *Callistemon rigidus* treatments. The larval density of *E. saccharina* decreased in the *P. glandulosus* treatment during sampling periods. All insecticide treatments were effective compared to the negative control, but Cypercot was once again the most effective insecticide.



Fig. 2. Larval population dynamic of *Spodoptera frugiperda* as influenced by insecticide treatments in 2018 (A) and 2019 (B).

Co: Control; C.p.: *Calotropis procera;* C.r.: *Callistemon rigidus;* P.g.: *Plectransthus glandulosis;* Cy: Cypercot; DAS: days after sowing.

Insecticidal effects of plant extracts on the larval population dynamic of Busseola fusca

Changes in the average larval density of *B. fusca* per maize plant in different insecticide treatments during the 2018 and 2019 sampling periods are shown in fig. 4. The larval population of *B. fusca* increased in the various treatments during the sampling period in 2018. Phyto-insecticide treatments had a positive effect on the larval density of *B. fusca* during the sampling period, with the exception of *Callistemon rigidus* extracts that recorded a higher larval density during the first and fourth sampling. In 2019, the larval population of *B. fusca* was more or less influenced by phyto-insecticides versus negative control. High levels of larval densities were recorded in the first sampling for treatments with *Callistemon rigidus* and *P. glandulosus* extracts. All phyto-insecticide treatments stabilized the larval population of *B. fusca* from the second sampling compared to control. Cypercot was the most effective insecticide on the larval population of *B. fusca* compared to all phyto-insecticide treatments during the sampling periods of the two cropping years.



Fig. 3. Larval population dynamic of *Eldana saccharina* as influenced by insecticide treatments in 2018 (A) and 2019 (B).

Co: Control; C.p.: *Calotropis procera*; C.r.: *Callistemon rigidus*; P.g.: *Plectransthus glandulosis*; Cy: Cypercot; DAS: days after sowing.

Insecticidal effects of plant extracts on the larval population dynamic of Sesamia calamitis

Changes in the average larval density of Sesamia calamitis per maize plant in different insecticide treatments during the 2018 and 2019 sampling periods are shown in fig. 5. Larval density variations were more or less stable in all insecticide treatments over the two cropping years. In 2018 and 2019, the larval density of Sesamia calamitis reached a peak at the third sampling. Treatment with Callistemon rigidus extract recorded a high larval density (20 larvae per plant) in 2019, compared to 2018 (about 4 larvae per plant). Yet Callistemon rigidus extract was the most effective phyto-insecticide in 2018 compared to 2019. The Cypercot treatment was most effective on the larval population of Sesamia calamitis than phyto-insecticides during the sampling periods of the two cropping seasons.



Fig. 4. Larval population dynamic of *Busseola fusca* as influenced by insecticide treatments in 2018 (A) and 2019 (B).

Co: Control; C.p.: *Calotropis procera*; C.r.: *Callistemon rigidus*; P.g.: *Plectransthus glandulosis*; Cy: Cypercot; DAS: days after sowing.



 $-\blacksquare$ C0 $-\clubsuit$ - C.p. $-\blacktriangle$ - C.r. \cdots - P.g. $-\clubsuit$ · Cy

Fig. 5. Larval population dynamic of *Sesamia calamitis* as influenced by insecticide treatments in 2018 (A) and 2019 (B).

Co: Control; C.p.: *Calotropis procera*; C.r.: *Callistemon rigidus*; P.g.: *Plectransthus glandulosis*; Cy: Cypercot; DAP: days after planting.

Influence of aqueous plant extracts on the maize plants part damages caused by lepidopteran larvae The different insecticide treatments had influence on the maize plant parts by lepidopteran larvae (Table 1). Concerning the maize leaves, the insecticide treatments did not significantly reduce leaf damage (P = 0.13) compared to the negative control in 2018. Calotropis procera extract provided better protection of leaves compared to others (27.93% reduction). P. glandulosus treatment had the same level of damages as the negative control, which was the less effective in leaf protection. In 2019, similar observations were noted. The various phyto-insecticides and the synthetic insecticide at best protected the maize leaves (30%). The Callistemon rigidus treatment best protected the leaves (44% reduction) compared to the others. In addition, there was a very significant decrease in damages to maize leaves from 2018 to 2019 (t = 8.55, P < 0.001).

For the maize stems damaged by lepidopteran larvae during the maize cropping seasons, all insecticide treatments significantly (P = 0.03) reduced damages to stems in 2018 compared to the negative control. The Callistemon rigidus treatment better protected maize stems compared to others (11.88% reduction). With more damages to the stems compared to negative control, P. glandulosus treatment was the less effective in protecting the stems. In 2019, insecticide treatments significantly reduced damages to the stems (P = 0.004), with the exception of the P. glandulosus treatment, which recorded more damages to the maize stems. The synthetic insecticide better protected maize stems with 82% reduction in damages. As for the 2018 growing season, the Callistemon rigidus treatment was the best phyto-insecticide in protection of maize stems (29% reduction). In addition, there was a significant decrease in stem damage from 2018 to 2019 (*t* = 4.59, *P* < 0.01).

With regard to the damages caused to the maize cobs by lepidopteran larvae influenced by different insecticide treatments during the experimental periods, there is no significant difference between the different insecticide treatments over the two years (P= 0.84 in 2018 and P = 0.69 in 2019). In 2018, *Calotropis procera* treatment has helped to induce better protection (21.37% reduction) of maize cobs against lepidopteran larvae than any other treatment. The *P. glandulosus* treatment and synthetic insecticide did not induced protection compared to the negative control. In 2019, all phyto-insecticides and synthetic insecticide induced fair protection (24.7% reduction) of maize cobs relative to the negative control. The *Calotropis procera* treatment induced better protection (58.79% reduction) to the maize cobs compared to others.

 Table 1. Plant parts damaged (%) of maize as affected by insecticide treatments during the 2018 and 2019 cropping seasons.

| | Damage (%) | | | | | | | | | |
|--------------------------|--------------------|--------------------|-------------|----------------------|--------------------|-------|--------------------|--------------------|------|--|
| Insecticides | Leaves | | | Stems | | | Cobs | | | |
| | 2018 | 2019 | t | 2018 | 2019 | t | 2018 | 2019 | t | |
| Control | $42.50 \pm$ | $31.25 \pm$ | 1.74 | $26.25 \pm$ | 25.63 ± | 0.11 | $8.75 \pm$ | $10.63 \pm$ | 0.49 | |
| | 5.70 | 5.77 | | 4.96 ^b | 4.84 ^b | | 4.54 | 4.08 | | |
| Calotropis procera | $30.63 \pm$ | $18.75 \pm$ | 1.47 | $23.75 \pm$ | $20.00 \pm$ | 0.56 | 6.88 ± | 4.38 ± | 1.16 | |
| | 5.62 | 4.40 | | 5.80^{ab} | 5.00^{ab} | | 4.00 | 2.27 | | |
| Callistemon rigidus | $35.00 \pm$ | $17.50 \pm$ | 2.30* | $23.13 \pm$ | $18.13 \pm$ | 0.81 | $8.75 \pm$ | $6.25 \pm$ | 0.84 | |
| | 5.01 | 4.48 | | 5.23^{ab} | 4.48 ^{ab} | | 3.64 | 3.45 | | |
| Plectranthus glandulosus | $42.50 \pm$ | $26.25 \pm$ | 1.85 | 31.88 ± | 26.88 ± | 0.78 | 13.13 ± | 9.38 ± | 0.69 | |
| | 6.17 | 4.43 | | 6.05 ^{ab} | 4.99 ^b | | 5.77 | 3.37 | | |
| Cypercot | $25.00 \pm$ | $15.63 \pm$ | 1.38 | $8.75 \pm$ | 4.38 ± | 1.16 | 13.13 ± | 9.38 ± | 1.00 | |
| | 2.56 | 4.33 | | 3.02^{a} | 2.08 ^a | | 5.55 | 3.83 | | |
| Means | $35.13 \pm$ | $21.88 \pm$ | 8.55^{**} | $22.75 \pm$ | 19.00 ± | 4·59* | $_{+}10.13 \pm$ | 8.00 ± | 2.04 | |
| | 2.52 | 2.15 | * | 2.37 | 2.09 | | 2.13 | 1.53 | | |
| <u>F</u> | 1.81 ^{ns} | 1.96 ^{ns} | | 2.78* | 4.11** | | 0.35 ^{ns} | 0.56 ^{ns} | | |

Means followed by same letter are not significantly different to P < 0.05 (Tukey's test). ns: non-significant, *: significant P<0.05, **: significant P<0.01, ***: high significant P<0.001.

Impact of aqueous plant extracts on and in maize stems damaged by lepidopteran larvae

Damages on maize stems (internodes damaged)

The assessment of internode damaged (holes) varied from one treatment to another (Table 2). Insecticide treatments significantly protected the internodes of maize stems compared to negative control (P =0.006). With the exception of the P. glandulosus treatment, which had more internodes attacked, all other phyto-insecticides offered better protection of the internodes of maize stems. However, the synthetic insecticide better protected stems compared to the phyto-insecticides with about 72% reduction of attacks. Calotropis procera extract was the phytoinsecticide that induced best protection of stems internodes compared to the others, with about 30% reduction of attacks. In 2019, insecticide treatments significantly (P = 0.002) protected stem internodes against lepidopteran larvae compared to the negative control. Of all the phyto-insecticide treatments, only Calotropis procera extract provided about 20% protection of internodes. Other phyto-insecticide treatments had more internodes attacked compared to negative control.

The synthetic insecticide protected the stems internodes better than all phyto-insecticides, with an 80% reduction of attacked internodes.

Damages in maize stems (Galleries length)

The average length of the galleries bored into maize stems by lepidopteran larvae is also shown in Table 2. The synthetic insecticide significantly protected maize stems (P < 0.00001) against lepidopteran larvae in 2018. All phyto-insecticides had larger galleries compared to the negative control.

In 2019, insecticide treatments significantly protected (P = 0.007) maize stems compared to the negative control. Only *Calotropis procera* treatment better protected maize stems against borers compared to others, with about 20% reduction in damage to the maize stems. The synthetic insecticide better protected maize stems than phyto-insecticides, with about 78% reduction in damage. In addition, there was a drastic decrease in the gallery length from 2018 to 2019 (t = 8.52; P < 0.001) with a significant reduction of damage in all bio-insecticide treatments

| Incontinidad | Internoo | les damaged (% |) | Galleries length (cm) | | | |
|--------------------------|-----------------------|-----------------------|--------|-------------------------------|----------------------|---------|--|
| Insecticides | 2018 | 2019 | t | 2018 | 2019 | t | |
| control | 31.20 ± 6.36^{b} | 20.71 ± 3.65^{b} | 1.26 | 13.65 ± 2.16^{b} | 6.65 ± 1.30^{b} | 2.63* | |
| Calotropis procera | 21.88 ± 4.78^{ab} | 16.83 ± 3.36^{ab} | 0.82 | $14.78 \pm 3.09^{\mathrm{b}}$ | 4.03 ± 1.00^{ab} | 4.03*** | |
| Callistemon rigidus | 29.29 ± 5.14^{b} | 23.39 ± 4.88^{b} | 0.99 | 18.23 ± 3.82^{b} | 5.43 ± 1.20^{ab} | 3.10** | |
| Plectranthus glandulosus | 32.66 ± 4.39^{b} | 26.07 ± 4.95^{b} | 0.93 | 24.62 ± 4.06^{b} | 5.40 ± 1.00^{ab} | 4.87*** | |
| Cypercot | 8.80 ± 4.02^{a} | 4.14 ± 2.81^{a} | 1.05 | 1.41 ± 0.55^{a} | 1.5 ± 0.44^{a} | 0.12 | |
| Means | 24.77 ± 2.36 | 18.23 ± 1.92 | 6.26** | 14.54 ± 1.46 | 4.60 ± 0.47 | 8.52*** | |
| F | 3.86** | 4.55** | | 7.89*** | 3.64** | | |

Table 2. Damages inflicted on maize internodes and galleries due as influenced by insecticides treatments during the 2018 and 2019 cropping seasons.

Means followed by same letter are not significantly different to P < 0.05 (Tukey's test). ns: non-significant, *: significant P<0.05, **: significant P<0.01, ***: high significant P<0.001.

Influence of aqueous plant extracts on maize yield

Maize crop yield was strongly affected by different insecticide treatments (Fig. 6). In 2018, *Calotropis procera* treatment yielded least (210 Kg.Ha⁻¹) compared to the negative control (375Kg.Ha⁻¹). *Callistemon rigidus* extract (452.17 Kg.Ha⁻¹) was the most effective with about 17% crop yield compared to all other insecticide treatments. There was no significant difference (P = 0.79) between the different insecticide treatments. In 2019, insecticide treatments did not have a significant impact (P =0.89) on maize yields.





Co: Control; C.p.: *Calotropis procera*; C.r.: *Callistemon rigidus*; P.g.: Plectr*ansthus glandulosis*; Cy: Cypercot

Bars affected with the same letters are not significantly different to P < 0.05 (Tukey's test).

The *Callistermon rigidus* treatment produced the lowest yield (404 Kg.Ha⁻¹) compared to negative control (535 Kg.Ha⁻¹). The *Calotropis procera* and *P. glandulosus*

extracts were the phyto-insecticides that induced the most production with *P. glandulosus* extract being better bio-insecticide (604.77Kg.Ha⁻¹), with 12% of crop yields induced. These crop yields induced by phyto-insecticide treatments were lower than the synthetic insecticide Cypercot (725.90Kg.Ha⁻¹).

Discussion

Different phyto-insecticides used as treatments for the improving maize production in this study have influenced the investigated parameters. As a result, all phyto-insecticides were more effective on *Spodoptera frugiperda* and *E. saccharina* larvae. *Callistemon rigidus* and *Plectransthus glandulosus* extracts were the least effective on *B. fusca* larvae in 2018 and 2019 respectively. For *Sesamia calamitis* larvae, all phyto-insecticides were less effective compared to the negative control, with the exception of *Callistemon rigidus* extracts in 2018.

The various phyto-insecticides were less effective than the Cypercot on the lepidopteran larvae with the exception of *Spodoptera frugiperda* larvae. Similar observation was reported by Phambala *et al.* (2020), Elizabeth *et al.* (2021) where the high larval mortality on *Spodoptera frugiperda* species were observed with using botanical insecticides. The effectiveness of phyto-insecticides on *Spodoptera frugiperda* larvae may be due to their anti-feeding activity. These results correspond to those of Girdhar *et al.* (1984), Verma *et al.* (1989), Jahan *et al.* (1991), Abbassi *et al.* (2003), and Ahmed *et al.* (2006) which demonstrate the larvicidal activity of *Calotropis procera*; the works of Naveed and Muhammad (2011), Danga et al. (2014) and Sumitra and Shiva (2014) concerning the larvicidal activity of Callistemon rigidus and; by Hazel et al. (2017) and Dessenbe et al. (2020a and 2020b) concerning the larvicidal activity of P. glandulosus. Of the different phyto-insecticides, P. glandulosus was the least effective. Indeed, this extract has more volatile, which would have reduced its adhesion on the plants. According to Ibrahim et al. (1999), adhesion is a factor that could improve the effectiveness of phyto-insecticides. The major efficacy of Calotropis procera is due to the colloidal latex it contains (Saha and Kasinathan 1963; Abbassi et al., 2004). In addition, recognition of volatile compounds emitted by host plants is a key factor in the development cycle of lepidopteran insects (Calatayud et al., 2014). To this end, the high efficacy of Callistemon rigidus extracts may be due to its high content of aromatic compounds characterized by a strong and persistent odor (Ndomo et al., 2009), which could lead to a disruption of the recognition of host plants by lepidoptera (Birkett et al., 2006). Also, the high efficacy of phyto-insecticides on Spodoptera frugiperda and E. saccharina larvae may be due to the fact that they are exposed to the different insecticide applications (Adiss, 2016). All larvae that feed on insecticide-soaked leaves may exhibit the various symptoms caused by the anti-feeding activity of these phyto-insecticides (Rao and Mehrotra 1977). On the other hand, the B. fusca and Sesamia calamitis larvae carry out most of their development cycle in the stems; this part of maize plant provides shelter for its and, as a result, they are less exposed to different insecticide treatments. The high efficacy of the Cypercot would be due to its wide action spectrum compared to different phyto-insecticides (Oparaeke et al., 2005; Samantha, 2007; Saxena et al., 2014).

Phyto-insecticides had similar activity on the population densities of lepidopteran larvae over the two years. All different phyto-insecticides used were less effective than the Cypercot on the larvae population density, except on the larval density of *Spodoptera frugiperda*. The larval population densities of lepidoptera depend on plant infestation

level. The larval number of *Spodoptera frugiperda* and *B. fusca* did not exceed 5 larvae per plant during the sampling period, unlike the *E. saccharina* and *Sesamia calamitis* larvae, whose number of larvae reached about 15 to 20 larvae per plant. It is possible to encounter at least two of these lepidopteran species on the same plant. This report was illustrated by Calatayud *et al.* (2014). Also, it was difficult to encounter the larvae of these four major lepidoptera on the same maize plant; this could be due to the preference on infested parts, the nature of predation and the living space of each species.

Spodoptera frugiperda larvae were mainly found on the leaves, *E. saccharina* on the inflorescences and, *B.* fusca and Sesamia calamitis larvae in stems. Callistemon rigidus and *P. glandulosus* had the most fluctuating population dynamics respectively with *B.* fusca, Sesamia calamitis and Spodoptera frugiperda larvae. The constant effectiveness of the Calotropis procera extract would be due to its large adherence to plants and its less elimination by the rains than others phyto-insecticides.

Cypercot stabilized lepidopteran larval populations better than phyto-insecticides, with the exception to the larval density of *Spodoptera frugiperda*. Indeed, the high effectiveness of phyto-insecticides on the larval population density of *Spodoptera frugiperda* compared to the Cypercot is due at the anti-feeding properties. *Spodoptera frugiperda* larvae being the most exposed to the different insecticide treatments because larvae feed mainly on the leaves (Elizabeth *et al.*, 2021).

The percentage of leaf damages caused by lepidopteran larvae to maize plants was lower in 2019 than in 2018. The low damage recorded with *Calotropis procera* and *Callistemon rigidus* extracts might have been be due to their greater effectiveness on the leaf-feeding larvae during sampling periods. The highest losses of leaves and inflorescences were caused by *Spodoptera frugiperda* and *E. saccharina* larvae respectively (Elizabeth *et al.*, 2021). Cochereau (1991) found in his work that *Spodoptera frugiperda* is a more phytophageous species than other lepidopteran larvae. The maize stems can presented various damages; the mains are the holes (on the stem internodes) and galleries (in the maize stems). The maize stems damaged were mainly caused by B. fusca and Sesamia calamitis larvae. These observations were showed by Kfir et al. (2002) and Calatavud et al. (2014) in their various works. The holes on the internodes were found to be made by the early larval stages of Sesamia calamitis on the young plants (Harris and Nwanze, 1992) and, also by B. fusca larvae later (Calatayud et al., 2014), where they continually dig galleries. The existence of significant difference between different insecticide treatments during the two cropping seasons could be conditioned by climatic variations and the larval species that parasitizing the stems. Moreover, Sesamia calamitis larvae perform descending galleries, unlike B. fusca larvae, which perform ascending galleries. The galleries drilled could be conditioned by the internodes attacked and the larval population densities of lepidoptera.

When the larval population densities of lepidoptera became very large, these larvae attack maize cobs. The damage to the cobs during the two cropping seasons was less important. *Calotropis procera* extract was the phyto-insecticide that induced the most protective activity than others, almost same as the synthetic insecticide. The *B. fusca* and *Sesamia calamitis* larvae were the main species recorded on the cobs. These larvae left from the stems to the cobs.

Grain yields were higher in the 2019 cropping season compared to 2018. This could be explained by the low density of lepidopteran larvae in plots treated with different insecticides and the low damage inflicted to the plants. *Calotropis procera* and *Callistemon rigidus* extracts, although the most effective phyto-insecticides in protecting plants from lepidopteran larvae during this work, produced the lowest crop performances. With a broad action spectrum, Cypercot effectively reduced the larval population densities of lepidoptera compared to the phyto-insecticides (Oparaeke *et al.*, 2005; Samantha, 2007; Saxena *et al.*, 2014), so it would not be surprising if the latter induced a higher crop yield than phyto-insecticides. According to Dereval *et al.* (2014), phyto-insecticides are less effective than synthetic insecticides. This would justify the low densities recorded of lepidopteran larvae, as well as the damage caused to leaves and stems by these larvae in Cypercot-treated plots.

Conclusion

At the end of this study, which sought sustainable ways to improve the control of lepidopteran larvae attacking maize plants using aqueous extracts of Callistemon rigidus Calotropis procera, and Plectransthus glandulosus. We found that these phyto-insecticides greatly reduced larval population densities of lepidoptera and stabilized their dynamics in the field. These phyto-insecticides also helped to reduce larval damage to the plant parts (leaves, stems and cobs) and helped improve crop yields accordingly. They could therefore be proposed as substitutes of chemical insecticides in the monitoring of these maize pests as well as for environmental preservation and consumer health.

References

Abate T, van HA, Ampofo JKO. 2000. Pest management strategies in traditional agriculture: an African perspective. Annual Review of Entomology **4(5)**, 631-659.

Abbassi AK, Kadiri ZA, Ghaout S. 2003. Biological effect of alkaloids extracted from three plants of Moroccan arid area on the desert locust. Physiological and Entomology **28**, 232-236.

Abbassi AK, Kadiri ZA, Ghaout S. 2004. Activité biologique des feuilles de *Calotropis procera* (Ait. R. Br) sur le criquet pèlerin (*Schistocerca gregaria*, Forsk. 1775). Zool. baetica **15**, 153-166.

Addis TT. 2016. Assessment on Distribution, Biology and Management of Maize Stem Borer (*Busseola fusca* Fuller) in Ethiopia. Journal of Biology, Agriculture and Healthcare **6(5)**, 115-127.

AGROBIO 47 (Association de développement de l'Agriculture Biologique de Lot et Garonne). 2012. Guide technique des préparations à base de plantes 1-28. Ahmed AMU, Shin Z, Nabil HHB, Kamal M, Hao Z, Guo Y. 2006. Evaluation of insecticidal potentialities of aqueous extract from *Calotropis procera* Ait. Against *Henosepilachna elaterii* Rossi. Journal of Applied Sciences **6(11)**, 2466-2470.

Alejandro OC. 1988. Insectes ravageurs du maïs : guide d'identification au champ. Mexico, D. F.: CIMMYT 1-115.

Birkett MA, Chamberlain K, Khan ZR, Pickett JA, Toshova T, Wadhams LJ. 2006. Electrophysiological responses of the lepidopterous stemborers *Chilo partellus* and *Busseola fusca* to volatiles from wild and cultivated host plants. Journal of Chemical Ecology **32**, 2475-2487.

Calatayud PA, Le Ru BP, van-den-Berg J, Schulthess F. 2014. Ecology of the African Maize Stalk Borer, *Busseola fusca* (Lepidoptera: Noctuidae) with Special Reference to Insect-Plant Interactions. Insects **5**, 539-563.

Charles AOM, Toby JAB, John AP, Zeyaur RK. 2015. Ecological management of cereal stem borers in African smallholder agriculture through behavioural manipulation. Insects and ecosystem services special issue; Ecological Entomology **40(1)**, 70-81.

Clive J. 2003. Global review of commercialized transgenic crops. Agri-Biotech. Applications **84(3)**, 20-23.

Cochereau P. 1991. Les travaux de recherché français en zone tropicale sur les noctuelles et pyrales du maïs : synthèse bibliographique. L'agronomie tropicale **44(2)**, 137-142.

Cox C. 2002. Pyrethrins/Pyrethrum Insecticide Factsheet. Journal of Pesticide Reform **22(1)**, 14-20.

Dabiré-Binso L. 1980. Biologie et écologie d'*Eldana* saccharina Walker (*Lepidoptera Pyralidae Galleriinae*) foreur du maïs en Côte d'Ivoire et inventaire des autre Lépidoptères foreurs du maïs. Paris: ORSTOM, 1-206. **Danga YSP, Esimone CO, Nukenine EN.** 2014. Larvicidal and phytochemical properties of *Callistemon rigidus* R. Br. (Myrtaceae) leaf solvent extracts against three vector mosquitoes. Journal of Vector Borne Diseases **51**, 216-223.

Dereval J, Krier F, Jacques P. 2014. Les biopesticides, compléments et alternatives aux produits phytosanitaires chimiques (synthèse bibliographique), Biotechnology Agronomy and Society Environment **18(2)**, 220-232.

Dessenbe T, Christophe HL, Nukenine EN. 2020a. Bio-efficiency of methanolic extracts of leaves of *Plectransthus glandulosus* on mortality and offspring of *Sitophillus zeamais* F_1 in maize protection. Algerian Journal of Natural Products **8(2)**, 780-786.

Dessenbe T, Christophe HL, Nukenine EN. 2020b. Inhibitory activity, reducing loss and repulsive of methanolic extracts from sheets of *Plectransthus glandulosus* against *Sitophillus zeamais.* Journal of Fundamental and Applied Sciences **12(1)**, 377-388.

DRADER (Délégation Régionale de l'Agriculture et du Développement Rural). 2013. Rapport sur le Développement Economique: Région de l'Adamaoua exercice **2013**, 1-216.

Elizabeth N, Phophi N, Karim M, Ruth M, Paul K, Arielle R, Dennis N. 2021. Experiences and Perspectives on *Spodoptera frugiperda* (Lepidoptera: Noctuidae) Management in Sub-Saharan Africa. Journal of Integrated Pest Management **12(1)**, 1-9.

FAO (Food and Agriculture Organization). 2011. The Status of Food Insecurity in the World. Food and Agriculture Organization of the United Nations, Rome, Italy, 1-55.

Girdhar G, Devel K, Mittel PR, Vasudevan P. 1984. Mosquito control by *Calotropis* latex. Pesticides **18(10)**, 26-29. Goudoum A, Ngamo TLS, Ngassoum MB, Mbofungem. 2012. Biodegradation of insecticidal compounds of *Clausena anisata* and *Plectranthus glandulosus* essential oils applied as protectant on stored grains. Agricultural Journal 7, 165-171.

Goudoum A, Tinkeu LSN, Ngassoum MB, Mbofungem. 2013. Persistence of active compounds of essential oils of *Clausena anisata* (rutaceae) and *Plectranthus glandulosus* (Labiateae) used as insecticides on maize grains and flour. African Journal of Food, Agriculture, Nutrition and Development **13(1)**, 7325-7238.

Harris KM, Nwanze KF. 1992. *Busseola fusca* (Fuller), the African maize stem borer: a handbook of information. Information Bulletin 33. ICRISAT. Oxon, UK: CABI, 1-92.

Ibrahim L, Butt T, Beckett A, Clark SJ. 1999. The germination oil-formulated conidia of the insect pathogen *Metarhizium anisopliae*. Mycological Research **103**, 901-907.

ICIPE (International Centre of Insect Physiology and Ecology). 2000. Agricultural Entomology, Biological control of cereal stemborers in East and Southern Africa at: http:// nbo.icipe.org/agriculture/stemborers/default.html.

Jahan S, Maman A, Khan AR. 1991. Insecticidal effect of akanda (*Calotropis procera*) on *Tribolium confusum* Duval. (Coleoptera: Tenebrionidae). Bangladesh Journal of Zoology **19**, 261-268.

Kfir R, Overholt WA, Khan ZR, Polaszek A. 2002. Biology and management of economically important lepidopteran cereal stem borers in Africa. Annual review of Entomology **47**, 701-731.

MINADER (Ministère de l'Agriculture et Développement Rural). 2009. Annuaire des statistiques du secteur agricole: campagnes 2006&2007. Direction des Enquêtes et Statistiques Agricoles; AGRI-STAT N°15, 1-111.

Moyal P, Tran M. 1989. Étude morphologique des stades pré-imaginaux en zone des savanes de côte d'ivoire des lépidoptères foreurs du maïs. Annuals Society of entomology **25(4)**, 461-472.

Naveed M, Muhammad S. 2011. Biological screening of *Viola betonicifolia* Smith whole plant. African Journal of Pharmacy and Pharmacology **5(20)**, 2323-2329.

Ndemah R, Schulthess F, Korie S, Borgesmeister C, Cardwell KF. 2001. Distribution, relative importance and effect of lepidopterous borers on maize yield in the forest and mid-altitude of Cameroon. Journal of economic entomology **94**, 1434-1444.

Ndomo AF, Tapondjou AL, Tendonkeng F, Tchouanguep FM. 2009. Evaluation des propriétés insecticides des feuilles de *Callistemon viminalis* (Myrtaceae) contre les adultes d'*Acanthoscelides obtectus* (Say) (Coleoptera; Bruchidae). Tropicultura **27(3)**, 137-143.

Ntambo MS, Walter M, Mtaita T, Onesime MK. 2015. Effect of *Lepidopterous stemborers, Busseola fusca* (Fuller) and *Chilo Partellus* (Swinhoe) on Maize (*Zea mays* L) Yield: A Review. International Journal of Innovative Research & Development **4(10)**, 181-188.

Nukenine EN, Adler C, Reichmuth C. 2010. Efficacy of *Clausena anisata* and *Plectranthus glandulosus* leaf powder against *Prostephanus truncatus* (Coleoptera: Bostrichidae) and two strains of *Sitophilus zeamais* (Coleoptera: Curculionidae) on maize. Journal of Pest Science **83**, 181-190.

Nukenine EN, Katamssadan HT, Adler C. 2011. Comparative efficacy of Neem Azal and local botanicals derived from *Azadirachta indica* and *Plectranthus glandulosus* against *Sitophilus zeamais* on maize. Journal of Pest Science **84**, 479-486.

Oparaeke AM, Dike MC, Amatobi CI. 2005. Evaluation of botanical mixtures for insect pest management of cowpea plant. Journal of Agriculture Rural Development Tropical and Subtropical **106**, 41-48. Phambala K, Tembo Y, Kasambala T, Kabambe VH, Stevenson PC, Belmain SR. 2020. Bioactivity of common pesticidal plants on fall Armyworm Larvae (*Spodoptera frugiperda*). Plants **9**, 1-12.

NPSMS (National Program of Support for Maize Sector). 2012. Recueil technique sur le maïs. Ministry of agriculture and rural development, Cameroon, 1-14.

Rao PJ, Mehrotra KN. 1977. Phagostimulants andantifeedantsfromCalotropisgiganteanforSchistocercagregaria.IndianJournalofExperimental Biology 15, 148-150.

Raveloson RLH. 2015. Méthodes de lutte agroécologique contre les insectes nuisibles des cultures vivrières. GSDM, Université d'Antananarivo, 1-30.

Saha JC, Kasinathan S. 1963. Histamine action of the latex of *Calotropis gigantea*. Ait Arch. Int. Pharmacodyn. Ther. **143**, 78-89.

Samantha MC, Zeyaur RK, John AP. 2007. The Use of Push-Pull Strategies in Integrated Pest Management. Annual Review of Entomology **52**, 375-400.

Saxena HO, Tripathi YC, Ganesh P, Kakkar A, Mohammad N. 2014. Botanicals as biopesticides: active chemical constituents and biocidal action. Tropical forest research institute 219-240

Sumitra S, Shiva. 2014. Genus callistemon: an update review. World Journal of Pharmacy and Pharmaceutical Sciences **3(7)**, 291-307.

Verma BS, Verma KK, Sangwan NK, Dhindsa KS. 1989. Toxicity of some indigenous plant extract to root-knot, seed gal and citrus nematodes. Pesticides (Biombay) **23**, 25-27.