



Original Research Article

Field and laboratory testing of new insecticides molecules against *Spodoptera frugiperda* (J. E. Smith, 1797) infesting maize in Benin

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**Gustave BONNI¹,
Thomas A. HOUNDETE²,
Emmanuel SEKLOKA¹,
Romaine ASSOGBA
BALLE²
and
O.K. DOURO KPINDOU³**

¹Institut National des Recherches Agricoles du Bénin/ Centre de Recherches Agricoles Coton et Fibres (INRAB/CRA-CF), Abomey-Calavi, Bénin, 01 BP 715 Cotonou.

²Institut National des Recherches Agricoles du Bénin/ Centre de Recherches Agricoles Coton et Fibres (INRAB/CRA-CF), Laboratoire d'entomologie de Cana, Bohicon.

³Institut International d'Agriculture Tropical (IITA), 08 BP 0932 Tri Postal, Cotonou, Bénin.

*Corresponding Author E-mail : gustavebonni@yahoo.fr

Tel. (+229) 97 44 96 09

Fall armyworm, (FAW, *S. frugiperda*) is a prime noctuid pest of maize on the American continents. The pest has currently become a new invasive species in West Africa, especially in Benin. In order to limit the use of non-registered synthetic insecticides on food crops, with its consequences on man's health and environment, the effect of some semi-organic and botanical insecticides was evaluated on *S. frugiperda* on maize, in a comparative test with synthetic insecticides registered for food crops. The two synthetic insecticide formulations contained Lambda cyhalothrin 15 g/l-Acetamiprid 10 g/l and the other one of Indoxacarb 50 g/l. The other semi-organic and botanical formulations contained Spinetoram 120 g/l, Azadirachtin 0.35 g/l and Emamectin benzoate 19.2 g/l. The experiment was carried out in the Entomology Laboratory of Cana (Benin) and in the maize fields in Sékou (South area Benin), Alafiarou (North-South area Benin) and in Gogounou (North area Benin). The two synthetic insecticide formulations used were Lambdace 25 EC (Lambda-cyhalothrin 15g/l-Acetamipride 10 g/l) and Indoxan 50 EC (Indoxacarb 50g/l). The semi-organic formulations used were Radiant 120 SC (Spinetoram 120 g/l), Ema 19.2 EC (Emamectin benzoate 19.2 g/l) and the botanical formulation was Neem (Azadirachtin 0.35 g/l). The laboratory tests were conducted following a Fisher bloc design with an untreated control with four (4) replicates. Each replicate consisted of ten (10) caterpillars weighing between 35 and 45 mg and placed individually in a Petri dish. Mortality was assessed at 24, 48 and 72 hours after application of the different molecules. In the field, the design was a Fisher block with six (6) treatments and four (4) repetitions with elementary plots of eight (8) rows of 9m, of which six (6) rows were treated. Three phytosanitary applications were made just after the first observation at the 2-3 leaf stage. The insecticide applications were carried out every 14 days with a Solo 425 backpack device. The results showed higher mortality rates in laboratory with Emamectin benzoate (94.16 ± 2.6%), Azadirachtin (80 ± 9.3%) and Spinetoram (79.16 ± 4.91%). In the field, Spinetoram proved to be the most effective with a reduction rate of 70.77% of maize plants attacked, followed by Emamectin benzoate (54.86%) and Azadirachtin (36.36%). This study indicates that these active ingredients could be used in an integrated management system for *S. frugiperda* caterpillars.

Keywords: Efficiency, spinetoram, emamectin, azadirachtin, *Spodoptera frugiperda*, maize.

INTRODUCTION

Maize, *Zea Mays* L. is one of the most important cereals grown in Africa. Almost 25 million hectares are cultivated, about 14% of the cultivated land (FAO, 2014; FAO, 2017b). Maize occupies, with other food crops, a preponderant place in Beninese agriculture by contributing for more than 70% to the agricultural gross domestic product (GDP) (MAEP, 2010; Adégbola et al., 2011). Furthermore, it is marketed in neighboring countries and thus constitutes an important source of income (Soglohoun, 2000).

Despite the favorable conditions enjoyed by this crop, production of maize is experiencing a sawtooth evolution, which causes its food balance to fluctuate sometimes in worrying proportions. This poses threats both to food security and to producers' incomes. Some producers obtain less than 0.5 t / ha of yield against a potential of 3 to 5 t / ha depending on whether or not they have applied mineral fertilizer (Azontondé et al., 2010). Coupled with this critical situation, the producers were faced in 2016 to an invasion of the fields by defoliating caterpillars, the first alerts of which started in southern Benin (Municipality of Djidja) and the attacks ended up reaching the northern zone of the country. The identification of samples highlighted the species *S. frugiperda* (Goergen, 2016). This polyphagous Lepidoptera of the family Noctuidae is a legionary species native to South America which until now had remained confined to the neotropical and narcotic regions (Molina-Ochoa et al., 2003; Casmuz et al., 2010; Knaak et al., 2013). Recently, it has become a new invasive species in West Africa and in Benin in particular. The losses caused by *S. frugiperda* are in the order of 8.3 to 20.6 million tonnes of maize each year in the absence of effective control methods for the 12 largest maize producers in Africa (Day et al., 2017). The pest causes also significant yield losses in the fields of sugar cane, rice, sorghum, peanuts, soybeans, cotton (Casmuz et al., 2010). The caterpillars are present at all phenological stages and attack all the organs of the corn plant. To protect their crops, producers use unregistered or fraudulent insecticides on food crops with the risk of creating food poisoning (Soulé et al., 2015).

Studying the impact of insecticides used on survival, sex ratio, and reproduction of *Trichogramma pretiosum* Riley, Jander et al. (2013) have shown that among the insecticides tested; only triflumuron was selective for *T. pretiosum* and can be recommended with this parasitoid in fall armyworm management programs in corn.

Spinosad is highly effective against lepidopteran and dipteran pests, among others, and has proved to have a very favorable ecotoxicological profile (Rache, 2006). It has been recommended for the control of FAW larvae; its application with phagostimulants may reduce the quantity of active ingredient required for effective pest control. Spinosad (Tracer®) was formulated in maize flour matrix granules (Tamez-Guerra et al., 2018), It is active by ingestion, and shows lower activity by contact (Salgado et al., 1998). The degradation of spinosad in the environment occurs primarily by photo degradation; the half-life by

photolysis in soil is <10 days and ~1–2 days in water (Kollman, 2003; Pérez et al., 2007)

In the search for safe and effective insect pest control products against FAW, the objective of this study was to assess the sensitivity of FAW to new generation molecules in the laboratory and field.

MATERIALS AND METHODS

Laboratory Bioassays

The caterpillars of *S. frugiperda* were collected in corn field in Cana and Sékou area in the south of Benin and at Okpara at the North. The different samples of *S. frugiperda* obtained during collections were raised on artificial nutrient medium based on corn flour at the Cana entomology laboratory. The artificial diet used is composed of maize flour (142,5 g/liter), brewer's yeast (37.5 g/liter), wheat germ (37.5 g/liter), oil of maize (*Zea mays* L.) (1.25 ml/liter), sorbic acid (1.5 g/liter), ascorbic acid (12.5 g/liter), agar (16,2 g/liter), zelitrex (valaciclovir) (500 mg/liter) and rifampicin (0.05 g/liter) as described by Couilloud and Giret (1980). Larvae used were 3rd instar and weighing 40 mg. Their sensitivities to five (5) insecticide formulations, namely: Lambdace 25 EC (Lambda-cyhalothrin 15g / l-Acetamipride 10g / l) concentrated at 0.064 g/l, Indoxan 50 EC (Indoxacarb 50 g/l) concentrated at 0.047 g/l, Radiant 120 SC (Spinetoram 120 g/l) concentrated to 0.046 g/l, Plant Neem, prepared formulation (Azadirachtine 0.50 g/l) concentrated to 0.003 g/l and Ema 19.2 EC (Emamectin benzoate 19.2 g/l) concentrated to 0.018 g/l through soaking tests on corn husks were studied. The experimental device used was a Fisher's block including an untreated control with 4 replicates. Each repetition is made up of 10 caterpillars weighing between 35 and 45 mg and placed individually in a petri dish. Mortality was assessed at 24 h, 48 h and 72 h after application.

Field Tests

Field tests were conducted at the Permanent Experimentation Centers (CPE) of Sékou in the south area and at Alafiarou and Gogounou in the North. The device was a Fisher block with 6 treatments and 4 repetitions with elementary plots of 8 lines of 9 m, of which 6 lines were treated. The treatments compared were: 1) the untreated plot, 2) plots treated with Lambda-cyhalothrin 15g / l-acetamipride 10g / l at a dose of 1 l / ha, 3) plots treated with Indoxacarb 50 g / l at 0, 5 l / ha, 4) plots treated with Spinetoram 120 g / l to 0.15 l / ha, 5) plots treated with Azadirachtin 0.35 g / l at 2 l / ha and 6) plots treated with Emamectin benzoate 19.2 g / l to 0.5 l / ha. Treatment with Lambda-cyhalothrin 15g / l-acetamipride 10g / l was used as a control to evaluate the effect of insecticides on *S. frugiperda*'s caterpillars. Plants attacked were counted every week from the 2-3 leaf stage of the corn plant (about 15 cm high). Three (3) phytosanitary applications were

Table 1. Mortality of *S. frugiperda* strain of Cana in 2018

Insecticide	Mean \pm s.e (%)		
	24h	48h	72 h
Control (distil water)	0.00 \pm 0.00 b	0.00 \pm 0.00c	0.00 \pm 0 d
Lambda-cyhalothrine 15 g/l-Acétamipride 10 g/l (0.064 g/L)	2.50 \pm 2.50b	15.00 \pm 2.88bc	47.50 \pm 4.78c
Indoxacarb 50 g/l (0.047g/L)	20.00 \pm 4.08b	30.00 \pm 4.08 b	65.00 \pm 5.00b
Spinetoram 120 g/l (0.046g/L)	70.00 \pm 4.08a	80.00 \pm 7.07a	92.50 \pm 4.78a
Azadirachtin 0,35g/l (0.003g/L)	70.00 \pm 16.83a	87.50 \pm 12.50a	92.50 \pm 7.50a
Emamectin benzoate 19.2 g/l (0,018g/L)	87.50 \pm 7. 50a	90.00 \pm 5.77a	100.00 \pm 0.00 a
p<0.05	<.001	<.001	<.001

Values in parenthesis = concentrations tested in the laboratory; means within the same column followed by different letters are significantly different.

Table 2. Mortality of *S. frugiperda* strain of Tori (Sekou) in 2018

Insecticide	Mean \pm s.e (%)		
	24h	48h	72 h
Contol (distil water)	0.00 \pm 0.00 b	0.00 \pm 0.00 b	0.00 \pm 0.00 d
Lambda-cyhalothrine 15 g/l-Acétamipride 10 g/l (0.064g/L)	7.50 \pm 4.78b	17.50 \pm 8.54 b	35.00 \pm 11.90c
Indoxacarb 50 g/l (0.047g/L)	5.00 \pm 2.88b	27.50 \pm 4.78b	60.00 \pm 10.80abc
Spinetoram 120 g/l (0.046g/L)	27.50 \pm 17.96b	42.50 \pm 16.52b	80.00 \pm 7.07ab
Azadirachtin 0.35g/l (0.003g/L)	27.50 \pm 16.01b	35.00 \pm 17.55 b	52.50 \pm 19.31bc
Emamectin benzoate 19.2 g/l (0.018g/L)	75.00 \pm 8.66 a	82.50 \pm 6.29a	97.50 \pm 2.50a
p<0.05	0.001	0.001	<.001

Values in parenthesis = concentrations tested in the laboratory; means within the same column followed by different letters are significantly different.

Table 3. Mortality of *S frugiperda* strain of Okpara in 2018

Insecticide	Mean \pm s.e (%)		
	24 h	48 h	72 h
Control (distil water)	0.00 \pm 0.00b	0.00 \pm 0.00 b	0.00 \pm 0.00d
Lambda-cyhalothrine 15 g/l-Acétamipride 10 g/l (0.064g/L)	2.50 \pm 2.50 b	5.00 \pm 2.88b	17.50 \pm 4.78d
Indoxacarb 50 g/l (0.047g/L)	0.00 \pm 0.00 b	10.00 \pm 4.08b	42.50 \pm 13.77 c
Spinetoram 120 g/l (0.046g/L)	45.00 \pm 6.45a	62.50 \pm 2.50a	65.00 \pm 2.88 b
Azadirachtine 0.35g/l (0,003g/L)	40.00 \pm 7.07a	67.50 \pm 6.29a	97.50 \pm 2.50 a
Emamectin benzoate 19.2 g/l (0,018g/L)	45.00 \pm 6,45a	72.50 \pm 2.50a	8.00 \pm 2.88a
p<0.05	<.0001	<.0001	<.0001

Values in parenthesis = concentrations tested in the laboratory; means within the same column followed by different letters are significantly different.

carried out just after the first observation with the various insecticides. The applications were carried out with a Solo 425 backpack device and spaced 14 days apart.

Plants attacked by *S. frugiperda* have been counted. Data were analyzed using SAS 9.1.3 software. (SAS Institute Inc., 2004) and the means were separated using the Student-Newman-Keuls (SNK) test.

RESULTS

Bioassays results

The highest mortality rates after 72 hours were observed

on samples of Cana with Emamectin benzoate (100 \pm 0.00%) followed by Spinetoram (92.5 \pm 4.78%) and Azadirachtin (92.5 \pm 7.50%) (Table 1).

The Tori samples were more sensitive to Emamectin benzoate, Spinetoram and Indoxacarb with mortality rates of 97.50 \pm 2.50; 80.00 \pm 7.07 and 60.00 \pm 10.80 respectively (Table 2).

The Okpara strain showed similar results to those of Cana and Tori with higher mortality rates with Azadirachtin (97.50 \pm 2.50) and Emamectin benzoate (85.00 \pm 2.88), followed by Spinetoram (65.00 \pm 2.88) after 72 hours. The other active ingredients showed less than 50% of mortality (Table 3).

Gogounou strain showed higher mortality rates with

Table 4. Mortality of *S. frugiperda* strain of Gogounou in 2019

Insecticides	Mean \pm s.e (%)		
	24 h	48 h	72 h
Control (distil water)	0.00 \pm 0.00b	0.00 \pm 0.00 d	0.00 \pm 0.00d
Lambda-cyhalothrine 15 g/l-Acétamipride 10 g/l (0.064g/L)	2.50 \pm 2.50 b	15.00 \pm 2.88cd	27.50 \pm 4.78c
Indoxacarb 50 g/l (0.047g/L)	7.50 \pm 2.50 b	22.50 \pm 7.50c	47.50 \pm 11.08 b
Spinetoram 120 g/l (0.046g/L)	40.00 \pm 4.08a	47.50 \pm 4.78 b	62.50 \pm 4.78 b
Azadirachtine 0.35g/l (0.003g/L)	32.50 \pm 8.54 a	65.00 \pm 8.66 a	82.50 \pm 2.50 a
Emamectin benzoate 19.2 g/l (0.018g/L)	50.00 \pm 8.16a	72.50 \pm 2.50a	82.50 \pm 2.50 a
P	<.0001	<.0001	<.0001

Values in parenthesis = concentrations tested in the laboratory; means within the same column followed by different letters are significantly different.

Table 5. Mortality of *S. frugiperda* strain of Sekou

Objet	Percentage of plants infested	
	in 2018	in 2019
Non- treated	56.83 \pm 2.75 a	17.50 \pm 2.79 b
Lambda-cyhalothrine 15 g/l-Acétamipride 10 g/l (Control)	28.82 \pm 1.49 c	9.52 \pm 2.28 ab
Indoxacarb 50 g/l	23.37 \pm 1.14 c	6.91 \pm 1.85 a
Spinetoram 120 g/l	12.71 \pm 0.63 d	11.55 \pm 2.51 ab
Azadirachtine 0.35g/l	46.04 \pm 2.70 b	9.88 \pm 1.83 ab
Emamectin benzoate 19.2 g/l	29.04 \pm 2.018 c	9.64 \pm 2.48 ab
p<0.05	<.0001	0.04

Means within the same column followed by different letters are significantly different, one-way ANOVA (SNK)

Emamectin benzoate (82.50 \pm 2.50) and Azadirachtin (82.50 \pm 2.50) followed by Spinetoram (62.50 \pm 4.78) after 72 hours. Treatments with Lambda-cyhalothrin-Acetamipride and Indoxacarb showed the lowest mortality rates (Table 4). The laboratory results indicated a higher sensitivity of *S. frugiperda* to Emamectin, Spinetoram followed by Azadirachtin.

Field tests results

Results of Sékou center

In 2018, the percentages of maize plants attacked by *S. frugiperda* were lower on plots treated with Spinetoram 120 g/l (12.71 \pm 0.63%) against (56.83 \pm 2.7 for untreated plot, followed by plots treated with Indoxacarb, Emamectin benzoate which were not significantly different from that of the reference control (28.82 \pm 1.49%). The percentage of plants attacked was high on the plots treated with Azadirachtin (46.04 \pm 2.70 %) (Table 5).

In 2019, the percentages of maize plants attacked on the different plots were not statistically different from those of the control plots (Table 5).

Results of Alafiarou Center

In 2018, the percentages of maize plants attacked by *S. frugiperda* were lower on plots treated with Spinetoram

120 g / l (8.39 \pm 1.55%) and those treated with Emamectin benzoate (13.05 \pm 3.15%) against 37.01 \pm 4.06 for NT. The percentages of plants attacked on the plots treated with Azadirachtin and on those treated with indoxacarb 50 g / l although significantly different from that of NT, remain high (19.26 \pm 3.68 and 20.59 \pm 4.85% compared to 31.60 \pm 4.23%) (Table 6).

In 2019, the rate of attacked plants was high on the plots treated with Azadirachtin, with indoxacarb and on those treated with Lambdacyhalothrin 15 gl-Acetamidrid 10 g / l. These rates of plants attacked were not significantly different from that of NT (Table 6).

Results of Gogounou Center

The results of the Gogounou site during the two years of experimentation did not show a significant difference between the percentages of plants attacked on the different plots (Table 7). Population densities of *S. frugiperda* have been low.

Field results showed better control of *S. frugiperda* with Spinetoram 120 g/l at 0.15 l/ha, followed by Emamectin benzoate 19.2 g/l at 0.5 l/ha. Azadirachtin 0.35 g/l at 2 l/ha and Indoxacarb 50 g/l at 0.5 l/ ha have been shown to be equivalent and rank in third position. These active ingredients have shown reduction rates of plants attacked by *S. frugiperda* of 70.77% (Spinetoram 120 g/l), 54.86% (Emamectin benzoate 19.2 g/l) and 36.36% (Azadirachtin 0.35 g/l).

Table 6. Plants infested by *S. frugiperda* at Alafiarou

Objet	% plants infested	
	in 2018	in 2019
Non- treated	37.01 ±4.06a	12.14±2.23 a
Lambda-cyhalothrine 15 g/l-Acétamipride 10 g/l (Control)	31.60±4.23ab	14.94±2.40 a
Indoxacarb 50 g/l	20.59 ± 4.85bc	9.58±1.89 a
Spinetoram 120 g/l	8.39 ± 1.55c	1.84±0.73 b
Azadirachtine 0.35g/l	19.26 ± 3.68bc	9.58±2.05 a
Emamectin benzoate 19.2 g/l	13.05 ± 3.15c	3.21±1.42 b
p<0.05	<.0001	<.0001

Means within the same column followed by different letters are significantly different, one-way ANOVA (SNK)

Table 7. Plants infested by *S. frugiperda* at Gogounou in 2018 and 2019

Objet	%of infested plants	
	in 2018	in 2019
Non- treated	6.57±1.58	1.97±0.53
Lambda-cyhalothrine 15 g/l-Acétamipride 10 g/l (control)	7.31±1.73	3.85±1.16
Indoxacarb 50 g/l	6.48±1.73	2.71±0.86
Spinetoram 120 g/l	2.96±0.68	1.15±0.55
Azadirachtine 0.35g/l	4.99±1.43	2.61±0.99
Emamectin benzoate 19.2 g/l	3.33±0.79	1.35±0.56
p<0.05	0.14	0.19

DISCUSSION

The objective of this study is to assess the sensitivity of *S. frugiperda* to some molecules in the laboratory and to study the effectiveness of these insecticides in the field in order to reduce the use of all-purpose products not approved on food crops and on corn in particular. In the laboratory, our results showed a greater sensitivity of the pest to Emamectin benzoate, Spinetoram and Azadirachtin. However, in the field, the effect of the neem-based insecticide has decreased compared to its effect in the laboratory. Neem insecticides often lose their effectiveness after exposure to the weather a few hours after their application (Koul et al., 1990; Schumutterer, 1990; Shoil et al., 2005). The dose of 2 l/ha of the neem-based insecticide may seem low for the control of *S. frugiperda* as it was the case on *Helicoverpa armigera* on cotton cultivation (Bonni et al., 2018). Higher dose studies are to be investigated. Spinetoram and Emamectin benzoate, on the other hand, confirmed their effectiveness in the field with reduction rates of plants attacked by *S. frugiperda* of 70.77 and 54.86% respectively.

Spinetoram is a non-systemic insecticide derived from active biological substances (spinosyn) produced by the soil actinomycete *Saccharopolyspora spinosa*. It is semi-synthetic discovered during studies of modification of natural substances by Dow Agro Sciences. Spinetoram has a broad spectrum of action and can be used on several crops. It is applied in low doses and has a low impact on most beneficial insects (Mertz and Yao, 1990). It stimulates the nervous system of insects by modifying the binding sites

known by other classes of insecticides.

Emamectin benzoate is one of the new generation of insecticides. It is a semi-synthetic insecticide from the Avermectins family and belongs to the Action Committee for Insecticide Resistance, in class 6 mode of action. Avermectins come from the fermentation isolation of soil microorganisms (*Streptomyces avermitilis*). It is much more used for its low toxicity on natural enemies (Hewa-Kapuge et al., 2003; Bostanian et al., 2004; Gonzalez-Zamora et al., 2004; Dinter et al., 2009) and is therefore very compatible for integrated pest management programs. It acts by contact and by ingestion (Wing et al., 2004; Temple et al., 2009). This active ingredient confirmed his effectiveness in the field in this study. Similar results have been obtained in Senegal (PR-PICA, 2019) this active ingredient is used in combination with acetamiprid in the West African in cotton cultivation to fight against the Lepidoptera-Sucking Sucker complex (PR-PICA, 2017).

CONCLUSION

Spodoptera frugiperda has become a new invasive species in West Africa and in Benin in particular. This pest causes significant crop losses in the corn fields. To fight against this armyworm, producers use unlicensed insecticides or those intended to fight against cotton pests, with all the risks for human health and the environment. The search for new molecules based on microorganisms or of botanical

origin adapted to food crops, enabled us to identify Spinetoram 120 g/l and Emamectin 19.2 g / l which made it possible to significantly reduce the number of plants attacked.

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Conflict of Interests

The authors declare that there is no conflict of interests regarding the publication of the paper.

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