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INSETICIDAL OILS FROM AMAZON PLANTS IN CONTROL OF FALL ARMYWORM

INSETICIDAL OILS FROM AMAZON PLANTS IN CONTROL OF FALL ARMYWORM[1]

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ABSTRACT - The potential insecticidal of oils from southwestern Amazon plants against *Spodoptera frugiperda* (J. E. Smith, 1797) (Lepidoptera: Noctuidae) was investigated. Initial bioassays were performed with undiluted oils from 11 plant species. The efficacy of the oils was evaluated against eggs and third-instar caterpillars of *S. frugiperda*. The oils of *Copaifera* sp. (Leguminosae), *Orbignya phalerata* (Arecaceae), and *Carapa guianensis* (Meliaceae) displayed a high efficacy against the caterpillars and were used in subsequent concentration-response bioassays, at concentrations established through preliminary tests. The highest nonlethal concentrations of oils and the lowest lethal concentrations were calculated. A completely randomized design was adopted in both bioassays. The LC_{50} of the oils varied from 7.50 to 60.84% (v/v). *Copaifera* sp. oil had the highest toxicity and was 6.84-fold more toxic than *O. phalerata* oil and 8.11-fold more toxic than *Carapa guianensis* oil. In general, oils from *Copaifera* sp., *O. phalerata*, and *Carapa guianensis* were effective in controlling *S. frugiperda* caterpillars under laboratory conditions, and are good candidates for use in integrated management programs of corn pests.

Keywords: *Spodoptera frugiperda*. Botanical insecticides. *Carapa guianensis*. *Copaifera* sp. *Orbignya phalerata*.

ÓLEOS INSETICIDAS DE PLANTAS DA AMAZÔNIA NO CONTROLE DA LAGARTA-DO-CARTUCHO

RESUMO - Investigou-se o potencial inseticida de óleos vegetais de plantas ocorrentes na Amazônia Sul Ocidental sobre *Spodoptera frugiperda* (J. E. Smith, 1797) (Lepidoptera: Noctuidae). Inicialmente, foram realizados bioensaios utilizando-se óleos puros (sem diluições), oriundos de 11 espécies vegetais. A eficiência dos óleos foi avaliada para ovos e para lagartas de terceiro ínstar de *S. frugiperda*. Os óleos de *Copaifera* sp. (Leguminosae), *Orbignya phalerata* (Arecaceae) e *Carapa guianensis* (Meliaceae) apresentaram elevada eficiência para as lagartas de *S. frugiperda*, sendo, portanto utilizados nos bioensaios subsequentes de concentração-resposta. As concentrações de cada óleo, utilizadas nestes testes de concentração-mortalidade, foram estabelecidas por meio de testes preliminares. Estabeleceram-se as maiores concentrações dos óleos em que não ocorresse morte de insetos e as menores concentrações em que ocorresse a maior mortalidade. Com base nos dados obtidos, foram estabelecidos os intervalos definitivos. O delineamento inteiramente casualizado foi adotado em ambos os bioensaios. As CL_{50} variaram de 7,50 a 60,84% (v/v) entre os óleos. O óleo de *Copaifera* sp. apresentou maior toxicidade para as lagartas, sendo 6,84 vezes mais tóxico que o óleo de *O. phalerata* e 8,11 vezes mais tóxico que o óleo de *Carapa guianensis*. Em geral, os óleos de *Copaifera* sp., *O. phalerata* e *Carapa guianensis* são eficazes no controle de lagartas de *S. frugiperda*, sob condições de laboratório, destacando-se como candidatos a uso nos programas de manejo integrado das pragas do milho.

Palavras-chave: *Spodoptera frugiperda*. Inseticidas botânicos. *Copaifera* sp.. *Orbignya phalerata*. *Carapa guianensis*.

INTRODUCTION

Production from Brazilian agriculture, primarily grains, has been increasing, owing to investments in new planting and harvesting technologies (ALVES et al., 2012). According to the National Resource Company (Companhia Nacional de Abastecimento, CONAB), the estimated Brazilian production of grain in the 2015–2016 harvest will be 210.3 million tons, with corn representing 40.93% of total production, around 83,519.1 million tons (CONAB, 2016).

One of the primary factors in low corn production is the damage caused by insect pests, which can occur in the field as well as during storage (FARONI; SOUSA, 2006). Pest damage can compromise the grain yield and quality, causing serious losses for farmers (COPATTI; MARCON; MACHADO, 2013). The fall armyworm, *Spodoptera frugiperda* (J. E. Smith, 1797) (Lepidoptera: Noctuidae), is an important field pest, and the corn weevil, *Sitophilus zeamais* (*Motschulsky,* 1885) (Coleoptera: Curculionidae), is an important storage pest (CRUZ et al., 2013).

The fall armyworm (*S. frugiperda*) may attack corn crops from seedling emergence to cob formation. It can cause high losses in grain productivity because the plants have no resistance to the pest (HAAS et al., 2014). Insecticides are applied in excess of the recommended dose during harvest in some regions in an effort to control this pest (FIGUEIREDO; MARTINS-DIAS; CRUZ, 2006).

Spodoptera frugiperda is normally controlled with synthetic insecticides; however, the use of these products has caused environmental contamination, human poisoning, extinction of natural predators, higher numbers of pests (MARTINS et al., 2009; LIMA JÚNIOR et al., 2010), and selection of resistant insects, requiring increasingly higher doses of insecticides (ALVES et al., 2012). Therefore, the search for alternative insecticides has become increasingly important.

Plants produce substances to defend themselves against insects, pathogens, and microorganisms (SOUSA; FARONI; FREITAS, 2014; ALVES et al., 2012). Secondary metabolites with insecticidal properties include terpenoids and limonoids, among others (SARRIA et al., 2011). These compounds may have different modes of action on insects; they may be acutely toxic, repellent, or inhibit feeding, growth, development, and reproduction. Plant-derived insecticides are usually available as powders, extracts, or oils, which are easy to obtain (GUERRA et al., 2009; HAAS et al., 2014; SOUSA; FARONI; FREITAS, 2014).

Given the problems associated with the use of synthetic insecticides in the control of *S. frugiperda* and considering the high plant diversity of the Amazon, the objective of this study was to determine the potential insecticidal activity of oils from plants occurring in the southwestern Amazon.

MATERIAL AND METHODS

Selective bioassays were performed by exposing the eggs and third-instar caterpillars of *S. frugiperda* to 11 undiluted oils. The most efficacious oils were evaluated with concentration-response bioassays to establish the degree of toxicity.

The bioassays were carried out in the Laboratory of Entomology of the Federal University of Acre, Rio Branco Campus, under constant temperature (25 ± 2°C) and relative humidity (70 ± 5%), using biochemical oxygen demand-type incubators. A stock colony of *S. frugiperda* was established in the laboratory according to the methods of Bavaresco et al. (2002), from specimens from the Agroforestry Research Center of Acre (EMBRAPA/AC). The caterpillars were fed on a diet composed of ascorbic acid (5.1 g), sorbic acid (1.7 g), agar (15.0 g), distilled water (1,200.0 mL), beans (1,665.5 g), 10% formalin (5.0 mL), wheat germ (79.2 g), brewer's yeast (50.7 g), methyl parahydroxybenzoate (nipagin) (3.2 g), and inhibitor solution (5.0 mL). The diet was produced according to Kasten Júnior, Precetti and Parra (1978).

Oils were extracted from the seeds of *Astrocaryum aculeatum* G. Mey. (Arecaceae), *Oenocarpus bataua* Mart. (Arecaceae), and *Orbignya phalerata* Mart. (Arecaceae); fruit pulp of *Mauritia flexuosa* L. f. (Arecaceae) and *Caryocar brasiliense* Cambess. (Caryocaraceae); seeds from *Anadenanthera colubrina* (Vell.) Brenan (Fabaceae), *Carapa guianensis* Aubl. (Meliaceae), *Platonia insignis* Mart. (Clusiaceae), and *Pentaclethra macroloba* (Willd.) Kuntze (Fabaceae); leaves of *Vernonia polysphaera* (Asteraceae); and wood of *Copaifera* sp. (Fabaceae).

The oils of *Platonia insignis*, *Pentaclethra macroloba*, and *A. aculeatum* were purchased from producers in Santa Bárbara do Pará, Pará. The Pará municipality is located at 01° 13′ 26″ south latitude and 48° 17′ 22″ west longitude, on the borders of the Augusto Meira Filho PA-391 highway. The other oils were acquired from the Technology Foundation of the State of Acre (FUNTAC), from producers in the municipality of Rio Branco, Acre. The Acre municipality is located at 9° 58′ 29″ south and 67° 48′ 36″ west, at an altitude of 153 meters above sea level.

In the selective bioassays, each oil was tested on the eggs and third-instar caterpillars of *S. frugiperda*. The tests were performed in Petri dishes (5.0 cm diameter × 1.2 cm height) containing a filter paper disc moistened with 200 µL of the oil; the oil quantity was determined in preliminary tests. Four Petri dishes were used as replicates for each egg bioassay and contained a disc with 40 eggs/plate. Four groups of 10 Petri dishes were used for each of

the caterpillar bioassays, with only one caterpillar per plate to prevent cannibalism. A completely randomized design was used. Eggs and caterpillars were exposed to the oils for 24 h. Subsequently, the numbers of hatched and unhatched eggs and live and dead caterpillars were counted. The egg count was repeated daily until the fifth day, as established in the preliminary tests.

The toxicity of *Copaifera* sp., *O. phalerata*, and *Carapa guianensis* oils was subsequently determined through estimates of lethal concentrations for 50% of the insects (LC$_{50}$). Initially, preliminary tests were performed to estimate the highest oil concentrations in which there were no insect deaths and the lowest concentrations in which the highest mortalities occurred. Five concentrations were established for the definitive bioassays with *Carapa guianensis* oil (0, 25, 50, 75, and 100% v/v), six for *Copaifera* sp. (0, 5, 10, 15, 20, and 25% v/v), and six for *O. phalerata* oil (0, 20, 40, 60, 80, and 100% v/v). The tests were performed in Petri dishes (5.0 cm diameter × 1.2 cm height) containing a disc of filter paper moistened with 200 µL of each oil at the previously established concentrations. The same experimental units as the selective bioassays were used. The caterpillar mortality rate was evaluated after 24 h of exposure to the oils.

The efficacy of the selective bioassay treatments compared to the controls was calculated with Abbott's formula. Data were evaluated with an analysis of variance (ANOVA) and the Scott-Knott test, with a significance level of 5%. The analyses were conducted with the program SISVAR

(FERREIRA, 2011).

Bioassay mortality data to determine the toxicity of *Carapa guianensis*, *Copaifera* sp., and *O. phalerata* oils were submitted to a Probit analysis to generate concentration-mortality curves (PROC PROBIT; SAS INSTITUTE, 2011). The toxicity ratios (TR) were calculated as described in Robertson and Preisler (1992).

RESULTS AND DISCUSSION

A significant effect was observed for undiluted oils on *S. frugiperda* eggs (F$_{10;29}$ = 7.04; *P* < 0.001) and caterpillars (F$_{10;29}$ = 27.59; *P* < 0.001) (Table 1). The efficacy varied from 14.38 to 91.50% on the eggs and from 0 to 100% on the caterpillars. Most of the oils were similarly efficacious on eggs, although the *P. macroloba* and *O. phalerata* oils were less efficacious. The most efficacious oils against the caterpillars were obtained from *Copaifera* sp., *O. phalerata*, and *Carapa guianensis*.

The caterpillar *S. frugiperda* causes heavy economic losses in all crops that it attacks, so a highly and rapidly effective insecticide is desirable (ALVES et al., 2012). The *Copaifera* sp., *O. phalerata*, and *Carapa guianensis* oils were the most suitable in this study because they demonstrated moderate toxicity in 24 h. In general, botanical insecticide efficiency may be associated with direct effects (caterpillar mortality) and indirect effects (increasing vulnerability to parasitoids, predators, and entomopathogens) (KNAAK et al., 2012).

**Table 1**. Efficiency of undiluted oils (200 µL) on eggs and third-instar caterpillars of *S. frugiperda*.

Species	Efficiency (%) ± S.E.M.[1]	
	Eggs	Caterpillars
Copaifera sp.	68.0 ± 7.1 a	100.0 ± 0.0 a
Orbignya phalerata	14.4 ± 2.9 b	100.0 ± 0.0 a
Carapa guianensis	64.7 ± 6.4 a	97.50 ± 2.2 a
Caryocar brasiliense	81.1 ± 7.4 a	72.50 ± 9.9 b
Astrocaryum aculeatum	66.0 ± 8.8 a	70.0 ± 3.7 b
Anadenanthera colubrina	91.5 ± 2.6 a	52.50 ± 5.6 b
Mauritia flexuosa	79.1 ± 8.9 a	37.50 ± 15.7 b
Oenocarpus bataua	57.5 ± 17.1 a	12.50 ± 8.5 c
Platonia insignis	58.2 ± 3.4 a	2.50 ± 2.2 c
Pentaclethra macroloba	30.1 ± 2.6 b	0.0 ± 0.0 c
Vernonia polysphaera	67.3 ± 11.1 a	0.0 ± 0.0 c

Means followed by the same letter within columns do not differ with the Scott-Knott test at 5% significance. [1]S.E.M. = standard error of the mean.

The concentration-response mortality curves of *Copaifera* sp., *O. phalerata*, and *Carapa guianensis*

oils are presented in Figures 1A, B, and C, respectively. The concentration-response mortality curves varied substantially. The *Copaifera* sp. oil was the most toxic (Figure 1 A). The LC$_{50}$ varied from 7.50 to 60.84% (v/v) (Table 2). The TRs indicated that *Copaifera* sp. oil was 6.84 and 8.11 times more toxic than *O. phalerata* and *Carapa guianensis* oils, respectively. The curves for *O. phalerata* and *Carapa guianensis* had steeper slopes than the curve

for *Copaifera* sp. (Table 2), suggesting that small variations in toxicity could cause *S. frugiperda* to develop resistance to these two oils, as other studies have demonstrated with similar concentration-mortality curves (SOUSA; FARONI; FREITAS, 2014). Additional studies are needed to assess the toxicity of these oils against *S. frugiperda* and the potential for resistance among populations.
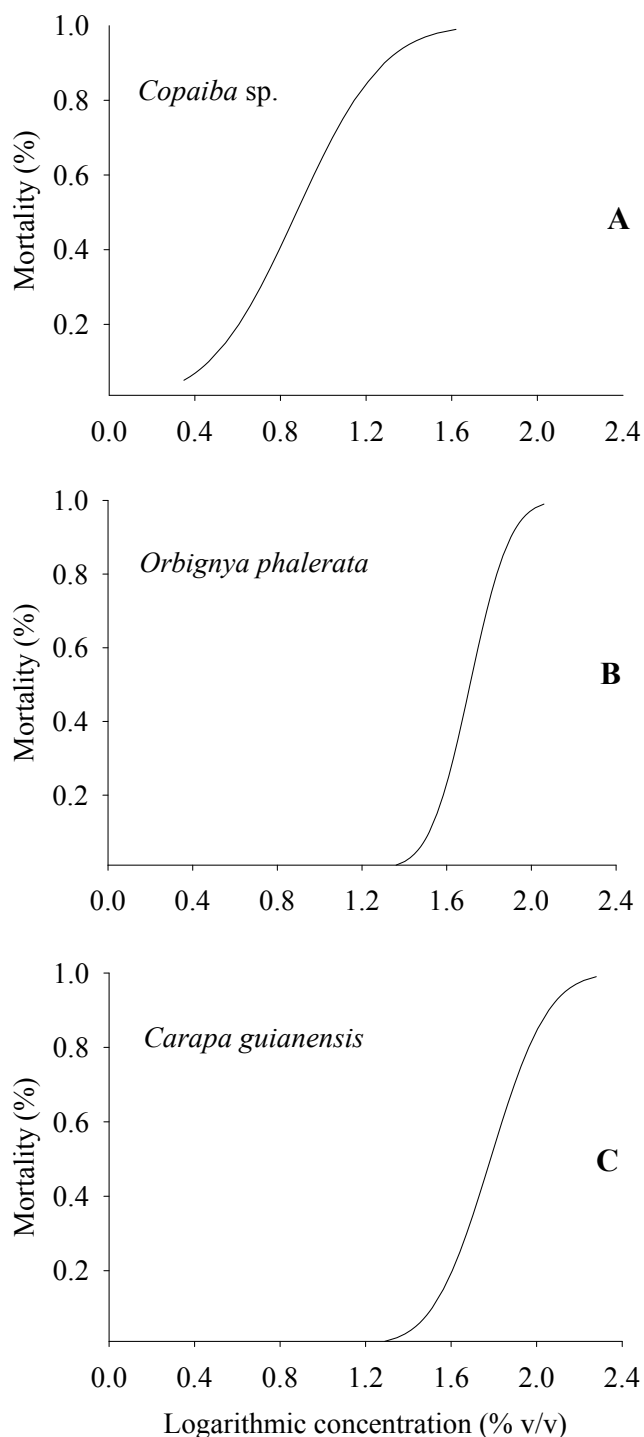
Figure 1. Mortality (%) of third-instar *S. frugiperda* caterpillars treated with different concentrations of the oils of *Copaifera* sp., *O. phalerata*, and *Carapa guianensis*.

Table 2. Toxicity of vegetable oils on third-instar caterpillar of *S. frugiperda*.

Plant species	Slope ± S.E.M.[1]	LC$_{50}$ (95% FI) % (v/v)	TR (95%CI) LC$_{50}$	χ^2	P
Copaifera sp.	3.12 ± 0.54	7.50 (5.91–9.10)	-	10.06	0.43
Orbignya phalerata	6.66 ± 0.85	51.31 (47.47–55.51)	6.84 (5.49–8.54)	24.20	0.14
Carapa guianensis	4.70 ± 0.69	60.84 (53.82–68.21)	8.11 (6.40–10.27)	15.56	0.34

[1]S.E.M. = standard error of the mean; LC$_{50}$ = lethal concentration to 50% of the insects; FI 95% = 95% fiducial interval; 95% CI =95% confidence interval; TR = Toxic ratio for LC$_{50}$; χ^2 = Chi-square test; P = probability.

Although the Amazon is internationally recognized as a biodiversity hotspot, in-depth studies of the flora as a source of insecticides are quite recent (COITINHO et al., 2006; SILVA et al., 2007; BARBOSA et al., 2011; SARRIA et al., 2011). In this study, most of the 11 oils displayed some activity against *S. frugiperda* eggs. However, *Copaifera* sp., *O. phalerata*, and *Carapa guianensis* oils were the most efficacious against *S. frugiperda* caterpillars.

The toxicity of *Copaifera* sp. (copaiba) oil demonstrated in this study corroborates the results of Alves et al. (2012). These authors found that methanolic extracts of leaves, bark, and fruits *Copaifera* were toxic to *S. frugiperda* caterpillars. Barbosa et al. (2011) found that commercial cultivation plots of tomato (*Lycopersicon esculentum* Mill., Solanaceae), after treatment with a copaiba extract, had reduced numbers of *Tuta absoluta* (Meyrick, 1917) (Lepidoptera: Gelechiidae) and *Bemisia tabaci* (Gennadius, 1889) (Hemiptera: Aleyrodidae) 24 and 72 h after spraying. The toxic effect of copaiba oil has been attributed to its terpenoid fraction (sesquiterpenes and labdanum-type diterpenes) (SILVA et al., 2007).

This is the first report of *O. phalerata* oil toxicity against *S. frugiperda*. Many other species of Arecaceae occur in the Amazon, but there are few studies on potential insecticides within this family. A previous study demonstrated *Attalea phalerata* Mart. ex Spreng. (Arecaceae) leaf extract toxicity against *S. zeamais* (SOUZA et al., 2008). The present study demonstrated the toxicity of *Carapa guianensis* oil against *S. frugiperda* caterpillars. Additionally, Sarria et al. (2011) demonstrated that limonoids isolated from *C. guianensis* arils prolonged caterpillar development and reduced pupa weight.

Plants used in this study were selected from reports on their toxicity, or of their genera and families, against insect pests. All the investigated species demonstrated some effectiveness against *S. frugiperda*. From an integrated management point of view, these results are of fundamental importance because they indicate that insecticidal compounds are present in these species (HAAS et al., 2014). These plants are common in the region and their oils can be easily manufactured, so they may be alternatives to synthetic insecticides for small producers. Locally produced pesticides have lower costs compared to synthetic insecticides, and botanical insecticides may be less damaging to the environment and are easy to prepare through relatively simple techniques (COITINHO et al., 2006; SOUSA; FARONI; FREITAS, 2014).

Copaifera sp., *O. phalerata*, and *Carapa guianensis* oils are ideal for incorporation into management programs for *S. frugiperda* in the Amazon region. However, botanical insecticides must be used with care because they can be toxic to mammals via ingestion (ELVIN-LEWIS, 2005), and some natural control agents (parasitoids and predators) are also susceptible to these products (BARBOSA et al., 2011).

CONCLUSIONS

The majority of the oils displayed moderate insecticidal activity against *S. frugiperda* eggs. *Copaifera* sp., *O. phalerata*, and *Carapa guianensis* oils effectively controlled *S. frugiperda* under laboratory conditions and should be considered as candidates for integrated management programs of corn pests. Additional studies are required to establish the maximum effectiveness of these oils against *S. frugiperda* eggs and caterpillars.

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