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Biological Activity of *Piper aduncum* extracts on *Anticarsia gemmatalis* (Hübner) (Lepidoptera: Erebidae) and *Spodoptera frugiperda* (J.E. Smith) (Lepidoptera: Noctuidae)

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ABSTRACT

Piper aduncum found naturally in the Amazon and southeastern Brazil, is known for its secondary metabolites that have activity on insects. *Anticarsia gemmatalis* and *Spodoptera frugiperda* are among the major insect pests associated with agricultural production. This research evaluated the biological activity of hexane, ethyl acetate, and ethanol extracts of *P. aduncum* leaves on mortality and duration of larval and pupal periods, as well as weight, width, and length of *A. gemmatalis* and *S. frugiperda* pupae. The mortality of *A. gemmatalis* larvae in trials with *P. aduncum* extracts were 93.3% (hexane) and 90% (ethyl acetate), estimating LC_{50} of 6.35 and 5.79 mg/mL, respectively. Mortality in *S. frugiperda* submitted to the hexane extract ranged from 3.33% to 96.66% (LC_{50} of 8.22 mg/mL). The ethanol extract induced low mortality (3.33% to 23.33%). The *P. aduncum* extracts did not affect the development of *S. frugiperda* pupae. In *A. gemmatalis* differences in weight and length occurred. The chemical characterization was by GC-MS, which revealed that the major constituent in the hexane extract of *P. aduncum* was apiol (90.7%). *P. aduncum* extracts are important and promising components to manage *A. gemmatalis* and *S. frugiperda*, which cause extensive production losses.

Key words: fall armyworm, velvetbean caterpillar, pest management, plant extracts.

INTRODUCTION

Soybean and corn crops are extensively cultivated in the state of Rio Grande do Sul, Brazil, thereby forming very simplified and vulnerable agroecosystems. Along with the benefits of these crops, problems have arisen such as the indiscriminate use of pesticides, which have adverse environment impacts. The overuse of these products has negative ecological consequences for human health, soil and water contamination, and the food chain.

The negative factors of pesticides have encouraged researchers to investigate less harmful control measures. One of the alternatives for controlling *Anticarsia gemmatalis* (Hübner) (Lepidoptera: Erebidae) and *Spodoptera frugiperda* (J.E. Smith) (Lepidoptera: Noctuidae) has been the

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use of natural products to facilitate environmentally safer production, which is fundamentally important for family farming and sustainable development.

The great diversity of plants in the world enables research about new products that may replace or reduce the use of synthetic chemical pesticides. Several plants of toxicological significance are from the Amazon region, such as *Piper aduncum* (Piperaceae). Surveys report that this plant species has antimicrobial (Abreu et al. 2015), insecticidal (Misni et al. 2008, Carmona-Hernández et al. 2014, Volpe et al. 2016), and acaricidal (Silva et al. 2007, 2009) effects. However, no reports in the literature have demonstrated the insecticidal effect of extracts from this plant on *A. gemmatalis* and *S. frugiperda.*

Phytochemical studies of the aerial parts of *P. aduncum* have isolated chalcones, flavonones, and dihydrochalcones (Morandim et al. 2009), a high amount of total flavonoid content, followed by phenols, and alkaloids with sesquiterpenes as the major compounds (Arroyo-Acevedo et al. 2015).

The objective of this research was to evaluate the biological activity on *A. gemmatalis* and *S. frugiperda* of hexane, ethyl acetate, and ethanol extracts of *P. aduncum* and chemically characterize these extracts.

MATERIALS AND METHODS

COLLECTION AND IDENTIFICATION OF BOTANICAL MATERIAL

P. aduncum leaves were collected in November 2012 (IBAMA - process 02001.004239 / 2013-05) from specimens grown in the Garden of Medicinal Plants of the Universidade Nilton Lins, located in the city of Manaus, AM, Brazil, at latitude 03°03' 36.5" S and longitude 060°00' 31.7" W. They were then identified and deposited in the herbarium of the Universidade de Caxias do Sul (HUCS 42569 and HUCS 42570, respectively).

EXTRACTION OF PLANT MATERIAL

The plant leaves were dried in an oven at 40 °C for five days and then ground. The extractions were performed using a Soxhlet extractor with eight hours for each extraction. For 200 g of crushed leaves, three liters of each solvent were used, with a sequence of solvents with increasing polarity: hexane, ethyl acetate, and ethanol. After extraction, the solvents were evaporated by rotary evaporator to obtain the crude extract.

PHYTOCHEMICAL ANALYSIS

The chemical composition of plant extracts was determined by gas chromatography-mass spectrometry (GC/MS). The analyzes were performed by gas chromatography coupled to mass selective detector, Hewlett Packard 6890 MSD 5973, equipped with HP Chemstation software and Wiley 275 spectral data, using the fused-silica capillary column HP-5MS (30 m X 250 m) and 0.5 m thick film (Hewlett-Packard, Palo Alto, CA, USA). The column temperature, 60 °C (8 min) to 180 °C at 3 °C/min, and 20 °C/min to 230 °C; injector 220 °C, interface 250 °C; Split ratio of 1:100; He carrier gas (56 kPa); flow 1.0 mL/min; ionization energy 70 eV. The components were identified by a combination of mass spectrum of the Wiley library and comparison with data from literature. (NIST 2016, USA: http://www.nist.gov/)

BIOLOGICAL TESTS

A. gemmatalis and *S. frugiperda* caterpillars were raised on an artificial diet (Greene et al. 1976) and kept in the insectarium of the Pest Control Laboratory at the Institute of Biotechnology of the Universidade de Caxias do Sul.

The hexane, ethyl acetate, and ethanol extracts of *P. aduncum* were solubilized in Tween-80 5% (v/v) at concentrations of 1.0, 2.5, 5.0, 10.0, and 15.0 mg/mL, previously determined, which were diluted, homogeneously in artificial diet. The

control group received diet with no extracts. For the biological tests, 30 third instar caterpillars were used for each treatment and one control group, for total of 180 *A. gemmatalis* caterpillars and 180 *S. frugiperda* caterpillars. Each caterpillar was individually placed in a 50 mL plastic cup, along with a moistened cotton ball and 1 g of artificial diet with extract, then left to feed for seven days. After this period, the surviving caterpillars were fed a diet without addition of extracts. Mortality rates were assessed daily until the pupal stage. The effects measured for the duration of larval and pupal stage included weight, length, and width of the pupa. Bioassays were maintained at room temperature (temperature 25 ± 1 °C and relative humidity 70 ± 5%).

STATISTICAL ANALYSIS

Data were analyzed using the parametric test for analysis of variance (ANOVA), in which the averages were compared by Tukey test (α = 0.05), with the aid of the Statistical Package for Social Sciences (SPSS for Windows 17.0). The graphics were prepared using the Graph Pad Prism 6.1 software. Probit analysis (Finney 1971) was used to determine the values of the median lethal concentration (LC_{50}).

RESULTS AND DISCUSSION

PHYTOCHEMICAL ANALYSIS OF *P. Aduncum*

The chemical characterization is an important step to relate the plants activity with their respective metabolites. The chemical constituents identified in the hexane extract of *P. aduncum,* 92.4% were phenylpropanoid, with the apiol as the major compound at 90.7%; 5.1% were sesquiterpenes (β-caryophyllene and β-selinene). In the ethyl acetate and ethanol extracts of *P. aduncum,* phenylpropanoid apiol was identified at 54.9% and 21.0%, respectively (Table I).

TABLE I
Chemical constituents identified in hexane, ethyl acetate, and ethanol extracts of *Piper aduncum* leaves.

Entry	Extract	Chemical constituent	Retention time	Abundance (%)
1	Hexane	apiol	21:57	90.7
	Ethyl acetate		21:52	54.9
	Ethanol		21:52	21.0
2	hexane	β-caryophyllene	18:41	3.6
3	hexane	β-selinene	19:54	1.5
4	hexane	myristicin	20:00	1.7

The presence of phenylpropanoids and sesquiterpenes (Table I) confirms what was found in the literature for plant species of Piperaceae family (Almeida et al. 2009; Riva et al. 2011; Cruz et al. 2014, Barros et al. 2016). Phenylpropanoid apiol is one of the major components of essential oil from *P. aduncum* (Santana et al. 2015).

Apiol (90.7%) was the major compound identified in hexane extract of *P. aduncum* (Table I). However, Silva et al. (2009) extracted the essential oil from hexane extract of *P. aduncum* leaves and identified phenylpropanoid dillapiol (94.84%) as the major constituent, and apiol was only 0.38%. Silva et al. (2013) also identified dillapiol (85.17%) as a major compound in the essential oil extracted from *P. aduncum* leaves. However, our research did not identify dillapiol in the hexane extract of *P. aduncum.* The absence of this chemical constituent may be due to factors such as plant cultivation system and environmental conditions in certain areas, which may interfere with the content and the presence or absence of some constituents of the plant.

The classes of secondary metabolites identified in *P. aduncum* (Table I) are one of the main features of the Piperaceae plant family. The Piperaceae family has one of the most versatile secondary metabolisms known in botanical families. The metabolites accumulated by species of this plant family are characterized as being derived from the mixed biosynthesis (shikimate/acetate), which

results mostly in the production of phenylpropanoid and terpenes (Fazolin et al. 2006).

This diversity of compounds reported in the literature depends on the environmental conditions during development or the age of the plant.

INSECTICIDAL ACTIVITY OF HEXANE, ETHYL ACETATE, AND ETHANOL EXTRACTS OF *Piper aduncum* ON *Anticarsia gemmatalis* AND *Spodoptera frugiperda*

Mortality rate of *A. gemmatalis* and *S. frugiperda,* subjected to different concentrations of hexane extract of *P. aduncum* leaves*,* ranged from 3.3 to 93.3% and from 3.3 to 96.6%, respectively. The concentrations of 5.0, 10.0, and 15.0 mg/mL showed statistical difference, in relation to the control, for the mortality percentage of evaluated caterpillars (Fig. 1a).

The average mortality of *A. gemmatalis* and *S. frugiperda* in trials with ethyl acetate extract ranged from 6.6% to 90.0%. The highest concentration, 15 mg/mL, induced 90.0% mortality for *A. gemmatalis* and 43.3% for *S. frugiperda* (Fig. 1b).

The ethanol extract of *P. aduncum* induced 23.3% mortality of *A. gemmatalis* and 26.6% mortality of *S. frugiperda,* for the highest concentration evaluated (15 mg/mL), with no statistically significant difference compared to other concentrations and the control group for both insect species (Fig. 1c).

Hexane and ethyl acetate extracts of *P. aduncum* induced the highest mortality rates of *A. gemmatalis* and *S. frugiperda.* These results can be attributed to the higher concentrations of phenylpropanoid in these extracts. These compounds derived from phenylalanine are present in most plant of the Piperaceae family. Several authors have found that these plant species have insecticidal and acaricidal activity (Salgado et al. 2012, Souto et al. 2012, Trindade et al. 2012, Lima et al. 2014, Santana et al. 2015).

Apiol was the major compound (90.7%) identified in the hexane and ethyl acetate extracts (54.9%). Santana et al. (2015) found that apiol was one of two major constituents of the *P. aduncum* essential oil and attributed larvicidal activity on the dengue vector *A. aegypti* (Diptera) to apiol. Khalaf (2004) confirmed the biological activity of apiol, because it interfered with growth and reduced fertility of adult flies of the species *Parasarcophaga dux* (Diptera). Studies with essential oil of *Petroselinum crispum* fruit*,* whose two main compounds were apiol and myristicin, demonstrated its high effectiveness in inhibiting oviposition of *Pseudaletia unipuncta* (Sousa et al. 2015).

The insecticidal activity of hexane and ethyl acetate extracts of *P. aduncum* may be associated with synergistic action of the phenylpropanoids apiol and myristicin, by inhibiting the function of cytochrome P450. According to Wilkinson et al. (1984), the secondary metabolites ingested by the insect form a complex with major detoxification enzymes, of dependent monooxygenase of cytochrome P450. According to Berenbaum (2002), these enzymes participate in many manufacturing processes in insects, which include ecdysteroid and juvenile hormone biosynthesis as well as detoxification from plant compounds and insecticides. Bernard and Philogene (1993) and Hodgson et al. (1995) argue that the synergists compounds bind to P450, inactivating this enzyme, which impedes metabolization of insecticide molecules. Li et al. (2007) report that inhibiting monooxygenase reduces the ability of the herbivorous insect to excrete xenobiotics, which results in mortality due to the accumulation of toxic substances in their digestive tract.

In addition, the insecticidal effect of these phenylpropanoid may be due to collaboration with other minor bioactive compounds, such as β-caryophyllene and β-selinene sesquiterpenes present in the hexane extract. According to Veiga

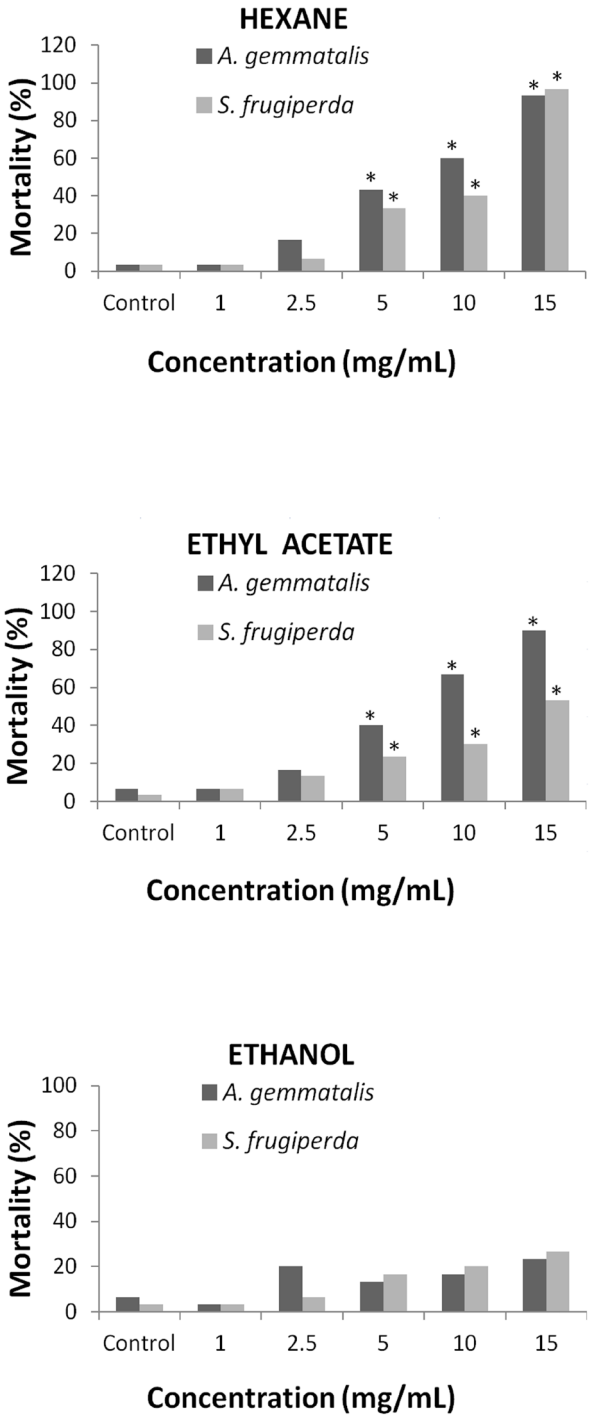

Figure 1 - Cumulative mortality of *Anticarsia gemmatalis* and *Spodoptera frugiperda* fed with artificial diet containing: **a**) hexane, **b**) ethyl acetate, and **c**) ethanol extracts of *Piper aduncum* leaves. *Averages mortality differs in relation to control as determined by Tukey test ($p \leq 0.05$).

Junior et al. (2005), β-caryophyllene is described in the literature to contain bactericide and insect repelling properties. Costantin et al. (2001) identified β-caryophyllene, α and β-pinene, bicyclogermacrene, spathulenol, and germacrene-D in *Piper cernuum* with antimicrobial activity. Other authors observed that the β-caryophyllene inhibits growth of the parasite *Trypanosoma cruzi* and *Leishmania brasiliensis* (Leite et al. 2013).

The mortality of *A. gemmatalis* and *S. frugiperda,* when compared to each other in the studied concentrations, presented statistical difference only in trials with ethyl acetate extract at concentration of 10 mg/mL. This may be related to the fact that *S. frugiperda* is polyphagous and therefore more resistant, which prevented mortality greater than 50% of this insect. Fortunato et al. (2007) stated that polyphagous insects, such as *S. frugiperda,* have a wide variety of digestive enzymes, due to the diverse chemical composition in their diet. Thus, polyphagous insects can best metabolize the compounds of their diet when compared to non-polyphagous insects, such as *A. gemmatalis.*

In relation to the estimated values of the median lethal concentration (LC_{50}) for *A. gemmatalis,* the hexane and ethyl acetate extracts presented the same toxicity. The hexane extract had the same efficacy of potential toxicity for both lepidopteran larvae. In tests with ethanol extract against *A. gemmatalis* and ethyl acetate and ethanol extract against *S. frugiperda,* the mortality percentages were lower than 50%, which prevented calculation of LC_{50} (Table II).

During the larval period, *A. gemmatalis* was fed a diet containing hexane extract of *P. aduncum* leaves, with increasing concentrations of extract. Statistical differences were observed between the control group and 5.0 mg/mL concentration. The duration of the pupal stage increased by approximately one day for the 2.5 mg/mL concentration. No statistical differences were found

TABLE II
Median Lethal Concentration (LC_{50}) of hexane, ethyl acetate, and ethanol extracts of *Piper aduncum* against *Anticarsia gemmatalis* and *Spodoptera frugiperda.*

Insect	Extract	LC_{50} (mg/mL)	Confidence Interval
A. gemmatalis	Hexane	6.35 [ab]	4.82 - 7.98
	Ethyl acetate	5.79 [a]	4.57 – 7.39
S. frugiperda	Hexane	8.22 [b]	7.87 – 9.11

Average mortality followed by different letters differs by Tukey test ($p \leq 0.05$).

in the pupal weight between the treatments and the control group. Only the 15.0 mg/mL concentration shortened the length of the pupae compared to control group. The width of the pupae did not change in the treated and the control groups (Table III).

In *S. frugiperda,* the larval period was prolonged in all concentrations of hexane extract compared to the control. However, the pupae treated with hexane extract at concentrations of 2.5, 5.0, and 10.0 mg/mL experienced the same larval duration. The 2.5 mg/mL concentration increased the pupal duration compared to the control group. The treated pupae showed no weight reduction compared to the control group. There was no statistical difference for the width and length of the pupae in all groups. With 15 mg/mL concentration,

TABLE III
Biological parameters of *Anticarsia gemmatalis* and *Spodoptera frugiperda* fed artificial diet containing the hexane extract of *Piper aduncum* leaves.

Anticarsia gemmatalis						
Biological parameters	Control	1 mg/mL	2.5 mg/mL	5 mg/mL	10 mg/mL	15 mg/mL
n	29	29	25	17	12	2
Larval duration (days)	8.9±1.87 a	9.2±1.75 ab	9.8±1.75 ab	10.3±2.47 bc	11.8±2.30 c	11.5±0.5 c
Pupal duration (days)	9.9±1.24 a	10.6±1.20 ab	11.3±1.68 b	11.4±1.61 b	11.7±1.22.0 b	11.5±1.34 b
Pupal weight (mg)	0.2±0.02 a	0.2±0.02 a	0.2±0.02 a	0.2±0.01 a	0.2±0.01a	0.2±0.002 a
Pupal width (mm)	5.2±0.22 a	4.9±0.35 a	5.0±0.24 a	5.1±0.20 a	5.0±0.32 a	4.8±0.32 a
Pupal length (mm)	18.1±0.59 a	17.9±0.78 ab	17.7±0.72 ab	17.6±0.65 ab	17.3±0.67ab	16.9±0.53 b
Spodoptera frugiperda						
Biological parameters	Control	1 mg/mL	2.5 mg/mL	5 mg/mL	10 mg/mL	15 mg/mL
n	29	29	28	20	18	1
Larval duration	19.8±1.34 a	21.5±1.36 b	22.8±1.25 bc	23.4±1.29 c	23.5±1.46 c	-
Pupal duration (days)	11.4±1.27 a	12.1±1.28 ab	13.2±1.34 bc	13.4±1.51 c	13.5±1.46 c	-
Pupal weight (mg)	0.2±0.02 a	0.2±0.02 a	0.2±0.01 a	0.2±0.02 a	0.2±0.39 a	-
Pupal width (mm)	4.8±0.30 a	4.6±0.49 a	4.6±0.34 a	4.7±0.42 a	4.7±0.39 a	-
Pupal length (mm)	15.8±1.05 a	15.0±0.76 a	15.3±0.85 a	15.4±1.07 a	15.6±1.28 a	-

For means followed by the same letters, the lines do not differ by Tukey test ($p \leq 0.05$).

TABLE IV
Biological parameters of *Anticarsia gemmatalis* and *Spodoptera frugiperda* fed artificial diet containing the ethyl acetate extract of *Piper aduncum* leaves.

Anticarsia gemmatalis						
Biological parameters	Control	1 mg/mL	2.5 mg/mL	5 mg/mL	10 mg/mL	15 mg/mL
n	28	28	25	18	10	3
Larval duration (days)	14.1±1.30 a	14.7±1.34 a	15.4±1.20 ab	16.2±1.22 b	17.3±1.46 b	17.8±1.45 b
Pupal duration (days)	9.2±1.25 a	10.1±1.27 ab	10.1±1.34 ab	11.3±1.31 b	13.1±1.21 c	13.2±1.23 c
Pupal weight (mg)	0.3±0.03 a	0.2±0.03 b	0.2±0.03 b	0.2±0.03 b	0.2±0.03 b	0.2±0.03 b
Pupal width (mm)	5.9±0.31 a	5.3±0.36 a	5.2±0.39 a	5.2±0.34 a	4.9±0.49 a	5.8±0.36 a
Pupal length (mm)	19.0±1.00 a	17.7±0.88 a	17.6±1.01 a	17.1±1.27 a	17.5±0.94 a	18.8±0.26 a
Spodoptera frugiperda						
Biological parameters	Control	1 mg/mL	2.5 mg/mL	5 mg/mL	10 mg/mL	15 mg/mL
n	29	28	26	23	21	16
Larval duration (days)	19.8±1.26 a	18.1±1.32 a	18.6±1.29 a	19.3±1.34 a	19.5±1.38 a	21.3±1.35 b
Pupal duration (days)	11.4±1.37 a	11.5±1.39 a	11.5±1.29 a	11.6±1.34 a	11.6±1.37 a	11.7±1.39 a
Pupal weight (mg)	0.2±0.16 a	0.2±0.02 a	0.2±0.24 a	0.2±0.03 a	0.2±0.02 a	0.2±0.03 a
Pupal width (mm)	4.8±0.30 a	4.8±0.34 a	4.5±0.41 a	4.4±0.52 a	4.4±0.51 a	4.3±0.62 a
Pupal length (mm)	15.8±1.05 a	15.7±1.07 a	16.0±1.23 a	16.5±0.81 a	16.2±1.08 a	15.7±0.91 a

For means followed by the same letters, the lines do not differ by Tukey test ($p \leq 0.05$).

96.6% mortality was obtained of larvae during the first five days, which made it impossible to analyze the biological parameters (Table III).

For the ethyl acetate extract of *P. aduncum* leaves, assays with *A. gemmatalis* found extended larval and pupal stages due to the 5.0 mg/ml concentration, compared to control. Pupal weight reduced at all concentrations of the extract. However, for width and length, the control group and studied concentrations presented no statistical difference (Table IV).

The larval period of *S. frugiperda* was prolonged by 1.8 days by the 15.0 mg/mL concentration of the ethyl acetate extract compared to the control group. The duration of the pupal period showed no statistical difference for treatment in all evaluated concentrations. However, the pupal weight showed no statistical difference between the treatments and the control group. The width and length of the pupae did not statistically differ between treatments (Table IV).

For the ethanol extract of *P. aduncum* leaves, the larval period of *A. gemmatalis* ranged from 17.9 to 19.8 days, and it was extended significantly with 2.5 mg/mL concentration compared to control. The duration of the pupal stage ranged from 15.2 to 18.1 days and increased beginning with 1.0 mg/mL concentration. The pupae presented reduced weight in all treatments compared to the control. The width and length showed no statistical difference between the control and the concentrations evaluated in the treatments (Table V).

The ethanol extract extended the duration of larval and pupal periods of *S. frugiperda* beginning

TABLE V
Biological parameters of *Anticarsia gemmatalis* and *Spodoptera frugiperda* fed artificial diet containing the ethanol extract of *Piper aduncum* leaves.

Anticarsia gemmatalis						
Biological parameters	Control	1 mg/mL	2.5 mg/mL	5 mg/mL	10 mg/mL	15 mg/mL
n	28	29	26	26	25	23
Larval duration (days)	16.4±1.24 a	17.9±1.29 ab	18.5±1.25 bc	18.2±1.24 bc	18.7±1.33 bc	19.8±1.37 c
Pupal duration (days)	13.3±1.32 a	15.2±1.35 b	16.2±1.35 bc	16.4±1.39 bc	17.2±1.45 c	18.1±1.43 d
Pupal weight (mg)	0.3±0.02 a	0.2±0.03 b	0.2±0.02 b	0.2±0.02 b	0.2±0.02 b	0.2±0.01b
Pupal width (mm)	5.8±0.58 a	5.3±0.46 a	5.4±0.31 a	5.2±0.20 a	5.4±0.37 a	5.4±0.40 a
Pupal length (mm)	18.7±0.74 a	17.2±1.48 a	17.4±0.57 a	17.9±0.81 a	17.8±0.87 a	17.6±0.62 a
Spodoptera frugiperda						
Biological parameters	Control	1 mg/mL	2.5 mg/mL	5 mg/mL	10 mg/mL	15 mg/mL
n	30	29	28	25	24	22
Larval duration (days)	14.9±1.31 a	18.6±1.34 b	19.2±1.29 b	21.3±1.35 c	21.3±1.39 c	21.5±1.41 c
Pupal duration (days)	11.2±1.25 a	13.3±1.29 b	13.5±1.32 b	13.5±1.29 b	13.6±1.32 b	13.6±1.34 b
Pupal weight (mg)	0.2±0.02 a	0.2±0.2 a	0.2±0.02 a	0.2±0.33 a	0.2±0.02 a	0.2±0.02 a
Pupal width (mm)	4.8±0.24 a	4.6±0.33 a	4.6±0.43 a	4.8±0.49 a	4.5±0.54 a	3.9±0.71 a
Pupal length (mm)	16.6±0.79 a	16.0±0.86 a	16.1±1.03 a	16.1±0.85 a	16.1±0.96 a	15.8±1.04 a

For means followed by the same letters, the lines do not differ by Tukey test ($p \leq 0.05$).

at 1.0 mg/mL concentration compared to control. The pupal weight did not statistically change between the treatments and the control group. No statistically significant differences appeared in the width and length of the pupae in the control group and the studied concentrations (Table V).

The extension of the larval and pupal stages by different extracts can be attributed to the presence of growth inhibitors or toxic substances (phenylpropanoid and sesquiterpenes) in the extracts. Studies have shown that organic compounds in plants with insecticidal activity can act as chitin synthesis inhibitors and may affect growth, development, reproduction, and diapause in insects (Aguiar-Menezes 2005). Phenylpropanoids are toxic to insects and may affect the life cycle of insects (Regnault-Roger et al. 2012, Kim and Lee 2014). Studies using leaf extracts from *A. coriacea* and *A. dioica,* with high content of phenolic compounds, found decreased weight gain, adult emergence, fecundity, fertility, and egg hatching of *S. frugiperda* (Freitas et al. 2014).

Morphological and physiological changes observed in this study may be attributed to the action of phenylpropanoid and sesquiterpenes, which can cause toxic interference in biochemical and physiological functions of insects. According to Trindade et al. (2000), some components of the extract interfere with the hormonal system that regulates insect larval development. Mordue and Blackwell (1993) argue that these changes result from reduced concentration of ecdysone or delay

of its release in the hemolymph, which affects larval development, thereby reducing the insect population and damage to plants.

Other researchers have found similar results in the duration of *S. frugiperda* larva as induced by the hexane extract of *P. aduncum* (Table III). Santiago et al. (2008) observed this with aqueous extracts of *Ruta graveolens* (26 days), *Momordica charantia* (22.6 days), and *Lippia sidoides* (24.8 days). Knaak et al. (2012) used aqueous extracts obtained by maceration and infusion of the species *Symphitum officinalis* (26.9 days), *Zingiber officinale* (28.6 days), and *Melissa officinalis* (26.6 days). However, the larvae treated with plant extracts in the studies of these authors showed no significant difference from the control group, unlike the hexane extract evaluated in our study. Under field conditions, extending the larval stage can increase the exposure time to the plague of natural enemies and reduce the number of generations (Torres et al. 2001). These factors are important for maize production, because this Lepidoptera can cause yield losses up to 54.5% (Figueiredo et al. 2006), depending on climactic conditions and development stage in which the plant is attacked (Dequech et al. 2013, Ribeiro et al. 2014).

With respect to the morphometric parameters (weight, length, and width), the larvae fed with diet containing extract produced smaller pupae and adults than the control group. Some authors have attributed this effect to the presence of peptidase inhibitors, which can be expressed by plants as a defense mechanism (Gahloth et al. 2011, Macedo et al. 2011, Cruz et al. 2013, Ghodke et al. 2013) resulting in delayed development of insects.

Lepidoptera accumulate reserves during larval development, and the adverse conditions in the process cause asynchrony with normal population which leads to problems with these insects' reproduction (Rodriguez and Vendramim 1997, Matos et al. 2006), fertility, and low performance of their offspring (Fantinou et al. 2008).

Based on these results, we conclude that the hexane and ethyl acetate extracts of *P. aduncum* have biological activity on *A. gemmatalis* and *S. frugiperda* species. Future studies with compounds isolated from these extracts may contribute to management strategies for these insect pests. The detection of new sources of insecticidal compounds from flora remains one of the principal gaps to be filled.

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