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Full Length Research Paper

Biological activity of *Ipomoea pauciflora* Martens and Galeotti (Convolvulaceae) extracts and fractions on larvae of *Spodoptera frugiperda* J. E. Smith (Lepidoptera: Noctuidae)

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Hexane, chloroform and methanol extracts of different parts of *Ipomoea pauciflora* were tested for their effects on the survival and development of Fall Armyworm (*Spodoptera frugiperda*), a Lepidoptera pest. For seven days, neonatal larvae (grown at 27 ± 2°C with a 16: 8 (L: D) h photoperiod) were exposed to different concentrations of crude *I. pauciflora* extracts (ranging from 0 to 4 mg/ml) that were incorporated into an artificial diet. Surviving larvae were weighed at days 6, 9 and 13 and were maintained until moths emerged. Eleven of the 18 crude extracts showed more than 30% larval mortality. The highest mortality was produced by hexane and chloroform extracts of seeds at 4 mg/ml (96.9 and 93.8%, respectively), with LC_{50} values of 1.85 mg/ml and 0.54 mg/ml, respectively. Fractions of both seed extracts were isolated by gravity column chromatography over silica gel and analyzed for their active compounds. Eight fractions of the hexane extract and six fractions of the chloroform extract from *I. pauciflora* seeds, exhibited larvicidal effects at 1 mg/ml (mortality from 33.3 to 88.9% and from 47.2 to 77%, respectively). Changes in larval weight were observed as compared with the control group. Phytochemical analysis through GC-MS and H^1 NMR revealed the presence of fatty acids and aldehydes in the active fractions. These results indicate that the bioactive extracts from the seed of *I. pauciflora* can induce lethal toxicity in *S. frugiperda* larvae or affect the weight of the surviving larvae.

Key words: *Ipomoea pauciflora*, bioassay, mortality, LC_{50} values, extracts, fractions, *Spodoptera frugiperda* larvae.

INTRODUCTION

Screening of active compounds from wild plants is an alternative approach to the development of agrochemical products (Dayane et al., 2009). Plant extracts have been screened for their biological activities against pests, such as their toxicity as well as their antifeedant, repellent, fumigant, growth-regulating and oviposition-deterret properties (Singh and Saratchandra, 2005; Isman, 2006).

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The use of these natural compounds instead of conventional pesticides as agrochemical agents can reduce environmental pollution, preserve non-target organisms and prevent pest resurgence (Singh and Saratchandra, 2005).

Recently, there has been a growing interest in investigating the insecticidal potential of extracts from plants used in folk medicine. There are about 20,000 species of tropical plants in Mexico, out of which 5, 000 are considered to possess medicinal values, which is yet to be documented and evaluated by means of scientific approaches (Aguilar and Martínez, 1993), such as several species of the family Convolvulaceae (Prakash and Rao, 1997). In this family, some species of the genus *Ipomoea* such as *Ipomoea tricolor*, *Ipomoea batata* and *Ipomoea murucoides* are traditionally used as nutricional, emetic, diuretic, diaphoretic, purgative (Pereda and Bah, 2003) and pesticidal agents (Jackson and Peterson, 2000; Vyvyan, 2002; Vera et al., 2009). These species have attracted considerable attention as a possible new source of biologically active natural products. Compounds such as resin glycosides from *Ipomoea* species have been reported for their purgative action and some other activities (Evans, 2002).

I. pauciflora, a perennial tree that grows in warm climates, is native of Mexico. It is popularly used as a medicinal plant for muscle pain, toothache, cough and stings (Monroy and Castillo, 2000); it is also part of the vegetation of the biosphere reserve "Sierra Huautla-Cerro Frío" at the State of Morelos, Mexico (Dorado et al., 2005) and its isothers regions (Bussmann and Sharon, 2006; Carranza, 2008). Although ethnobotanical information about the use and distribution of *I. pauciflora* is available, phytochemicals and toxicologicals studies have not been reported. Thus, the objective of this study was to determine whether hexane, chloroform and methanol extracts and fractions of leave, stem, bark, flower, bud and seed from *I. pauciflora* possess any biological activities against *S. frugiperda*.

MATERIALS AND METHODS

Plant material

I. pauciflora wild plant material was collected in Yautepec, State of Morelos, Mexico (1213 m asl) and authenticated by Biol. Manuel Castro from the Facultad de Ciencias, UNAM. A voucher specimen has been deposited (voucher number 15520) at Medicinal Herbarium IMSSM of the Mexican Institute for Social Security (National Medical Center Century XXI, Mexico, DF, Mexico).

Extraction

Fresh leaves (L), stems (St), barks (B), flowers (F), buds (Bu) and seeds (S) were collected from *I. paucifora* plants, dried at room temperature (26 - 28°C) and grinded. Extracts were obtained by maceration of the plant material (1500 g) followed by sequential extraction in 1500 ml of hexane (Hex) (purity 99.5%), chloroform ($CHCl_3$) (purity 99.5%) and methanol (MeOH) (purity 99.8%) in amber glass flasks. Each extraction was repeated three times over a period of three days each time. The solvents of the extracts were later removed with a rotatory evaporator. The crude extracts were weighed and stored at 4°C in amber glass flasks before used.

Bioassays to assess the toxicity of the crude extracts

For bioassays, a concentration of 4 mg/ml was used. 480 mg of each extract was dissolved in 1.8 ml of solvent (acetone or methanol for the methanol extracts and acetone or hexane for the hexane and chloroform extracts). The mixture was gently shaken to ensure a homogeneous solution. An aliquot of the solution (450 µl) was mixed with 30 ml of hot artificial diet. The extract-diet mixture was stirred and placed in 24-wells cell culture plates (Cell Well No. 3527, Corning Incorporated Costar® Corning NY) at 1 ml per well and allowed to solidify at room temperature.

S. frugiperda eggs were obtained from a colony maintained at the Biological Control Laboratory of the Biotechnology Research Center, of the Autonomous University of the Morelos State (UAEM), Morelos, Mexico. To obtain a cohort of *S. frugiperda* larvae, some eggs were placed in an incubator one to two days before the experiment. After hatching, one neonatal larva from the cohort was placed into one plate well (one larva/well). Larvae exposed to the solvent-diet mixture (450 µl of acetone, methanol or hexane) as well as normal artificial diet was used as a negative control and Fosdrim® (an organophosphate insecticide) mixed with 30 ml of artificial diet was used as a positive control.

One experimental unit was defined as one 24-wells cell culture plates with 24 neonatal larvae. Each treatment was performed in four replicates with a total of 2400 larvae tested. All larvae were incubated for seven days at 27 ± 2°C, with a photoperiod of 16: 8 h (L:D) and 60% ± 5 relative humidity (RH) in a growth chamber, for seven days. They were checked every day and the numbers of live and dead larvae were recorded. The mortality percentage (%M) was recorded and analyzed at day seven of the treatment. After seven days of incubation, the surviving larvae were fed with the normal diet and their weight was recorded at 6, 9 and 13 days of incubation to determine sublethal effects of extracts on larvae.

Determination of lethal concentration (LC_{50}) values

The median lethal concentration (LC_{50}) was determined for the extracts that showed lethal toxicity (> 50% mortality at the highest dose) in the previous bioassays. Different concentrations of each crude extract (0.4, 0.8, 1.0, 1.5, 2, 2.5, 3.0, 3.5 and 4.0 mg/ml) together with the controls (acetone, methanol, hexane and normal diet) were tested on larvae as previously described. Each treatment involved 96 neonatal larvae of *S. frugiperda* in four replications. Larvae were incubated at 27 ± 2°C, with a photoperiod of 16: 8 (L: D) h and 60% ± 5 RH. Mortality data were recorded after seven days of incubation.

Chromatographic separation of extracts

Extracts with major biological activity on *S. frugiperda* (> 90%M) were fractionated by gravity column chromatography packed with 36 g of silica gel 60 (70 - 230 mesh, Merck Darmstadt, Germany) and eluted with dichloromethane-methanol (100: 0, 95: 5, 90: 10, 85: 15 and 80: 20). Fractions collected with similar chromatographic behavior in thin layer chromatography (TLC) were combined and weighed. TLC was carried out on precoated Kieselgel 60 F_{254} (0.25 mm thick, Merck Darmstadt, Germany) plates. The elution pattern

Table 1. Mortality percentage (M %) of *S. frugiperda* neonatal larvae after seven days of exposure to different extracts of *I. pauciflora* (4 mg/ml).

Treatments	M% ± SE[1]	
Fosdrim	100 (± 0)	a
S/Hex	96.875 (± 1.99)	a
S/$CHCl_3$	93.750 (± 3.99)	a
S/MeOH	57.292 (± 2.62)	b
F/Hex	53.125 (± 1.04)	bc
Bu/MeOH	51.042 (± 1.99)	bcd
L/Hex	48.611 (± 1.39)	bcde
L/$CHCl_3$	46.875 (± 2.62)	bcde
St/MeOH	43.750 (± 1.21)	bcde
Bu/ $CHCl_3$	35.417 (± 1.20)	cdef
F/ $CHCl_3$	32.292 (± 1.99)	cdefg
Bu/Hex	31.250 (± 2.08)	defg
B/ $CHCl_3$	28.125 (± 1.04)	efgh
L/MeOH	27.083 (± 3.99)	efgh
F/MeOH	20.833 (± 1.70)	fgh
B/Hex	16.667 (± 1.70)	gh
St/Hex	13.542 (± 3.12)	h
St/$CHCl_3$	10.417 (± 2.08)	hi
MeOH	3.125 (± 1.04)	ij
Acetone- MeOH	3.125 (± 1.99)	ij
B/MeOH	1.042 (± 1.04)	j
Acetone- Hex	1.042(± 1.04)	j
Hexane	0 (± 0)	j
Acetone	0 (± 0)	j
Diet	0 (± 0)	j

ANOVA (P = 0.0001). C.V.= 14.34. Means followed by the same letter do not differ statistically (Tukey, α = 0.05).
Data were transformed [Arcsine (√(score value/100))] for analysis and expressed in the table in terms of the original variables.
[1]Standard error (SE), Leaves (L), stems (St), barks (B), flowers (F), buds (Bu) and seeds (S). Hexane (Hex), chloroform ($CHCl_3$) and methanol (MeOH).

of each fraction was determined using dichloromethane as the mobile phase and spots were visualized by spraying the plates with 10% H_2SO_4 solution followed by heating. A total of eight fractions of hexane seed extract and six fractions of chloroform seed extract were obtained which were numbered according to their elution order from the column.

For bioassays, fractions of the extract were used at a concentration of 1 mg/ml. First, 51 mg of the fractions was dissolved in 765 µl of hexane and 255 µl of these solutions was then mixed with 17 ml of hot artificial diet. All the solutions were tested as described above.

Mortality was recorded after seven days of incubation and all surviving larvae were weighed at 6, 9 and 13 days of treatment. For each treatment group, 36 neonatal larvae were used in three replications. Fractions with significant biological activities (1A, 1B, 1C, 1D, 1F, 1H, 2B, 2C, 2D, 2E, 2F, 2G) were further analyzed by nuclear magnetic resonance (NMR) and gas chromatography coupled to mass spectrometry (GC-MS).

Characterization of fatty acids and aldehydes

The GC-MS system consisted of a HP 6890 gas chromatograph equipped with a flame ionization detector and a HP 5970 mass selective detector in the electron-impact ionization mode. The chromatographic conditions were: 5 m x 0.5 mm HP-5 column; He, 6 mL/min; 50°C, 2 min, 50 - 220°C, □ 10°C/min, 220°C, 10 min; temperatures of the injection port and the detector 300°C; split 20: 1.

NMR spectra were obtained on a Varian spectrometer model Gemini 200 at 25°C using standard Varian software. Proton chemical shifts were referenced to internal tetramethylsilane (TMS), with 20 mg of each chromatographic fraction dissolved in ca. 0.65 mL of chloroform*d*.

Statistical analysis

Mortality percentage data for seven days were transformed [Arcsine (√(score value/100)] for their analysis and together with the larval weights at day 6, 9 and 13 were analyzed by analysis of variance using PROC GLM and by Tukey's test (Zar, 1999; SAS, 2005). Normality and homogeneity of variance were determined using the Kolmogorov-Smirnov test and the Levene´s test, respectively (Zar, 1999). LC_{50} values (concentration at which 50% of the larvae died) were calculated using probit analysis according to calculation outlined in POLO-PC (LeOra, 2002). Mortality data were also analyzed by chi-square goodness-of-fit to determine whether these data fit the probit or logit models.

RESULTS AND DISCUSSION

Biological activity of *I. pauciflora*

Bioassay results of the toxic effect of the different *I. pauciflora* extracts on *S. frugiperda* are shown in Table 1. Significant difference among the treated larvae were observed (P = 0.0001) after seven days of exposure to the extracts in the diet. All extracts except the hexane and chloroform seed extracts induced a below 60% mortality. None of the negative controls (MeOH, acetone, hexane and normal diet) caused mortality while the organophosphate pesticide Fosdrim® (positive control) induced 100% mortality. Only the methanol barks extracts (10.42% mortality) was not statistically different from the negative controls. Hexane and chloroform seed extracts induced higher mortality than the other extracts (96.875 and 93.75%, respectively). This lethal toxicity was statistically equal to that of Fosdrim® (positive control).

In the present study, crude extracts (of seeds) of *I. pauciflora* showed lethal toxicity against neonatal larvae of *S. frugiperda* been the most active. Xie et al. (1994) observed that wood and bark extracts of the species of the genus *Trichilia* were more active than foliar extracts. As for *I. pauciflora*, the seed extracts were more active than the other plant parts, which suggested an evolutionary advantage to have higher levels of these active compounds in the seed rather than other parts of the plant (Wheeler et al., 2001).

The seeds of many plant species can synthesize metabolites (proteins, carbohydrates and lipids) which in addition

Table 2. Mortality percentage (M%) of *S. frugiperda* neonatal larvae after seven days of exposure to different fractions of hexane extractof *I. pauciflora* seeds in an artificial diet (1 mg/ml).

Treatments	M% (± SE[1])	
Fosdrim	100 (± 0)	a
1B	88.9 (± 5.5)	ab
1F	88.9 (± 5.5)	ab
1D	77.8 (± 5.5)	bc
1C	66.7 (± 4.8)	c
1A	33.3 (± 4.8)	d
1H	33.3 (± 4.8)	d
1E	0 (±0)	e
1G	0 (±0)	e
Diet	0 (±0)	e
Hexane	0 (±0)	e

ANOVA (P = 0.0001). C.V.= 16.14. Means followed by the same letter do not differ statistically (Tukey, α = 0.05). Data were transformed [Arcsine (√(score value/100)] for their analysis and expressed in the table in terms of the original variables. [1]Standard error (SE). 1 = Hexane extract fractions of *I. pauciflora* seed.
A, B,...,H = Fractions obtained and numbered according to their elution order from the column.

to serving as energetic reserves for the embryo, can protect the tissue of the seeds from predators (Powell, 2009).

Many natural products isolated from seeds show functions ranging from plant growth regulators, fungicides, insecticides and repellents of herbivores. These compounds may also be present in other plant parts of the plants, but probably at lower concentrations (Powell, 2009).

Several authors have reported lethal toxicity against Lepidoptera larvae induced by seed extracts, such as the methanolic extract of *Annona cherimolia* (Colom et al., 2007) , extracts of plants from the family Meliaceae, such as *Azadirachta indica*, *Melia azedarach* and *Aglaia cordata* (Mikolajczak and Reed, 1987; Mikolajczak et al., 1989).

Experiment performed in larvae of *Helicoverpa armigera* Hubner (Lepidoptera: Noctuidae) exposed to different concentrations of hexane, chloroform and ethyl acetate extracts of *Atalantia monophylla* leaves at 5, 10, 25 and 50 mg/ml showed that the highest larval mortality was induced by the hexane extracts at different concentrations (Baskar et al., 2009).

As reported previously, neonatal larvae of *S. frugiperda* were exposed to different concentrations of methanol extracts of flower/seed of *Galphimia glauca* (Malpighiaceae) and the LC_{50} was 0.412 mg/cm^2 (Román, 1998). The finding that the flower/seed mix extract contain more active compounds against *S. frugiperda* than all the other extracts tested is in agreement with ours.

Lethal concentration (LC_{50}) values

Mortality data recorded with different concentrations of hexane seed extract for seven days of incubation fit the probit model better than the logarithmic model (X^2 = 2.03) and the LC_{50} of this extract was 1.68 mg/ml (1.58 - 1.77 mg/ml, α = 0.05). Mortality data for the chloroform seed extract also fit the probit model (X^2 = 4.39) and the LC_{50} of this extract was determined as 0.55 mg/ml (0.46 - 0.63 mg/ml, α = 0.05), indicating a two-fold higher toxicity over that of the hexane extract.

Linear regression probit analysis (concentration - mortality) revealed that the larvicidal activity of the crude extracts of seeds is directly proportional to their concentration. Baskar et al. (2009) observed that as compared with the chloroform and ethyl acetate extracts (LC_{50} value of 49 mg/ml and 52.6 mg/ml, respectively), the hexane extract of *A. monophylla* leaves showed the highest toxicity (LC_{50} 24.6 mg/ml) against the larvae of *H. armigera.*

Effect of fractions of the hexane and chloroform extracts of *I. pauciflora* seeds on *S. frugiperda*

The bioassays results demonstrated that the different fractions of hexane seed extract are statistically different (P = 0.0001). Fractions 1A, 1B, 1C, 1D, 1F and 1H caused larval mortality from 33.3 to 88.9%, with fractions 1B and 1F been the most active, causing 88.9% mortality (Table 2).

Similarly, the mortalities caused by chloroform seed extract fractions, were statistically different from each other (P = 0.0001) where fractions 2B, 2C, 2D, 2E, 2F and 2G yielded mortalities from 47.2 to 77% (Table 3).

Eight fractions, 1B, 1C , 1D, 2D, 2E, 1F, 2F and 2G (tested at 1 mg/ml concentration), showed lethal toxicity statistically equal to that of the hexane and chloroform crude extracts of seed at 4 mg/ml. These results agreed with those of Baskar et al. (2009), who reported that the larvicidal activity of the fraction of the hexane extract of *A. monophylla* at 1 mg/ml was higher than that of the crude extract on *H. armigera*.

The average weights of larvae fed on active fractions from *I.* pauciflora (1 mg/ml in an artificial diet) after 6, 9 and 13 days of incubation are shown in Table 4. Significant differences among fractions were observed (P = 0.0001). There was a significant weight reduction in the surviving larvae when they were fed with fractions 1A, 2G, 1H, 1B, 2D, 2F, 1F, 1C, 2E, 1D, 2B and 2C (P = 0.0001) in agreement with weight evolution observed in Table 4. Despite continuous feeding with normal diet, larvae on these treatments failed to recover to the normal weight. After 13 days of incubation, their maximum weights ranged from 186.4 mg ± 1.9 mg to 6.2 mg ± 0.25 mg which represented a weight reduction from 47 to 98% as compared to the control larvae (Figures 1a and b).

Under these conditions, the larvae were probably unable

Table 3. Mortality percentage (M%) of *S. frugiperda* neonatal larvae after seven days of exposure to different fractions of chloroform extract of *I. pauciflora* seeds in an artificial diet (1 mg/ml).

Treatments	M% (± SE [1])
Fosdrim	100 (± 0) a
2E	77.8 (± 5.55) b
2F	72.2 (± 2.77) bc
2G	69.4 (± 5.55) bc
2D	66.7 (± 0) bcd
2C	52.8 (± 5.55) cd
2B	47.2 (± 5.55) d
Hexane	5.55 (± 0) e
Diet	0 (± 0) e

ANOVA (P = 0.0001). C.V. = 13.45. Means followed by the same letter do not differ statistically (Tukey, α = 0.05). Data were transformed [Arcsine (√(score value/100))] for their analysis and expressed in the table in terms of the original variables. [1]Standard error (SE). 2 = Chloroform extract fractions of *I. pauciflora* seed. A, B,…,H = Fractions obtained and numbered according to their elution order from the column.

Table 4. Average weight (P) of surviving *S. frugiperda* larvae after different days of treatment with active fractions of hexane and chloroform extracts of *I. pauciflora* seeds (1 mg/mL).

Treatments	Six days		Nine days		13 days	
	N	P (mg) (% Effect on weight) [-1]	N	P (mg) (% Effect on weight) [-1]	N	P (mg) (% Effect on weight) [-1]
1E	36	15.76 (+ 159.2) a	36	96.33 (- 45.2) b	36	459.3 (+31.34) a
1G	36	11.97 (+ 96.9) b	36	91.41 (- 47.97) c	36	403.4 (+ 15.35) b
Hexane	36	6.08 (-) c	36	175.72 (-) a	36	349.72 (-) c
Diet	36	6.75 (+ 11.02) c	36	177.5 (+ 1.01) a	36	342.68 (+10.7) c
1A	27	1.89 (- 68.9) e	19	21.19 (- 87.94) e	18	186.37 (- 46.7) d
2G	14	1.49 (- 75.5) ef	10	26.47 (- 84.94) d	10	68.55 (- 80.4) e
1 H	29	0.82 (- 86.5)f g	22	4.97 (- 97.2) gh	16	54.14 (- 84.5) f
1 B	11	0.42 (- 93.1) g	8	6.02 (- 96.6) g	7	51.06 (- 85.4) fg
2D	15	1.65 (- 72.9) e	12	12.55 (- 92.9) f	11	45.89 (- 86.9) fgh
2F	13	0.36 (- 94.1) g	8	14.44 (- 91.8) f	8	43.05 (- 87.7) fghi
1F	9	1.21 (- 80.1) efg	9	4.76 (- 97.3) gh	9	39.99 (- 88.6) ghi
1C	9	1.21 (- 80.1) efg	13	4.29 (- 97.6) gh	13	34.54 (- 90.12) hi
2E	14	0.35 (- 94.1) g	11	2.6 (- 98.5) h	7	20.65 (- 94.1) i
1D	8	2.83 (- 53.5) d	8	4.67 (- 97.34) gh	7	9.3 (- 97.34) j
2B	20	0.52 (- 91.5) g	20	2.31 (- 98.7) h	15	6.39 (- 98.2) j
2C	20	0.604 (- 90.1) g	17	2.59 (- 98.52) h	14	6.17 (- 98.24) j
ANOVA		(P < 0.0001)		(P < 0.0001)		(P < 0.0001)
		C. V. = 8.3		C.V. = 4.95		C. V. = 3.08

Means followed by the same letter do not differ statistically (Tukey, α = 0.05). [1]% Effect on weight = Percentage of decrease (-) or increase (+) of the larval weight with respect to the hexane control. TR = Treatments. 1 = Hexane extract fractions; 2 = Chloroform extract fractions of *I. pauciflora* seeds. A, B,…,H = Fractions obtained and numbered according to their elution order from the column.N = Number of surviving larvae at different days of weighing.

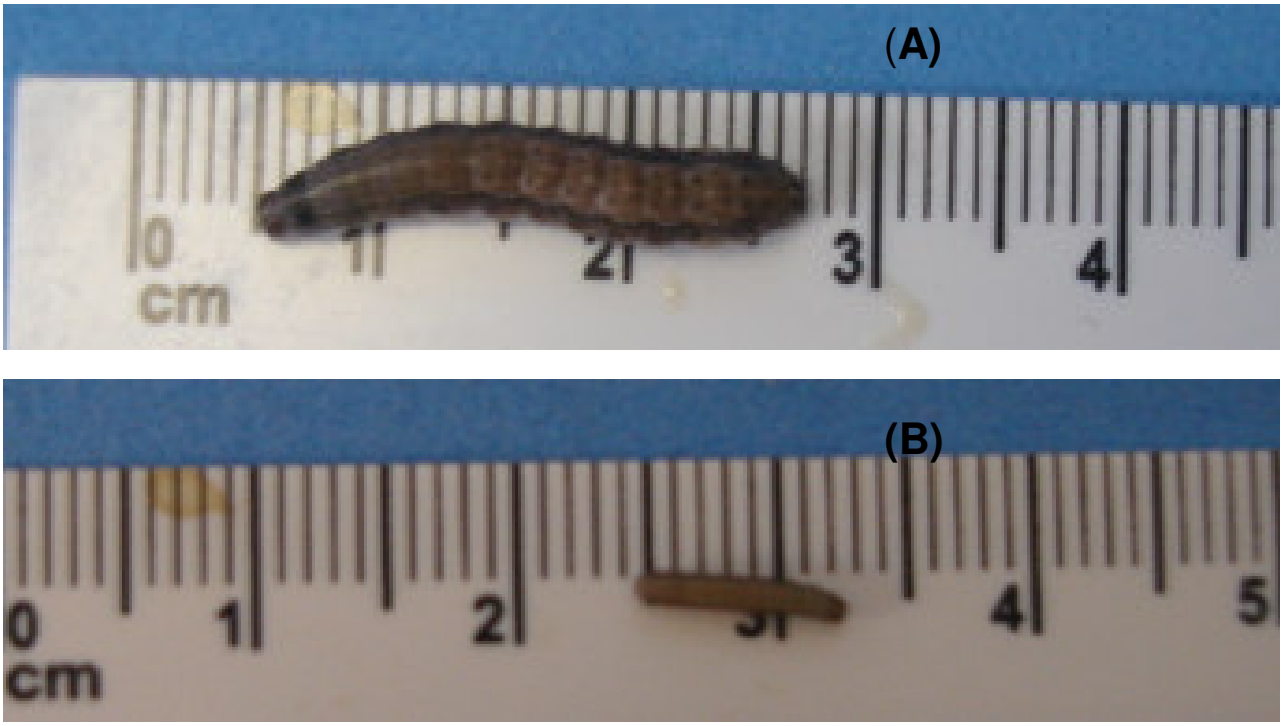

Figure 1. Effect of different fractions of hexane extract of *I. pauciflora* seeds on the weight of *S. frugiperda* larvae: (A) Normal larva (23 mm), (B) Larva fed with fraction 1D (7 mm). Larvae were 14 days old.

to eliminate the toxic compounds in the fractions, due to a loss of appetite. They produced little excrement and probably had less energy for the production of biomass needed for detoxification. These results agreed with those obtained with other Lepidoptera, such as *Ostrinia nubilalis* (Arnason et al., 1985).

Larvae fed with fractions 1E and 1G manifested a constant weight gain during the bioassay. After 13 days of incubation, they reached maximum weights of 459.31 mg ± 1.45 mg and 403.4 mg ± 0.77 mg which represent 31% (fraction 1E) and 15% (fraction 1G) increases over the controls (349.7 mg).

Similar effects were also noted by Abassay et al. (1977) with leaf extracts of *Acokanthera spectabilis* Hook. (Apocynaceae) which possess phagostimulant properties against larvae of *S. littoralis* when sprayed on castor leaves (Prakash and Rao, 1997). Bergaoui et al. (2008), tested the extracts and fractions of petroleum and chloroformic extracts of *Anacyclus cyrtolepidioides* (Asteracea) on 7-day-old larvae of *Tribolium confusum* du Val (Coleoptera: Tenebrionidae) and found phagostimulant and antifeedant activities against larvae after seven days of incubation. These data indicated that chromatographic fractionation of the crude extracts could further classify the specificity of the activity and allow for the separation of the antifeedant and phagostimulant compounds in some fractions (Bergaoui et al., 2008).

Our results suggested high variations of the activities of the fractions from *I. pauciflora* seed extracts: some were lethally toxic while some others only affected the larval weight (decrease or increase).

Phytochemical analysis of the active fractions from the hexane and chloroform extracts of the seeds was carried out. Through coelution with authentic standards and matching of their retention time data and mass spectra by GC-MS, revealed the presence of the following fatty acids and aldehydes in the fractions: hexanoic acid (t_R 8.3 min) *m/z* $[M]^+$ 116 (1), 60 (100); decanoic acid (t_R 15.5 min) *m/z* $[M]^+$ 172 (7), 60 (100); hexadecanoic acid (t_R 21.2 min) *m/z* $[M]^+$ 256 (73), 73 (100); oleic acid (t_R 22.92 min) *m/z* $[M]^+$ 280 (10), 55 (100); octanal (t_R 8.7 min) *m/z* $[M]^+$ 130 (0.1), 41 (100); tras-4-nonenal (t_R 10.1 min) *m/z* $[M]^+$ 140 (1), 55 (100); and trans, trans-2, 4 -decadienal (t_R 13.2 min) *m/z* $[M]^+$ 152 (7), 81 (100). Proton NMR spectra showed an intense signal at 1.2 ppm of the long chain of methylenes, a triplet signal (0.85 ppm) of the methyl group and a triplet signal (2.2 ppm) of the methylene next to the carboxylic group corroborating the presence of carboxylic acids. The signals at 9.9 ppm suggested the presence of aldehydes.

Unsaturated fatty acid metabolites, such as oleic acid, linoleic acid, capric acid and lauric acid exhibited toxic properties against insects (Dev and Koul, 1997; Fatope et al., 2000; Chaubey, 2007). They are antioxidant, anti-fungal (Sang et al., 2002) and are cyclooxygenase-2 and -1 inhibitors (Henry et al., 2002).

In summary, the obtained results suggest that the extracts of *I. pauciflora* could be useful in the search for natural larvicidal compounds.

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