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## Insecticidal Activity of Some Wild Plant Extracts Against Cotton Leafworm, *Spodoptera littoralis* (Boisd.) (Lepidoptera: Noctuidae)

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**Abstract:** The biological and phytochemical studies of four Egyptian wild plants/weed were studied against *Spodoptera littoralis*. *M. senegalensis* had highly significant antifeedant activity on the 3rd instar larvae of *S. littoralis* followed by *Cl. amblyocarpa* and *S. argel*, with 95.52, 45.23 and 32.12%, respectively. Larval mortality was relatively higher in case of *M. senegalensis* and *S. argel*. Extract of *Cl. amblyocarpa* was the only extract which causes malformation to pupae of *S. littoralis*. The highly pupal mortality was recorded in case of *M. senegalensis* extract (80%). The most effective extracts on reduction of pupal weight were *Cl. amblyocarpa* and *A. setifera*. The adult emergence percentage in case of *M. senegalensis* was 20% as compared to 95% in the control and all females moths resulting were deformed and all died before oviposition. The lowest number of eggs was observed in case of *A. setifera* extract and the eggs did not hatch. *A. setifera* and *M. senegalensis* possess both behavioural effects and post ingestive toxicity on the developmental stages of *S. littoralis*. *A. setifera* extract had the superior ovicidal activity on the viability of *S. littoralis* egg masses aged 72, 24 and 48 h, with 72.22, 39.77 and 31.82% reduction, respectively. The longer the post treatment period, the lower the bioefficacy of plant extracts against the target insect. *M. senegalensis* extract showed high latent mortality by the lapse of time, where 88% mortality occurred after 15 days from treatment at initial time and the residual effect remained for 5 days with 18% mortality after 15 days at 5 days post treatment. The lowest effective extract was *S. argel* which was effective for 3 days after treatment.

**Key words:** Botanical extracts, phytochemical constituents, *Spodoptera littoralis*, antifeedant, ovicidal, bioresidual

### INTRODUCTION

Serious problems of genetic resistance by insect species, pest resurgence, residual toxicity, photo toxicity, vertebrate toxicity, widespread environmental hazards and increasing costs of application of the presently used synthetic pesticides have directed the need for effective, biodegradable pesticides (Glenn *et al.*, 1994; Ewete *et al.*, 1996; Guedes *et al.*, 1997; Abd El-Aziz and Sharaby, 1997). This awareness has created worldwide interest in the development of alternative strategies, including the re-examination of using plant derivatives against agriculturally important insect-pests. Plant-derived materials are more readily biodegradable. Some are less toxic to mammals, may be more selective in action and may retard the development of resistance. Their main advantage is that they may be easily and cheaply produced by farmers and small-scale industries as crude, or partially purified extracts. In the last two decades, considerable efforts have been directed at screening

plants in order to develop new botanical insecticides as alternatives to the existing insecticides.

Saleh (1986) mentioned that *Anabasis setifera* is a desert plant from Egypt and its main components were carvacrol and thymol. Kemabonta and Okogbue (2002) tested the insecticidal and ovipositional activity of *Chenopodium ambrosioides* (Chenopodiaceae) against *Callosobruchus maculatus*. Adults of *Ca. maculatus* that emerged from treated eggs decreased significantly in number when compared with the control. Application of *Ch. ambrosioides* (5.0% extract) caused 54% mortality of *Ca. maculatus* adults after 5 days, reduced oviposition by 72.5% as compared to the control and thereafter, reduced emergence of F1 adults to 55% as compared to 81% in the control.

*Solenostemma argel*, is a shrub occurring in wild state in Egypt and Sudan. The herb of the Plant locally called El Hargel. Methanolic extracts of aerial parts of the medicinal plant Argel (*S. argel* (Del.) Hayne), incorporated into rearing media of *Culex pipiens* L.

showed effect on oviposition, egg hatchability and larval viability. The ovicidal effect of *S. argel* was relatively less pronounced, however, the 0.1% concentration reduced egg hatch by 33.7%. Complete suppression of oviposition within the first 2 days was observed, however its action was gradually lost thereafter. Bioactive effects were mainly attributed to the presence of a variety of bioactive organic substances mainly terpenes, pergenine glucosides, alkaloids and sterols (Al-Doghairi *et al.*, 2004).

An active component isolated from *Maytenus rigida* root extracts was reported to show a high degree of antifeedant activity when tested against *Pieris brassica* and *Locusta migratoria* (Monache *et al.*, 1984). The natural products nortriterpene quinone methides (pristimerin, tingenone and 20- $\alpha$ -hydroxytingenone) were isolated from *M. sp.* (Celastraceae) and their effects tested on larvae of codling moth (*Cydia pomonella*, Lepidoptera: Tortricidae). 20- $\alpha$ -Hydroxytingenone was the most active compound, showing lethal, antifeedant and insect growth regulation activities. Pristimerin showed also a high antifeedant activity together with its molt effect suppression. Tingenone showed the lowest activity (Avilla *et al.*, 2000). *Cleome visosa* leaf extract in water and also its juice were reported to show insect repellent and insecticidal activities when tested against the cotton leaf armyworm, *S. litura* (Krishnamurthy Rao, 1982).

The present research is a biological and phytochemical studies of four Egyptian wild plants/weed representing four families collected from the south eastern regions of Egypt, on feeding, growth and metamorphosis against the cotton leaf worm *Spodoptera littoralis*.

## MATERIALS AND METHODS

**Insect maintenance:** A standard laboratory culture of *S. littoralis* was maintained in the laboratory on castor leaves *Ricinus communis* at a temperature of 28 $\pm$ 2°C and 65 $\pm$ 5% RH.

**Preparation of plant extracts:** Four plants belonging to 4 families (Asclepiadaceae, Celastraceae, Chenopodiaceae and Cleomaceae) were extracted according to the method described by Zidan *et al.* (1994).

**Phytochemical screening of the tested plants:** The phytochemical screening of the tested plants was made to detect alkaloids, flavonoids, steroids, terpenoids and tannins according to Romo (1966), Geissman (1962), Wall *et al.* (1964) and Trease (1961), respectively. While, anthraquinones, coumarins and saponins were detected according to Harbone (1973). The major chemical constituents are shown in Table 1.

**Antifeedant activity (choice test):** The antifeedant activity (choice test) of the tested plant extracts were experimented against the third instar larvae of *S. littoralis* as described by (Abd El Aziz and El-Hawary, 1997). Twenty replicates were used for each tested extract. The amount of food consumed per leaf disc was estimated using the following equation (Lwande *et al.*, 1985).

$$\text{Deterrence \%} = (1 - T/C) \times 100$$

Where: T and C represent the percent leaf area consumed (cm<sup>2</sup>) per larva of the treated and control sets, respectively.

**Insecticidal properties of tested plant extracts:** The insecticidal properties of tested plant extracts were tested on newly hatched larvae of *S. littoralis*, according to the procedure described by (Abd El Aziz and El-Hawary, 1997). The percentages of larval mortality, pupal mortality, pupal malformation, pupal weight, adult emergence, adult malformation, number of deposited eggs and egg hatchability were recorded.

**Ovicidal tests:** The ovicidal effects of methanolic plant extracts were tested against *S. littoralis* egg masses, aged

Table 1: Plant classification and the phytochemical screening of tested plants

Species	Family	Chemical constituent							
		Carbohydrates and/or glycosides	Sterols and terpenes	Flavonoids	Tannins	Alkaloids	Saponins	Coumarins	Anthraquinone
<i>Anabasis setifera</i> (Moq.)	Chenopodiaceae	+	+	+	+	+	+	+	-
<i>Cleome amblyocarpa</i> (Linn.)	Cleomaceae	+	+	++	-	-	-	+	-
<i>Maytenus senegalensis</i> (Lam.)	Celastraceae	+	+	+	-	+	±	-	-
<i>Solenostemma argel</i> (Del.) Hayne)	Asclepiadaceae	+	+	++	-	-	±	-	-

(-) Non-detected; (±) Rare; (+); Moderate intense (++) Intense

24, 48 and 72 h old according to the method described by (Abd El Aziz and Sharaby, 1997). The percent hatchability was recorded.

**Bioresidual efficacy of plant extracts:** Cotton plants, *Gossypium barbadense* var. Giza-77, three months old (cultivated in April, 2005) were grown in 60 large pots (30 cm diam.), divided into groups each of 10 pots in greenhouse in NRC. The bioresidual efficacy of the tested plant extracts was experimented according to Zidan *et al.* (1994). Percentages of accumulated larval mortality were estimated 3, 7 and 15 days after feeding on the treated leaves for 24 h and followed by feeding on untreated cotton leaves. Data were corrected for untreated mortality using Abbot's formula (Abbot, 1925).

Results were subjected to statistical analysis according to Snedecor and Cochran (1980).

## RESULTS AND DISCUSSION

**Antifeedant activity (choice test):** Data in (Table 2) showed varying degrees of the antifeedant activities of the methanolic extracts of some plants in choice test against *S. littoralis*. The extract of *M. senegalensis* had highly significant antifeedant properties on the 3rd instar larvae of *S. littoralis*. The antifeedant% reached 95.52%. While, *Cl. amblyocarpa* and *S. argel* showed moderate effects, being 45.23 and 32.12%, respectively. On the other hand, *A. setifera* had strong stimulant effects (-33.03%). From the foregoing results, it appears that *M. senegalensis* had a strong antifeedant activity, while *A. setifera* had highly stimulant effect (phagostimulant activity) against *S. littoralis* larvae. These results in agreement with the finding by Monache *et al.* (1984) who isolated an active component from *M. rigida* root extracts which show a high degree of antifeedant activity when tested against *Pieris brassica* and *L. migratoria*. The natural products nortriterpene quinone methides (pristimerin, tingenone and 20-alpha-hydroxytingenone) were isolated from *M. sp.*

Table 2: Antifeedant activity of methyl extract of different plants against 3rd larval instar of *S. littoralis* in choice test

Plant extract	Leaf disc consumed (cm <sup>2</sup> )/larva in 48 h		
	Treated (T)	Control (C)	Deterrence (%)
<i>Anabasis setifera</i>	29.4	22.1	-33.03
<i>Cleom amblyocarpa</i>	15.5	28.3	+45.23
<i>Maytemus senegalensis</i>	3.0	67.0	+95.52
<i>Solenostemma argel</i>	26.2	38.6	+32.12

(Celastraceae) and their effects tested on larvae of codling moth (*Cydia pomonella*, Lepidoptera: Tortricidae). 20-alpha-Hydroxytingenone was the most active compound, showing lethal, antifeedant and insect growth regulation activities. Pristimerin showed also a high antifeedant activity together with its molt effect suppression. Tingenone showed the lowest activity (Avilla *et al.*, 2000). Itokawa *et al.* (1993) isolated seven novel alkaloids from *M. ebenifolia* Reiss. Also, Saleh (1986) mentioned that the main components of *A. setifera* were carvacrol and thymol. Two constituents of essential oils, carvacrol and thymol, caused a slight reduction of the feeding damage of *Frankliniella occidentalis* (Sedy and Koschier, 2003).

**Insecticidal properties of tested plant extracts:** As shown in Table 3, larval mortality was relatively higher in case of *M. senegalensis* and *S. argel* 71 and 60%, respectively. *Cl. amblyocarpa* was the only extract which causes malformation to pupae of *S. littoralis*. The highly pupal mortality was recorded in case of *M. senegalensis* extract (80%). The pupal weight was highly significantly reduced in all tested plant extract compared with control. The most effective extracts on reduction of pupal weight were *Cl. amblyocarpa* and *A. setifera* with (190.9±1.83 mg) and (192.9±1.89 mg), respectively and there were no significant difference between them. *Cl. visosa* leaf extract in water and also its juice were reported to show insect repellent and insecticidal activities when tested against the cotton leaf armyworm, *S. litura* (Krishnamurthy Rao, 1982). The adult emergence percentage in case of *M. senegalensis* was 20% as compared to 95% in the control and all resulting female moths were deformed and all died before ovipositing. The lowest number of eggs was observed in case of *A. setifera* extract (39.9±2.34) and the eggs did not hatch. In conclusion, *M. senegalensis* and *A. setifera* possess both behavioural effects and post ingestive toxicity on the developmental stages of *S. littoralis*. This finding is in agreement with (Ce'spedesa *et al.*, 2001). 9-benzoyloxy-1 $\alpha$ ,2 $\alpha$ ,6-,8 $\alpha$ ,15-penta-acetoxy-dihydro-agarofuran (1) and from seeds of *M. boaria* 9-furoyloxy-1 $\alpha$ ,6-,8 $\alpha$ -triacetoxy-dihydro-agarofuran (2) were isolated from the aerial parts of *M. disticha*. Compounds 1 and 2 caused 100% larval mortality at 25 and 15 ppm, respectively. MeOH and hexane/EtOAc extracts caused 100% larval mortality at 25.0 ppm, respectively, they also increased the development time of surviving larvae and a significant delay for the time of pupation and adult emergence. Acute toxicity against adults of *S. frugiperda*

Table 3: Insecticidal properties of methanolic extracts of some plants on the developmental stages of *S. littoralis*

Plant extract	Larval mortality (%)	Pupal mortality (%)	Malformed pupae (%)	Pupal weight (mg) ±SE (range)	% Adult emergence	Malformed adult (%)	No. of deposited eggs±SE (range)	Egg hatchability (%)
<i>Anabasis setifera</i>	46.67	37.50	0.00	192.9±1.89 (184-202)d	62.50	0.00	39.9±2.34 (28-51)d	0.00
<i>Cleom amblyocarpa</i>	50.00	33.00	15.00	190.9±1.83(180-199)d	55.56	5.0	1031.1±2.97 (1017-1045)c	65.00
<i>Maytenus senegalensis</i>	71.00	80.00	0.00	287.3±1.84(279-296)b	20.00	0.00	0.00	0.00
<i>Solenostemma argel</i>	60.00	0.00	0.00	243.7±1.86 (235-252)c	90.00	0.00	1167.5±6.51 (1140-1200)b	85.00
Control	10.00	0.00	0.00	319.5±1.43 (312-327)a	95.00	0.00	1505±6.99 (1470-1540)a	85.00

Means followed by the same letter are not significantly different ANOVA (LSD) p>0.05

Table 4: Effect of methanolic extract of different plants on viability of *S. littoralis* egg masses

Plant extract	(% Hatchability of different egg ages)		
	24 h	48 h	72 h
<i>Anabasis setifera</i>	53.0c (39.77)	60.0c (31.82)	25.0e (72.72)
<i>Cleom amblyocarpa</i>	85.0a (3.41)	84.0a (4.55)	41.0c (54.44)
<i>Maytenus senegalensis</i>	61.0b (30.68)	86.0a (2.27)	60.0b (33.33)
<i>Solenostemma argel</i>	87.0a (1.14)	71.0b (19.32)	30.0d (66.67)
Control	88.0a	88.0a	90.0a

Number between brackets represents percent decrease than check, Means followed by the same letter(s) (vertical difference) are not significantly different

was also found, for hexane/EtOAc extract and 2 had the most potent activity with LD50 value of 4.7 and 1.9 ppm, respectively. MeOH extract, hexane/EtOAc extract, 1 and 2 caused acetylcholinesterase inhibition with 78.0, 89.2, 79.3 and 100% inhibition at 15.0 ppm, respectively (Ce'spedes *et al.*, 2001).

**Ovicidal activity of tested plant extracts:** Data concerning the ovicidal activity of the tested plant extracts (Table 4) clearly indicate the important role played by age of eggs and nature of tested plant in determining the ovicidal activity against *S. littoralis* egg masses.

Considering the age of eggs, data indicate, in general, the higher susceptibility of the older eggs (72 h old) to the tested plant extracts followed by 24 h old eggs in comparison to egg masses (48 h old). Data indicated the superior ovicidal activity of *A. setifera* extract on the viability of egg masses of *S. littoralis*, aged 72, 24 and 48 h, with 72.22, 39.77 and 31.82% reduction, respectively. *M. senegalensis* extract showed moderate reduction in egg hatchability. Only the older eggs (72 h old) had affected with *Cl. amblyocarpa* and *S. argel* treatments.

This finding is in agreement to some extent with the results obtained by Abd El-Aziz and Sharaby (1997) on *S. littoralis* egg masses treated with different concentrations of white mustard oil. This may be due to the inhibiting influence of the chemical agent on the vital enzymes which have been directed at the late stage of embryonic development of eggs (Smith and Salked, 1966).

**Bioresidual efficacy of plant extracts:** Data in Table 5 indicated the important role of post treatment period and

plant species in determining the bioresidual activity of the tested extracts against the 4th instar larvae of *S. littoralis* under potted condition. The longer the post treatment period, the lower the bioefficacy of plant extracts against the target insect. A moderate knock down effect was recorded with initial time samples. Larvae fed on treated cotton leaves for one day and followed by untreated ones, showed percentage of mortality after 3 days, 50, 43, 31 and 20% in case of *M. senegalensis*, *Cl. amblyocarpa*, *A. setifera* and *S. argel*, respectively. *M. senegalensis* extract showed high latent mortality by the lapse of time, where 88% mortality occurred after 15 days from treatment at initial time and the residual effect remained for 5 days with 18% mortality after 15 days at 5 days post treatment. The lowest effective extract was *S. argel* which was effective for 3 days after treatment. Kivan (2005) investigated the effect of azadirachtin (NeemAzal T/S) on different stages of the sunn pest, *Eurygaster integriceps* Put. in the laboratory. No effect was observed for 1st instar nymphs at 1 day after application, although adults had slightly effect (20%). Adults and nymphs were influenced 7 days after treatment and mortality rates for adults and nymphs were recorded 44.0 and 51.9%, respectively.

The foregoing results indicate that the tested plant extracts have properties which cause feeding deterrence, larval mortality, retardation in the developmental stages, pupal and adult morphogenesis, reduction in fecundity and viability of *S. littoralis* and persistent on cotton plants and this may be correlated to the chemical constituents of these plants (Table 1). For instance, flavonoids were reported to be toxic to some insects (Salama *et al.*, 1970). The effect of tannins in growth inhibition of lepidopterous larvae was reported by Klock and Chan (1982). Kogan (1986) mentioned that alkaloids had repellent, toxic and feeding inhibition effects on some insect species. Sterols were also reported by Nayer and Fraenkel (1962) to inhibit the feeding of some insects. So, it is evident that these chemicals as constituents of the tested plants have properties which inhibit feeding and cause retardation in the larval development, pupal and adult morphogenesis of *S. littoralis*.

Table 5: Accumulated corrected mortality on 4th instar larvae of *S. littoralis*, fed on cotton leaves, 0-5 days after being treated with plant extracts

Plant extract	Days after treatment of cotton plants																	
	Corrected percentage larval mortality (days after treatment)																	
	Initial			1			2			3			4			5		
	3	7	15	3	7	15	3	7	15	3	7	15	3	7	15	3	7	15
<i>Anabasis setifera</i>	31	54	73	26	44	56	20	36	42	14	20	25	0	15	20	0	6	8
<i>Cleom amblyocarpa</i>	43	66	78	34	51	71	25	41	50	18	31	37	15	0	0	0	10	0
<i>Maytenus senegalensis</i>	50	60	88	46	52	82	40	56	62	31	45	49	22	29	0	0	14	18
<i>Solenostemma argel</i>	20	40	60	15	20	35	0	13	19	0	0	12	0	0	0	0	0	0

Therefore, it can be concluded that, *M. senegalensis* and *A. setifera* extracts were effective in suppressing the population size of *S. littoralis* either directly through their acute toxic effects on the larvae and egg masses or indirectly through their delayed effects on the pupae and adults and minimizing the cotton infestation by the cotton leaf worm, *S. littoralis* at the vegetative growth stage.

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