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Research Article

ANTI-INSECT PROPERTIES OF *ARGEMONE MEXICANA* L. PLANT PART SOLVENT EXTRACTS AGAINST *SPODOPTERA LITURA* FAB

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ABSTRACT

Studies on the effect of acetone, ethyl acetate, petroleum ether and methanol extract of various plant parts of *Argemone mexicana* L. against *Spodoptera litura* Fab. revealed presence of various anti insect properties such as feeding deterrence, insecticidal and insect growth regulatory activities. Among the solvents tested acetone imparted maximum antifeedant action of 98.01 per cent followed by methanol. Between the plant parts tested the seed extract had shown higher feeding deterrence followed by leaf extract. Various solvent extracts of other plant parts failed to exhibit significant feeding deterrence (> 60%). Supreme insecticidal action was noticed only in methanol extract of seed (80% larval mortality) and it caused complete death of all the treated insects (Nil adult emergence). Insect growth regulatory activity alone was noticed as the supreme anti insect action in ethyl acetate solvent extract. It caused nil adult emergence by imparting 40 and 60 per cent larval and adult malformations respectively. Among the solvent extracts, petroleum ether exhibited minimum anti insect effects in all the plant parts tested.

INTRODUCTION

The global losses due to insect pests have been estimated as 10.8% towards the beginning of this century and in India, the crop losses have been around 15.7% at present. In terms of monetary value, Indian agriculture currently suffers an annual loss of about US$ 36 billion (Dhaliwal *et al.*, 2015). An estimated one third of global agricultural production valued at several billion dollars is destroyed annually by over 20,000 species of insect pests in field and storage (Mariapackiam and Ignacimuthu, 2008). Indiscriminate use of chemical pesticides over the years to combat these menace, has adversely affected non-target organisms, human health and resulted in environmental contamination. It has also promoted development of resistance in many insect species. In such a scenario, eco-friendly, biodegradable botanicals are being considered as viable alternatives (Ignacimuthu, 2004). Botanicals often cause varieties of anti insect actions such as repellence, feeding deterrence, growth regulatory, insecticidal *etc*. Among many promising botanicals seeds of herbaceous medicinal plant Mexican prickly poppy *Argemone mexicana* L. (Papaveraceae) contains toxic alkaloids *viz.*, sanguinarine, dihydrosanguinarine and berberine. The effective insecticidal action of *A. mexicana* extract against phytophagous insects and mosquitoes has been proved by many authors (Sakthivadivel *et al*., 2012; Kangade and Zambare, 2013; Abou-Elnaga, 2015; Sharma *et al.*, 2016 and Sivaraman *et al*., 2016). However, the information regarding the suitable extraction solvent and their effects on target species are scanty. Hence, the present investigation was conducted to study the anti-insect properties of various solvent extracts of different plant parts of *A. mexicana* against third instar larvae of *S. litura*.

MATERIALS AND METHODS

Mass culturing of Spodoptera litura Fab

Tobacco caterpillar, *Spodoptera litura* Fab. (Noctuidae: Lepidoptera) egg masses were collected from the castor plants grown in and around Annamalainagar (Latitude:11°N, Longitude: 79°E). The emerging larvae were maintained in castor leaves up to second instar. Then, they were reared in Bengal gram flour based semi synthetic diet till pupation. The pupae were collected, cleaned, surface sterilized with 0.05% sodium hypochlorite, sexed and transferred into an oviposition cage. Egg masses laid were collected daily, sterilized with 0.05 per cent sodium hypochlorite solution and a continuous culture was maintained. The rearing is done at 26 ± 1°C and 75 per cent relative humidity (PDBC, 1998).

Collection and extraction of plant materials

A. mexicana whole plants were collected from Annamalainagar and shade dried. Leaves, flowers, seeds, stem and roots were

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separated, powdered by using wiley mill. Cold solvent extraction method described by Jaglan *et al*. (1997) was followed for extracting active principle from leaf powder.

Powdered plant material was packed as 100 g packets by using Whatman No.40 filter paper and placed inside stoppered round bottom flasks (Capacity: 2 lit.) at the rate of five packets per flask. The flasks were then filled with one liter of respective solvents separately and kept for 72 h at room temperature. Then the extract was filtered and concentrated under reduced pressure by a rotary flash vacuum evaporator. The semisolid extract thus obtained was stored in deep freezer at –20°C (Selvamuthukumaran and Arivudainambi, 2010).

Anti-insect activity bioassay

A no-choice leaf disc assay was carried out using 4 h pre-starved third instar *S. litura* larvae (Bentley *et. al*., 1984). Castor leaf discs (3 cm diameter) were cut out and treated with 300 µl of undiluted solvent extracts separately on both the sides. After shade drying for one minute, leaf discs were placed separately inside a Petri plate (9 cm diameter) lined internally by moist filter paper to avoid early drying. Each Petri plate was provided with one 4 h pre starved third instar larvae and each treatment was replicated ten times. Solvent and absolute controls were also maintained.

Treated leaf discs were collected from the containers after six hours. Then, the leaf area fed was measured graphically and per cent feeding deterrence was computed. The larvae alive were reared using untreated castor leaves till adult emergence and mortality and malformations were recorded (Selvamuthukumaran and Arivudainambi, 2008).

$$\text{Percent feeding deterrence} = \frac{\text{Leaf disc consumed by the larvae in control} - \text{Leaf disc consumed by the larvae in treated}}{\text{Leaf disc consumed by the larvae in control} + \text{Leaf disc consumed by the larvae in treated}} \times 100$$

RESULTS AND DISCUSSION

The result of the leaf disc bioassay with various solvent extract of different parts of *A. mexicana* revealed presence of various anti insect properties such as feeding deterrence, insecticidal and insect growth regulatory activities.

Among the solvents tested acetone imparted maximum feeding deterrence action (Fig. 1) followed by methanol. Between plant parts tested the seed extract deterred feeding effectively followed by leaf extract. The acetone extract of seed provided maximum feeding deterrence (98.01%) and resulted in minimum adult emergence (20%). Further, it exhibited very minimal (20% to nil) deformities in growth stages except seed extract. Such similar result was also obtained for larval or pupal mortality.

Supreme insecticidal action was noticed only in methanol extract of seed (80% larval mortality) (Fig. 2). It also resulted in complete death of all the treated insects (Nil adult emergence). However methanol extract of other plant parts showed minimum or nil insecticidal activity (Table 4). This finding was in accordance with the report of Pandey *et al*. (1981). They established the supremacy of *A. mexicana* seed extract which caused 93.33 per cent insecticidal activity in *Bagrada cruciuferarum* (Krik) attacking turnip. They further elaborated that the leaf extract was having lesser action than seed extract which resulted in 86.66 per cent insecticidal activity. Sanguinarine is one of the main insecticidal ingredients. Measurements taken on the effects of sanguinarine on six metabolic enzymes of third instar larvae of *Pieris rapae* showed that the activities of carboxyl esterase, alkaline phosphatase, acid phosphatase and cytochrome P450 in treated larvae were activated by sanguinarine while the activity of acetylcholinesterase was inhibited (Chunmei *et al*., 2013). This finding also corroborated the reason for presence of insecticidal action.

The methanol extract failed to show any significant (>50%) malformations in any of the growth stages. Similarly apart from seed extract (79.93 %) other plant parts exhibited minimum feeding deterrence (>50%) when extracted with methanol (Table 4).

Insect growth regulatory activity alone was noticed as the supreme anti insect action in ethyl acetate solvent extract. It caused nil adult emergence by imparting 40 and 60 per cent larval and adult malformations respectively (Fig. 3). The ethyl acetate extract failed to show any insecticidal effect and as well significant feeding deterrence (>50%) (Table 2). Similar trend of growth regulatory activity was noticed by Malarvannan *et al*. (2008). They reported that different treatments differed significantly in their efficacy. They further reported occurrence of growth deformities such as larval-pupal intermediates, malformed moths, dead pupae etc. Among the solvent extracts, petroleum ether exhibited minimum anti insect effects in all the plant parts tested (Table 3).

The present study revealed presence of multifarious anti insect properties of *A. mexicana*, which provides scope for futher product development after in depth mode of action and toxicological studies.

Figure 1. Leaf disc showing the maximum feeding deterrence activity when treated with acetone seed extract

Figure 2. Insecticidal action noticed in *S. litura* larva after treatment with methanol extract

(a) **(b)** **(c)**

Figure 3. Malformation noticed in *S. litura* (a) larva, (b) Pupa and (c) Adult after treatment with ethyl acetate seed extract

Table 1 Anti insect effects of *Argemone mexicana* L. acetone extract on third instar *Spodoptera litura* Fab.

Plant parts	Percent feeding deterrence activity*	Percent mortality*		Percent malformations*			Percent adult emergence*
		Larva	Pupa	Larva	Pupa	Adult	
Seed	98.01 (81.87)	20 (26.56)	0 (0.0)	20 (26.56)	40 (39.23)	0 (0.0)	20 (26.56)
Leaf	53.12 (46.78)	0 (0.0)	0 (0.0)	20 (26.56)	0 (0.0)	0 (0.0)	80 (63.44)
Stem	31.12 (33.89)	0 (0.0)	0 (0.0)	0 (0.0)	20 (26.56)	0 (0.0)	80 (63.44)
Root	40.29 (39.41)	0 (0.0)	20 (26.56)	20 (26.56)	20 (26.56)	20 (26.56)	80 (63.44)
Flower	21.21 (27.42)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	90 (71.56)
Solvent control	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	90 (71.56)
Absolute control	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	90 (71.56)
S.Ed	0.699	0.095	0.095	0.134	0.121	0.095	0.105
CD	1.541	N.S.	N.S.	0.296	0.266	N.S	0.231

*Mean of ten replications
Values within parentheses are arc sine transformed

Table 2 Anti insect effect of ethyl acetate extract of various plant parts of *Argemone mexicana* L. on third instar of *Spodoptera litura* Fab.

Plant parts	Percent feeding deterrence activity*	Percent mortality*		Percent malformations*			Percent adult emergence*
		Larval	Pupal	Larval	Pupal	Adult	
Seed	45.74 (42.53)	0 (0.0)	0 (0.0)	40 (39.23)	0 (0.0)	60 (50.77)	0 (0.0)
Leaf	33.12 (35.12)	0 (0.0)	0 (0.0)	0 (0.0)	20 (26.56)	0 (0.0)	80 (63.44)
Stem	30.16 (33.34)	0 (0.0)	0 (0.0)	20 (26.56)	0 (0.0)	0 (0.0)	80 (63.44)
Root	38.19 (38.17)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	20 (26.56)	80 (63.44)
Flower	20.12 (26.64)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	90 (71.56)
Solvent control	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	90 (71.56)
Absolute control	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	90 (71.56)
S.Ed	0.093	-	-	0.101	0.095	0.101	0.164
CD	0.205	-	-	0.224	0.209	0.224	0.362

*Mean of ten replications
Values within parentheses are arc sine transformed

Table 3 Anti insect effect of petroleum ether extract of various plant parts of *Argemone mexicana* L. on third instar of *Spodoptera litura* Fab

Plant parts	Percent feeding deterrence activity*	Percent mortality*		Percent malformations*			Percent adult emergence
		Larval	Pupal	Larval	Pupal	Adult	
Seed	56.04 (48.45)	0 (0.0)	0 (0.0)	20 (26.56)	20 (26.56)	0 (0.0)	60 (50.77)
Leaf	31.33 (34.02)	0 (0.0)	0 (0.0)	0 (0.0)	20 (26.56)	20 (26.56)	60 (50.77)
Stem	28.13 (32.01)	0 (0.0)	0 (0.0)	0 (0.0)	20 (26.56)	0 (0.0)	80 (63.44)
Root	25.12 (30.07)	0 (0.0)	0 (0.0)	0 (0.0)	20 (26.56)	0 (0.0)	80 (63.44)
Flower	16.14 (23.66)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	90 (71.56)
Solvent control	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	90 (71.56)
Absolute control	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	90 (71.56)
S.Ed	0.110	-	-	0.095	0.134	0.095	0.140
CD	0.243	-	-	0.209	0.296	0.209	0.309

*Mean of ten replications
Values within parentheses are arc sine transformed

Table 4 Anti insect effect of methonal extract of various plant parts of *Argemone mexicana* L. on third instar of *Spodoptera litura* Fab.

Plant parts	Percent feeding deterrence activity*	Percent mortality*		Percent malformations*			Percent adult emergence*
		Larval	Pupal	Larval	Pupal	Adult	
Seed	79.93 (63.44)	80 (63.44)	0 (0.0)	0 (0.0)	20 (26.56)	0 (0.0)	0 (0.0)
Leaf	40.12 (39.29)	20 (26.56)	20 (26.56)	0 (0.0)	20 (26.56)	0 (0.0)	40 (39.23)
Stem	33.16 (35.18)	0 (0.0)	0 (0.0)	20 (26.56)	0 (0.0)	20 (26.56)	60 (50.77)
Root	40.12 (39.29)	0 (0.0)	20 (26.56)	20 (26.56)	0 (0.0)	20 (26.56)	40 (39.23)
Flower	23.17 (28.79)	0 (0.0)	20 (26.56)	0 (0.0)	0 (0.0)	0 (0.0)	80 (63.44)
Solvent control	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	90 (71.56)
Absolute control	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	90 (71.56)
S.Ed	0.099	0.123	0.134	0.123	0.123	0.123	0.168
CD	0.217	0.271	0.296	0.270	0.270	0.270	0.371

*Mean of ten replications
Values within parentheses are arc sine transformed

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