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Evaluation of antifeedant activity of different parts of *Calotropis gigantea* against *Helicoverpa armigera*

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Abstract

Milkweed plant or erukku (*Calotropis gigantea* R. Br.). (Family Asclepiaceae) a common waste land weed distributed in tropical and subtropical region of Asia. It possess many toxic principles which protects the crops from pest attack. *Helicoverpa armigera* (Hubner), is a polyphagous noctuid pest causing heavy damage to agricultural, horticultural and ornamental crops and developed resistance to insecticides posing problem to the farmers. Therefore alternate control measures needs to be developed which would be eco friendly and safe to the environment. In the present study, the antifeedant activity of *C.gigantea* was evaluated from its leaf, flower, stem, root and whole plant against third and fourth instar larvae, *Helicoverpa armigera*. The results suggested that 10 per cent concentration of leaf, flower, stem, root and whole plant extract recorded the minimum area consumption (20.36%, 30.46%, 48.03 %, 59.00 % and 35.06%) on third instar larvae. Similarly, 30.93%, 38.23%, 48.46%, 52.00% and 36.13% of area consumption in leaf, flower, stem, root and whole plant against fourth instar larvae of *H.armigera*. Higher the concentration of plant extracts, higher the antifeedant activity. Among the different plant parts, leaf extract showed strong antifeedant effect.

Keywords: milk weed, *Calotropis gigantea*, *Helicoverpa armigera*, antifeedant

Introduction

Milk weed plant or erukku (*Calotropis gigantea* R. Br.). (Family Asclepiaceae), a common waste land weed distributed in tropical and subtropical region of Asia (Mueen Ahmed *et al.*, 2005) is gaining importance in recent years as a potential pesticidal source against insect pests, besides having several medicinal and industrial uses (Meshram, 1995) [12]. It is known to have insecticidal (Solunke and Deshpande, 1991) [24], antifeedant (Pari *et al.*, 1998) [15], nematicidal (Philip *et al.*, 1993) [16], antitermiticidal (Badshah *et al.*, 2004) [5], antibacterial and antifungal properties (Anil Srivatsava *et al.*, 2000) [3]. The plant extract of *C.gigantea* proved effective against lepidopterous and sucking pests of several crops (Pugalenthi *et al.*, 1994 [18]; Muhammad *et al.*, 2003) [14]. Arulprakash and Senthilkumar (2005) [4] evaluated the efficacy of *C. gigantea* plant parts extract against *Callosobruchus maculatus* (Fab.) and the results showed that mortality of pulse beetle, seven days after treatment was highest in whole plant powder (68.28%) treatment followed by leaf, flower, stem and root respectively.

Calotropis procera exhibited excellent antimicrobial and insecticidal activity against common microbial contaminants and insect pests of pulses and mortality of *C. chinensis* was found to be dose dependent with maximum mortality (99%) at 90 µg ml⁻¹ after 9 hrs of treatment (Mendki *et al.*, 2005) [11]. Aqueous extract of leaf, flower and roots of *C. procera* proved most effective in the control of *Henosepilachna elaterii* showing strong repellent activity. Five per cent extract of different plant parts gave 100 per cent protection of cucurbit leaf and no larva survived after exposure to 5 per cent extract. One and 2.5 per cent concentrations of *Calotropis* highly reduced the fecundity and longevity of the insect (Ahmed *et al.*, 2006) [2]. Aqueous extract of *C. gigantea* plant parts, 5 per cent leaf extract provided maximum mortality of *Aphis gossypii* Glover (60.00%), *Epilachna vigintioctopunctata* Fab. (76.66%), *L. orbonalis* G. (63.33%), *B. tabaci* Genn. (60.00 %), *E. vitella* F. (56.67%), *H. armigera* (76.66%) and *Liriomyza trifolii* Burgess (23.33%) (Kanimozhi, 2006) [8].

Helicoverpa armigera (Hubner), is a polyphagous noctuid pest causing heavy damage to agricultural, horticultural and ornamental crops (Talekar *et al.*, 2006) [27]. Also it developed resistance to insecticides posing problem to the farmers (Liu *et al.*, 2000) [9]. *Helicoverpa armigera* developed resistance to conventional insecticides. Several insecticides and pesticides are used to control *H. armigera*. However, harmful effects and persistent nature of the chemical pesticides demand for eco-friendly alternatives (Ramya *et al.*, 2008) [19]. Therefore in the present study, an attempt has been made to evaluate the antifeedant activity of *C.gigantea* against *Helicoverpa armigera*.

Materials and Methods

Collection and preparation of *Calotropis gigantea*

C. gigantea R. Br plant parts viz., leaves, stem, flower and roots were collected from waste lands. Fresh plant materials were used to prepare the aqueous suspensions. The plant materials were washed thoroughly with water to remove dirt, if any and chopped into small pieces with a sharp knife. Chopped pieces were then crushed into a fine paste with the help of a pestle and mortar. The paste thus obtained was diluted to 100 ml solution with distilled water and filtered through a fine muslin cloth. Two, four, six, eight and 10 per cent concentrations were prepared and 0.1 ml Teepol was added as a sticking agent to the filtrate (Rani, 2005) [20].

Evaluation of antifeedant activity of *Calotropis gigantea*

Leaf disc bioassay method was adopted for evaluating the antifeedant property of *C. gigantea* plant extracts. Leaf discs of uniform area were made from the host plants and measured prior to the commencement of the experiment. The leaf discs were dipped for two minutes in aqueous extracts at different concentrations (2, 4, 6, 8 and 10 %) and then transferred to petridishes of 10 cm diameter. Third and fourth instar larvae were pre-starved for six hours and released into each experimental unit. Larvae were also released on leaf discs dipped in water served as control. Three replications were kept for each treatment. The larvae were allowed to feed on the leaf discs for 24 hrs and then the leaf area consumed/protected was measured using the formula stated by Jacob and Sheila (1994) [7] as given below.

$$\text{Per cent leaf area protection over control} = \frac{\% \text{ leaf area protection in treatment} - \% \text{ leaf area protection in control}}{100 - \% \text{ leaf area protection in control}} \times 100$$

The antifeedant activity of the different plant extracts was rated as per the scale given below.

- | | |
|---|---|
| 1. Strong inhibition (++++)
(High antifeedant activity) | When the feeding ratio is below 20 |
| 2. Medium inhibition (+++)
(Medium antifeedant activity) | When the feeding ratio is between 20-50 |
| 3. Weak inhibition (++)
(Weak antifeedant activity) | When the feeding ratio is between 50-80 |

- | | |
|---|------------------------------------|
| 4. Low inhibition (+)
(Low antifeedant activity) | When the feeding ratio is above 80 |
|---|------------------------------------|

Statistical analysis

The data collected in various experiments were statistically analysed using randomized block design. The percentage data obtained in the experiments were converted into corresponding angles (Arc-Sine Percentage for Statistical interpretation) and analysed (Snedecor and Cochran, 1968) [23].

Results and Discussion

Antifeedant effect of aqueous extract of *Calotropis gigantea* on third and fourth instar larvae of *H. armigera*

The antifeedant effect of various aqueous extract of *C. gigantea* viz., leaf, flower, stem, root and whole plant were tested on third instar larvae of *Helicoverpa armigera*. From the data, it was observed that 10 per cent concentration of leaf, flower, stem, root and whole plant extract recorded the minimum leaf area consumption (20.36%, 30.46%, 48.03 %, 59.00 % and 35.06%), which was on par with eight per cent (24.33 %, 33.53 %, 54.26%, 68.46 and 36.23 %) concentration. The maximum leaf area consumption was observed in 2 per cent concentration of leaf (84.13%), flower (89.33%), stem (93.03 %), root (94.33%) and whole plant (90.13%) which showed low antifeedancy (Table 1).

Similarly, the antifeedant effect of various aqueous extract of *C. gigantea* on fourth instar larvae of *H. armigera* were also studied. The minimum leaf area consumption was recorded at ten per cent concentration of leaf (30.93%), flower (38.23%), stem (48.46%), root (52.00%) and whole plant extract (36.13%) (Table 2). The maximum leaf area consumption was observed at two per cent concentration leaf (88.56%), flower (90.73%), stem (93.03%), root (94.33%) and whole plant extract (92.33%).

Among the different plant parts, the leaf extract showed minimum leaf area consumption and it was maximum in root extracts. The leaf extract at 10 per cent concentration showed that the average leaf area consumed was very little than other concentration tested and control. The leaf area consumed was 20.36 per cent in case of third instar and it was 30.83 per cent in fourth instar respectively. The leaf area consumed was directly proportional to the various concentrations used. From the present study, it is clear that antifeedant activity was found to be concentration dependent and the less leaf area consumed by the *H. armigera* was due to the phytochemicals produced by *Calotropis gigantea*

Table 1: Antifeedant effect of aqueous extract of leaf, flower, stem, root and whole plant of *Calotropis gigantea* on third instar larvae of *Helicoverpa armigera*

Treatments	Leaf		Flower		Stem		Root		Whole plant	
	Leaf area consumed*	Rating								
2%	84.13 (66.52) ^b	+	89.33 (70.93) ^b	+	93.03 (75.03) ^b	+	94.33 (76.23) ^b	+	90.13 (71.69) ^b	+
4%	73.53 (59.03) ^c	++	76.46 (60.93) ^c	++	87.76 (69.52) ^c	+	89.53 (71.09) ^c	+	78.46 (62.35) ^c	++
6%	30.66 (33.62) ^d	+++	48.26 (44.00) ^d	+++	63.73 (52.95) ^d	++	74.26 (59.51) ^d	++	52.33 (46.31) ^d	++
8%	24.33 (29.53) ^e	+++	33.53 (35.36) ^e	+++	54.26 (47.44) ^e	++	68.46 (56.78) ^d	++	36.23 (37.00) ^e	+++
10%	20.36 (26.82) ^e	+++	30.46 (33.50) ^f	+++	48.03 (43.87) ^f	+++	59.00 (50.18) ^e	++	35.06 (36.31) ^e	+++
Control	100 (85.94) ^a	+								
SED	0.25		0.20		1.39		0.51		0.29	
CD (P = 0.05)	0.57		0.46		3.11		1.15		0.64	

*Values in parentheses are arc-sine transformed values

Mean values with various alphabets differ significant

(+) low inhibition, (++) weak inhibition, (+++) medium inhibition, (++++) strong inhibition.

Table 2: Antifeedant effect of aqueous extract of different plant parts of *Calotropis gigantea* on fourth instar larvae of *Helicoverpa armigera*

Treatments	Leaf		Flower		Stem		Root		Whole plant	
	Leaf area consumed*	Rating								
2%	88.56 (70.23) ^b	+	90.73 (72.28) ^b	+	93.03 (75.03) ^b	+	94.33 (76.36) ^b	+	92.33 (73.92) ^b	+
4%	80.96 (64.13) ^c	+	87.76 (67.00) ^c	+	91.83 (73.37) ^b	+	92.06 (73.64) ^c	+	88.56 (70.18) ^b	+
6%	50.46 (45.26) ^d	++	55.73 (48.29) ^d	++	66.83 (54.81) ^c	++	71.33 (57.60) ^d	++	58.36 (49.81) ^c	++
8%	33.26 (35.22) ^e	+++	42.56 (40.68) ^e	+++	50.38 (44.22) ^d	++	68.23 (55.69) ^d	++	45.66 (42.51) ^d	+++
10%	30.83 (33.72) ^f	+++	38.23 (38.17) ^f	+++	48.46 (44.08) ^d	+++	52.00 (46.14) ^e	++	39.13 (38.72) ^e	+++
Control	100 (85.94) ^a	+								
SED	0.23		0.42		1.36		0.92		0.28	
CD (P = 0.05)	0.52		0.94		3.05		2.06		0.63	

*Values in parentheses are arc-sine transformed values

Mean values with various alphabets differ significant

(+) low inhibition, (++) weak inhibition, (+++) medium inhibition, (++++) strong inhibition.

Plants produced a range of secondary metabolites like alkaloids, terpenoids, flavonoids, phenols, glycosides, sitosterols and tannins which are easily degradable when applied in a crop ecosystem. These phytochemicals help in protecting the plants from the attack of insect-pests (Ahmad, 2007) [1]. However, production of phytochemicals varies from plant to plant. Further, parameters like age of plant, part of plant (root, stem, leaf, fruit, flower, seed and bark) have been reported to affect the production of phytochemicals. The phytochemicals produced in response to insect-pest attack, affect feeding and oviposition of insects on the plants. The active compounds may act as antifeedants, disturb insect growth, development and inhibit oviposition.

Jacob and Sheila (1994) [7] reported that antifeedant activity of aqueous leaf extract of *Calotropis* sp. at five per cent concentration on *Pericallia ricini* F. showed feeding of 28.91 per cent. Similar antifeedant effect of *Calotropis procera* was reported by Meshram (1995) [12] against teak skeletonizer. Dodia *et al.* (1995) [10] showed the antifeedant property of aqueous extracts of *C. procera* against castor semilooper, *Achaea janata* F. Pari *et al.* (1998) [15] reported the antifeedant activity of gigantocine, a novel non-protein amino acid isolated from root bark of *C. gigantea* using petroleum ether as solvent, against nymphs of the desert locust *Schistocerca gregaria* (Forsk.) in the present study.

Saikia and Paresmeswaran (2000) [21] also found similar results with neem, *Vitex negundo*, *Allium cepa* and *C. gigantea* plant extracts against *Cnaphalocrocis medinalis*. So the results of the present study confirmed that *C. gigantea* plant parts possessed repellent property. The antifeeding property of crude foliar extracts of *C. gigantea* on third instar larvae of *S. litura* has been reported by Praveen *et al.* (2002) [17]. Results of present investigation were also found to be similar with the results of Manikantan (2003) [10] who reported that leaf extracts of *C. gigantea* at five per cent gave maximum leaf area protection with minimum leaf area (45.12%) consumed by *S. litura*.

Researchers have reported the presence of effective phytochemical constituents which are responsible for its biological activities especially its insecticidal activity, in the various parts of *Calotropis gigantea* especially in the leaves (Suresh Kumar *et al.*, 2013) [26]. Sumathi *et al.* (2017) [25] studied the bioefficacy of *Calotropis gigantea* flower extracts tested against papaya mealy bug infestation in *Ailanthus excelsa* found that insecticidal activity (90-95%) at 2000 ppm within 24 hours of treatment. The present study proved that *C. gigantea* found to possess antifeedant activity. Shumaia Parwin *et al.* (2014) [22] evaluated therapeutic potentials and insecticidal effects of *gigantea* leaves against pathogenic bacteria, fungi, and insects. The insecticidal activity was better

with 80% mortality rate at a dose of 50 mg/ml in 48 hours whereas n-hexane fraction showed 40% mortality rate.

Conclusion

The antifeedant effect of *Calotropis gigantea* viz., leaf, flower, stem, root and whole plant were evaluated for its repellent effect on third and fourth instar larvae of *Helicoverpa armigera*. Higher the doses of plant extracts greater the antifeedant activity. The active principles present in the *calotropis* may produce repellent effect leading to avoidance of host plants. Plants based antifeedants not only played a major role in protecting crops from pest attack and also it helps in replacing the chemicals which inturn save the environment.

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