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Prabhu S
Teaching Assistant, Department of Sericulture, Forest College and Research Institute, Tamil Nadu Agricultural University, Mettupalayam, Tamil Nadu, India

Priyadharshini Pachiappan
Assistant Professor, Department of Sericulture, Forest College and Research Institute, Tamil Nadu Agricultural University, Mettupalayam, Tamil Nadu, India

Thangamalar A
Teaching Assistant, Department of Sericulture, Forest College and Research Institute, Tamil Nadu Agricultural University, Mettupalayam, Tamil Nadu, India

Veeravel R
Professor, Division of Entomology, Annamalai University, Annamalai Nagar, Chidambaram, Tamil Nadu, India

Corresponding Author:
Prabhu S
Teaching Assistant, Department of Sericulture, Forest College and Research Institute, Tamil Nadu Agricultural University, Mettupalayam, Tamil Nadu, India

Effect of dust formulation of Milkweed (*Calotropis gigantea* R. Br.) plant parts against *Helicoverpa armigera* (Hubner)

Prabhu S, Priyadharshini Pachiappan, Thangamalar A and Veeravel R

Abstract
An experiment was conducted to study the effect of dust formulation of milkweed plant *(Calotropis gigantea* R. Br.) plant parts *Viz.*, leaf, flower, stem, root and whole plant against *Helicoverpa armigera* (Hubner) (Noctuidae : Lepidoptera). The insecticidal action of dust formulation of *C. gigantea* plant parts were applied on II, III instar stages of *H. armigera* larvae with concentrations of 2, 4, 6, 8 and 10 percent. Among the different plant parts, the leaf dust @ 10 percent proved its superiority over other plant parts dust by recording 86.66 percent of mortality followed by eight percent (60.00%), six percent (53.33%), four percent (43.33%) and two percent (33.33%) mortality respectively in second instar larvae of *H. armigera*. Leaf dust at 10 percent recorded maximum mortality rate of 83.33 percent whereas eight, six, four and two percent recorded mortality of 76.66, 46.66, 40.00 and 26.66 percent respectively in third instar larvae. Hence, the present study revealed that leaves from *C. gigantea* R. Br have insecticidal properties and could be an alternative candidate for chemical pesticides.

Keywords: *Helicoverpa armigera, Calotropis gigantea*, Dust formulation, Insecticidal activity, Mortality

Introduction
Milkweed plant or erukku (*Calotropis gigantea* R. Br.) belongs to the family Asclepediaceae, a common waste land weed widely distributed in tropical and subtropical region of Asia[1]. It is gaining more attention in recent years as a potential pesticidal source against insect pests, besides having several medicinal and industrial uses [2]. It is known to have insecticidal [3], antibacterial and antifungal properties [4]. The plant extract had proved effective against lepidopterous and sucking pests of several crops [5, 6]. Extracts of milkweed plant containing insect toxic principles such as glycosides or flavonoids are easily degradable when applied in a crop ecosystem and are effective against an array of insect pests [7].
American Bollworm, *Helicoverpa armigera* (Hubner) (Noctuidae: Lepidoptera) is one of the most destructive pests of various crops and more or less of universal occurrence except in regions where extremes of climate prevails. It has been reported that it feeds on more than 120 cultivated food plants all over the world. Host species for *H. armigera* include broad spectrum of families and important agricultural crops such as cotton, maize, chickpea, pigeon pea, sorghum, sunflower, soyabean and groundnuts. This pest alone accounts for losses of more than 50-80 percent in several crop [8]. Although, several conventional insecticides were found to be effective against this pest, *H. armigera* had developed resistance to commonly available insecticides posing great problem to farmers [9, 10] and pollute the environment. Management of *H. armigera* using insecticides has become now difficult due to the development of resistance. Therefore alternate measures of control that will be more efficient, more economical and safer needs to be developed. Botanical based insecticides have long been touted as attractive alternations to synthetic chemical insecticides for pest management.
Taking all these into considerations, the present investigations were under taken to obtain precise information on the effect of *C. gigantea* plant parts on developmental stages of *H. armigera*.

Materials and Methods
Mass culturing of test insect *H. armigera*
The eggs of *H. armigera* were collected from the field, transferred on a piece of cloth, surface sterilized with 0.1% sodium hypochlorite solution and kept on plastic trays.

The emerged neonates were transferred to the chickpea plant using a fine camel hairbrush. When the larvae were four to five days old (second instar), they were transferred to multicavity tray containing fresh bhendi fruit pieces to avoid cannibalism [11]. The larvae were allowed to remain in the multicavity tray until they reached the prepupal stage and then transferred to sand medium for pupation. The pupae obtained from culture were placed in petridishes inside adult emergence cage. Adults were fed with 10 percent sucrose solution with multi vitamin drops. Ten pairs of healthy adults were transferred to plastic buckets for mating and oviposition. The top of plastic bucket was covered with sterile blue coloured muslin cloth, which served as an oviposition substrate.

Preparation of Plant material

Fresh *Calotropis gigantea* R. Br plant parts *viz.*, leaves, stem, flower and roots were collected and shade dried to prevent the loss of active principle from the plant parts. The shade drying process was continued until the plant materials were dried enough to be powdered. The dried plant materials were taken individually and ground into powder with the help of Willey Mill. Whole plant powder was prepared by mixing leaf, flower, stem and root powder in equal proportions. The plant parts dust was stored in plastic containers for further use.

Preparation of Dust formulation

Dust formulations of *Calotropis gigantea* plant parts were prepared by dissolving the crude extracts in methanol made to 10 g stock solution. Different concentrations *viz.*, 2, 4, 6, 8 and 10 percent of insecticidal dust were formulated by adding the requisite quantity of 10 g stock solution to a known weight of the diluents talc (90 g) required for particular concentration. The contents were thoroughly stirred with help of glass rod so as to ensure complete impregnation of the diluent's particles with plant extract by placing the beaker in an electric oven where the temperature was maintained at 29 ± 1 °C to evaporate the solvent for 48 hrs when the dust was completely free from the solvent. Thereafter, the impregnated dust was thoroughly ground with the help of pestle and mortar [12].

Results and Discussion

Indiscriminate use of pesticides has led to the accumulation of toxic residues in food, soil, air, water and development of resistance in pests [13, 14]. Also, pesticides affect soil microbes, which are essential in governing soil quality and health. Promising results have been obtained using botanical compounds derived from plants for the control of agricultural pests. The compounds of botanical origin may be highly effective, with multiple mechanisms of action and minimum toxicity towards nontarget organisms.

Insecticidal action of *C. gigantea* plant parts dust against second instar larvae of *H. armigera*

The data presented in Table 1 exerted that percent mortality of second instar larvae of *H. armigera* was directly proportional to the concentration of plant part dust. The leaf dust proved its superiority over other plant parts dust by giving 86.66 percent of mortality at 10 percent concentration followed by eight percent (60.00%), six percent (53.33%), four percent (43.33%) and two percent (33.33%) mortality respectively. Flower dust treatment recorded the maximum mortality of 66.66 percent at 10 percent which was followed by eight, six, four and two percent recorded that 53.33, 43.33, 36.66 and 30.00, percent mortality respectively. Next to flower dust, the whole plant dust and stem dust evinced mortality at 10 percent concentration (63.33% and 43.33%), while six and four percent concentrations of whole plant extract were on par with each other recording 36.66 and 33.33 percent mortality compared to other dusts, root dust exerted low mortality percentage in two percent concentration. Prabhu *et al.* [15] studied the ovicidal activity of *Calotropis gigantea* plant parts on *Helicoverpa armigera*.

Bakavathippan, *et al.* [16] reported larvicidal activity of chloroform leaf extract of *Calotropis procera* against *Spodoptera litura*. Begum *et al.* [17] examined the insecticidal activity of *Calotropis procera* against various stages of *Musca domestica*. The efficacy of aqueous extract of leaf, flower and roots of *C. procera* proved most effective in the control of *Henosepilachna elaterii* [18].

Table 1: Effect of dust formulation of *C. gigantea* plant parts against second instar larvae of *H. armigera*

Treatments	Mortality in Percentage*				
	Leaf	Flower	Stem	Root	Whole plant
2%	33.33 (35.21)^{e}	30.00 (33.21)^{e}	13.33 (21.14)^{d}	10.00 (18.43)^{e}	26.66 (30.99)^{d}
4%	43.33 (41.15)^{d}	36.66 (37.22)^{d}	20.00 (26.56)^{c}	16.66 (23.85)^{d}	33.33 (35.21)^{c}
6%	53.33 (46.92)^{c}	43.33 (41.15)^{c}	23.33 (28.78)^{c}	20.00 (26.56)^{c}	36.66 (37.22)^{c}
8%	60.00 (50.76)^{b}	53.33 (46.92)^{b}	30.00 (33.21)^{b}	26.66 (30.99)^{b}	50.00 (45.00)^{b}
10%	86.66 (68.85)^{a}	66.66 (54.78)^{a}	43.33 (41.15)^{a}	36.66 (37.22)^{a}	63.33 (52.77)^{a}
Control	0.0 (0.0)^{f}	0.0 (0.0)^{f}	0.0 (0.0)^{e}	0.0 (0.0)^{f}	0.0 (0.0)^{e}
SED	2.64	1.95	2.30	1.81	2.46
CD (p = 0.05)	5.89	4.36	5.13	4.05	5.49

*Values are mean of three replications

Values in parentheses are arc-sine transformed values

Mean values with various alphabets differ significantly by DMRT.

The dust treatment of all the five plant parts of *C. gigantea* caused considerable larval mortality of *H. armigera* but the mortality was more when leaf dust at 10 percent concentration was used on second instar larvae showed maximum of 86.66 percent mortality. Next to leaf dust, flower dust showed the maximum mortality of 66.66 percent whereas whole plant dust at 10 percent recorded mortality of 63.33 percent on second instar larvae. The stem and root dust were less effective and recorded mortality percentage of 43.33 and 36.66 percent at 10 percent concentration. Dodia *et al.* [19] reported that *Calotropis procera* and *Datura stromonium* showed 90 percent feeding protection against *H. armigera.* Jitendra Singh *et al.* [20] evaluated insecticidal activities of *Calotropis procera* against *Pieris brassicae* and *Helicoverpa armigera* which showed larval mortality of 18-50 percent.

Insecticidal action of *C. gigantea* plant parts dust against third instar larvae of *H. armigera*

Leaf dust at 10 percent recorded maximum mortality rate of 83.33 percent whereas eight, six, four and two percent recorded mortality of 76.66, 46.66, 40.00 and 26.66 percent respectively (Table 2). Next to leaf extract, flower and whole plant dust at 10 percent concentration recorded mortality of 76.66 and 73.33 per cent. Stem dust showed the mortality of 56.66% at 10 percent. Root dust was found to be least effective. As the dust concentration of plant parts increased, proportional larval mortality percentage was observed. The present study confirmed the efficacy of dust formulation of calotropis and larval mortality increased with increasing concentration. Deepali Lall *et al.* [21] studied the larvicidal effect of *Calotropis procera* leaf extracts against *Helicoverpa armigera* and proved that increased concentration increased mortality. The pesticidal effects of *Calotropis gigantea* latex on *Helicoverpa armigera* were studied by Singh *et al.* [22] and recorded mortality of 37.02%.

Table 2: Effect of dust formulation of *C. gigantea* plant parts against third instar larvae of *H. armigera*

Treatments	Mortality in Percentage*				
	Leaf	Flower	Stem	Root	Whole plant
2%	26.66 (30.99)^{e}	23.33 (28.78)^{d}	10.00 (18.43)^{e}	6.66 (13.64)^{e}	16.66 (23.85)^{e}
4%	40.00 (39.23)^{d}	33.33 (35.21)^{c}	16.66 (23.85)^{d}	13.33 (21.14)^{d}	26.66 (30.99)^{d}
6%	46.66 (43.07)^{c}	36.66 (37.22)^{c}	23.33 (28.78)^{c}	20.00 (26.56)^{c}	33.33 (39.14)^{c}
8%	76.66 (61.21)^{b}	66.66 (54.78)^{b}	36.66 (37.22)^{b}	33.33 (35.21)^{b}	60.00 (50.76)^{b}
10%	83.33 (66.14)^{a}	76.66 (61.21)^{a}	56.66 (48.84)^{a}	50.00 (45.00)^{a}	73.33 (59.00)^{a}
Control	0.0 (0.0)^{f}	0.0 (0.0)^{e}	0.0 (0.0)^{f}	0.0 (0.0)^{f}	0.0 (0.0)^{f}
SED	2.41	2.77	2.76	3.03	3.02
CD (p = 0.05)	5.38	6.17	6.16	6.76	6.74

*Values are mean of three replications
Values in parentheses are arc-sine transformed values
Mean values with various alphabets differ significantly by DMRT.

The presence of phytochemicals *viz.,* Sapogenins, holarrhetine; cyanidin-3-rhamnoglucoside, taraxasterol isovalerate. mudarine and three glycosides calotropin uscharin, calotoxin along with phenol [23] in the milkweed plant extracts shall be the most important factors for causing mortality of *Helicoverpa armigera* larvae. Alam *et al.* [24]. proved the insecticidal activity, residual film toxicity, fumigant toxicity and repellent effect of methanol extract of *Calotropis gigantea* against *Tribolium castaneum*. The present study also confirmed the insecticidal activity of *Calotropis gigantea* against *Helicoverpa armigera*.

Conclusion

Dust formulation of milkweed plant *(Calotropis gigantea* R. Br.) parts *Viz.,* leaf, flower, stem, root and whole plant showed insecticidal activity against *Helicoverpa armigera* (Hubner). Botanical derived insecticides are easily biodegradable, low toxicity, inexpensive and keeps the environment clean and safe to non target organisms including humans. From the present study, it is concluded that *Calotropis gigantea* could be an alternative to synthetic chemical pesticides for management of pests.

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