

Antifeedant Activity of Leaf Aqueous Extracts of Selected Medicinal Plants on VI instar larva of *Helicoverpa armigera* (Hübner)

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ABSTRACT

Phytochemicals, especially botanical insecticides are currently of interest because of their successful application in plant protection as biocontrol agents. Biological activity of leaf aqueous extract of ten selected medicinal plants were evaluated against the fourth-instar larvae of gram pod borer *Helicoverpa armigera* (Hübner), (Lepidoptera: Noctuidae). Antifeedant activity of leaf aqueous extracts of *Abutilon indicum* L., *Achyranthus aspera* L., *Aerva lanata* L., *Albizia amara* (Roxb), *Andrographis paniculata* Ness., *Cardiospermum halicacabum* L., *Cassia tora* L., *Catharanthus roseus* L (G) Don., *Datura metal* L. and *Tribulus terrestris* L. were evaluated in this study. Preliminary screening after 24 h of exposure with leaf aqueous extracts of the selected plants at a concentration of 1,000 ppm exhibited significant larval mortality rate. The percentage mortality rate ranged considerably from 10.8 to 72.8. The mortality rate was observed in the decreasing order of *A. paniculata* > *C. roseus* > *D. metal* > *A. amara* > *C. halicacabum* > *A. indicum* > *C. tora* > *T. terrestris* > *A. aspera* > *A. lanata* against the larvae of *H. armigera*. The results imply that leaf aqueous extract of *A. paniculata*, *C. roseus* and *D. metal* can potentially be used as eco-friendly pest control agents against the larva of *H. armigera*.

INTRODUCTION

India is basically an agro-based country more than 80% of Indian population depends on agriculture. Indian economy is largely determined by agricultural productivity. Insect-pests are known to cause significant damage to crops and affect agricultural productivity. The monetary loss due to feeding by larvae and adult insects alone contributes to billion dollars per annum (Jacobson, 1982). Among the Lepidopteran, Pod borer - *Helicoverpa armigera* is the key pest that cause severe damage to crop plants and bring about significant yield loss. *Helicoverpa armigera*, commonly known as cotton bollworm or American bollworm, is a major polyphagous noctuid pest in Asia, causing heavy damage to agricultural, horticultural and ornamental crops (Talekar *et al.*, 2006). *H. armigera* infests several plants like cereals, pulses, cotton, and vegetable besides it has been reported to attack wild hosts. In central and north India, it is the major pest affecting cotton. The larvae feed extensively on cotton plant parts including the newly emerging bolls causing severe loss of crop. Bollworms are relatively safe from natural enemies because of the cryptic feeding habits of the larvae within cotton bolls. Therefore, large numbers of *H. armigera* in cotton and other vegetables survive to adults that may disperse widely, producing progeny that damage high-value

crops (Cabanillas and Raulston 1995; Michael and Donald 1996). Economic loss due to this pest in India accounts for 5,000 cores (Manjunath *et al.*, 1985). *Helicoverpa armigera* has a long history of resistance to conventional insecticides. Variety of chemical insecticides and pesticides are used to control *H. armigera*. However, harmful effects and persistent nature of the chemical pesticides demand for eco-friendly alternatives.

During the last 50 years, worldwide use of synthetic insecticides to control insect pests has led to both insecticide resistance and environmental persistence (Roush, 1990). Plant derived phytochemicals have been widely used in the management of agricultural pest since time immemorial (Choudary, 2001). Plant derived pesticides are eco-friendly, non-toxic to non target organisms, non persistent in nature, besides they do not promote drug resistance (Liu *et al.*, 2000). Therefore, researchers world over are engaged in a mission to hunt for novel phytochemicals that could potentially be used in the management of insect-pests.

Plants are endowed with a potential to produce a range of secondary metabolites like alkaloids, terpenoids, flavonoids, phenols, glycosides, sitosterols and tannins. These phytochemicals are known to protect the plants from the attack of insect-pests (Ahmad, 2007). 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. Application of bio-pesticides has been reported to have positive impacts on bollworm population management (Ge and Ding 1996).

A number of plants have been shown to have pesticidal and antifeedant activity against *H. armigera* of which Neem has been subjected to extensive investigation by Chopra *et al.*, (1994). Studies have shown that *Acorus calamus*, *Annona squamosa*, *Vitex negundo* are effective in the management of *H. armigera* (Murugan *et al.*, 1998). Sundararajan and Kumuthakalavalli, (2001) evaluated antifeedant activity of aqueous extract of *Gnidia glauca* and *Toddalia asiatica* against *H. armigera*. In this view, effect of leaf aqueous extracts of selected medicinal plants viz., *A. indicum*, *A. aspera*, *A. lanata*, *A. amara*, *A. paniculata*, *C. halicacabum*, *C. tora*, *C. roseus*, *D. metal* and *T. terrestris* have been evaluated for their efficacy in the management of the insect pest *H. armigera*.

MATERIALS AND METHODS

Preparation of extract

Selection of plants used in the present study was made on the basis of their availability and absence of damage by the insect-pest. Healthy plant materials were collected from the wild in poly bags and brought to lab and their botanical identity was established. Fresh leaves from the selected plants were collected and washed separately. About 1gm of fresh leaf material was ground with distilled water using mortar and pestle. The extract was filtered and the filtrate was made upto 100ml and was maintained as stock. The leaf aqueous extract was diluted with distilled water to obtain 1% test solution used in the bioassay studies.

Test organism

The larvae used for the study were collected from the host plants in the cotton fields and brought to lab. They were reared on artificial diet under laboratory conditions. Studies were carried out using VI instar larvae of *H. armigera* against the aqueous extract of all the selected plant species. The percentage mortality was calculated after a period of 24h by using bioassay studies.

Bioassay studies

Studies were conducted for a period of 24h in the laboratory in transparent plastic containers of 4x2.5 cm size capped with perforated plastic lids. Fresh leaves of *Lycopersicum esculentum* (tomato) were collected from the field

and washed in water. Excess moisture was removed from the leaves and the leaves were dipped in 1% test solution, shade dried and served to the VI instar larvae of *H. armigera*. Extract free leaves served as control. For each treatment 10 larvae were singly introduced in separate containers after six hour starvation. Three replicates each of ten larvae were maintained for each treatment. All the experiments were conducted at 27 ± 1 °C, 75% humidity and 14h dark period. Twenty four hour larval mortality was observed and the percentage mortalities were corrected using Abbott's formula (Abbott, 1925).

RESULTS

The aqueous extracts of selected plant species collected from Salem and Vellore Districts of Tamil Nadu were subjected to bioassay studies against the VI instar larva of *H. armigera* (24h) and the percentage mortality rate was observed. The results are given in Table 1. It was observed that all the selected plant species exhibited antifeedant activity against the larvae of *H. armigera*. However, the percentage mortality rate varied significantly among the plants tested. The percentage mortality rate of the insects ranged from 10.8 to 72.8. The mortality rate was observed in the decreasing order of *A. paniculata* > *C. roseus* > *D. metal* > *A. amara* > *C. halicacabum* > *A. indicum* > *C. tora* > *T. terrestris* > *A. aspera* > *A. lanata* against the larvae of *H. armigera*. The leaf aqueous extracts of three plants namely *A. paniculata*, *C. roseus* and *D. metal* exhibited high rate of mortality (72.8, 67.8 and 62.2 percent respectively) against the larvae of *H. armigera*.

DISCUSSION

Prohibitive expense to meet the challenges of increasing resistance in insects, resurgence of pests and escalating environmental pollution caused by synthetic pesticides call for the discovery of less-expensive, non-hazardous alternatives in the management of insect-pests. Plants are endowed with a potential to produce a wide spectrum of allelo-chemicals (Norduland and Sauls, 1981). Insects have been influential in the evolution of allelo-chemicals in plants which in turn affects the insects. Some of compounds affect the feeding behavior of the insects and inhibit feeding, while few others disrupt hormonal balance there by inhibits growth, metamorphosis and reproduction. Due to aforesaid reasons there is resurgence of interest in plant derived compounds for developing them novel eco-friendly insecticides on commercial scale (Jacobson and Crosby, 1971). Despite hundreds of plants have insecticidal property, only few compounds like Azadirachtin (known to disrupt the action of moulting hormone) and pyrethroids (brings about paralysis of the insects) have been commercialized. However, an understanding of structure-activity relationship and knowledge on the mode of action is required for large-scale production. For successful exploitation of a bio-insecticide, screening of phytochemicals for wide spectrum of behavioral and physiological activities in poly-phagous insects is essential.

Koul *et al.*, (2000) administered phytochemical extracts orally through food to determine the toxicity or efficacy of plant materials for antifeedancy, inhibition of growth or emergence as adults. Murugan *et al* (1998), followed no-choice method in which the test insect was provided with treated leaf disc. Dual choice method mimics the situation in the field, and is valuable in assessing the antifeedancy of phytochemicals. However, this method does not facilitate administration of apt quantity of plant extract to the insects. On the other hand, no-choice bioassay method provides an opportunity to precisely administer the desired quantity of extract to the insect. Hence in the present study, no-choice bioassay method was followed for assessing the insecticidal activity of the different plant extracts. Usually larger doses of plant extracts inflict mortality either by inhibiting feeding or reducing digestibility or inhibiting growth. Smaller doses of extracts may not be adequate for killing the insects however it may sometimes induce malformation (Ahmad, 2007). Induction of morphogenetic deformities during larval development or

metamorphosis has greater impact on population build up. Malformed adults are unable to participate in reproductive activities and hence do not help building up the population. High antifeedancy (low ED₅₀) has been reported for pure compounds isolated from different plants by Simmonds *et al.*, (1990). Aqueous extracts of *Calotropis procera* and *Datura stromonium* display about 90% feeding protection against *H. armigera* (Dodia *et al.*, 1998).

Likewise, it has been reported that the effectiveness of the phytochemicals depend on the extraction method used. Janarthan *et al.*, (1999) reported that petroleum ether extracts of *Parthenium hysterophorus* at a concentration of 0.2 and 0.5% cause 100% mortality in the larvae of *H. armigera*. Antifeedant property of plant extracts brings about retardation of growth and ultimately results in death of the insect. However, compounds which do not display antifeedant property are reported to have growth regulatory activity (Kraus *et al.*, 1987). On the other hand, a few plant extracts display bimodal activity. At high concentrations they are as feeding deterrents and at low concentrations as growth inhibitors (Nawrot *et al.*, 1991). Jermy (1990) reported that extracts with antifeedant and toxic effect are more successful in practical application as they evoke behavioral effect of antifeedancy. Jaglan *et al* (1997) evaluated the effect of *Azadirachta indica* extracts against *H. armigera* and reported that Chloroform : methanol (9:1) extracts of Neem seed kernels and leaves showed better insecticidal properties than methanol extracts. On the other hand Koul *et al.*, (2000) reported that *H. armigera* larva fed on azadirachtin treated leaf suffered dose dependant effect on growth. Young larvae fed on 4 ppm azadirachtin treated leaf initially gained weight however, older larvae fed on 4 ppm azadirachtin leaf suffered 75% decrease in growth compared with the control larva (Koul, 1985). Similar observations were made by Murugan and Babu (1998) on growth and feeding physiology of *H. armigera* larva on extracts of *Ricinus communis*, *Glycosmis pentaphylla*, *Vitex negundo* and *Nerium oleander*. Murugan *et al.*, (1998) reported that larvae fed on 0.3% ethylacetate fraction of *Glycosmis pentaphylla*, *Vitex negundo* and *Nerium oleander* showed significant effect on initial weight (259, 181 and 177%) in 48h compared to weight gained by the control larva (898, 972 and 890%). However, earlier it has been reported that the aqueous leaf extracts of *Gnidia glauca* showed more than 50% larval mortality at 0.8-1.0% and 86.1% mortality observed at 1.0% on *Toddalia asiatica* extract against the sixth-instar larvae of *H. armigera* (Sundararajan and Kumuthakalavalli, 2001). In the present study all the tested plant extracts exhibited antifeedant activity on *H. armigera*. Of the 10 species of the plants screened for insecticidal property, three plant species (*A. paniculata*, *C. roseus* and *D. metal*) showed high rate of mortality, i.e. above 60%, on application of aqueous extracts against the VI instar larva of *H. armigera*. In conclusion, the study reveals that leaf aqueous extracts of *A. paniculata*, *C. roseus* and *D. metal* can potentially be used as eco friendly bio-pesticide to control the devastating damage caused by VI instar larva of *H. armigera*.

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Table 1 Effect of leaf aqueous solutions on the larvae of *H. armigera*.

S. No	Botanical name	Family	Mortality Rate (%)
1.	<i>Aerva lanata</i> L.	Amaranthaceae	10.8
2.	<i>Achyranthus aspera</i> L.	Amaranthaceae	16.8
3.	<i>Tribulus terrestris</i> L.	Zygophyllaceae	25.2
4.	<i>Cassia tora</i> L.	Caesalpiniaceae	31.8
5.	<i>Abutilon indicum</i> L.	Malvaceae	35.2
6.	<i>Cardiospermum halicacabum</i> L.	Sapindaceae	42.8
7.	<i>Albizia amara</i> (Roxb).	Minosaceae	51.8
8.	<i>Datura metal</i> L.	Solanaceae	62.2
9.	<i>Catharanthus roseus</i> L (G) Don.	Apocanaceae	67.8
10.	<i>Andrographis paniculata</i> Ness.	Acanthaceae	72.8