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Larvicidal Activity of Solvent Leaf Extracts of *Cassia fistula* (Linn) and *Clerodendron inerme* (Gaertn) on the *Spodoptera litura* (Insecta: Noctuidae): A Potential Botanical Alternative

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Abstract

Spodoptera litura is a polyphagous pest affecting several agricultural crops. Chemical insecticides have been used in the past for controlling this pest. This has resulted in the development of resistance to several classes of pesticides. Bioactive molecules of plant origin hold potential alternative for the chemical pesticides. In the present study leaves of *Clerodendron inerme* and *Cassia fistula* were extracted with methanol and ethanol. The efficacy of the extracts as insecticidal agents were assessed on third instar larvae of *Spodoptera litura*. The results suggest that ethanol extracts of *Cassia fistula* (LD_{50} = 1.70389) and methanol extracts of *Clerodendron inerme* (LD_{50} = 3.84670) shows a higher toxicity on third instar larvae of *Spodoptera litura*. The present study shows that the solvent extracts from the above plants can be used for the control of armyworm.

Key Words: Armyworm, Golden shower, Phytochemical, Kashmir Bouquet

Introduction

Spodoptera litura is a polyphagous pest damaging several crops in Southeast Asia [1, 2, 3]. Use of chemical insecticides to control *Spodoptera litura* has proved futile as it has developed resistance to several classes of insecticides [4, 5]. The adverse impact of chemical pesticides on non-target organisms including humans and long term environmental persistence has necessiciated the search for alternative methods for pest management [6] using botanicals [7].This can provide an environment friendly way of management of insect pests without contaminating the soil and protecting the harmful effects on non-target organisms [8].

The plant kingdom is by far the most efficient producer of chemical compounds, synthesizing secondary metabolites that are used in defense against herbivores. The essential constituents of secondary metabolites are tannins, alkaloids, polyphenols, terpenoids, and essential oils which have wide range of anti-insect properties[9] which include insecticidal activity, repellence, antifeedant effects, insect growth regulation [10, 11, 12].

Clerodendron inerme Gaertn. (Family: verbinaceae), commonly known as Kashmir bouquet is a biennial, hardy plant and widely grown as a hedge plant along home gardens. The leaf extracts of the plant has been shown to have larvicidal and pupicidal activities in mosquitoes [13, 14]. *Cassia fistula* L (family: leguminosae) is a semiwild India tree (known as Golden Shower) having wide ranging distribution in Asia, parts of Africa and South America. It has been used by tribal people to treat ring worm and skin infections [15]. It is also used by tribal people in the treatment of fever, diarrhoea, leprosy and abdominal pain [16].

The leaf extracts of the plant has been known to have larvicidal and ovicidal activity in mosquitoes [17]. In the present study, Methanol and ethanol leaf extracts of *Cassia fistula* and *Clerodendron inerme* were tested for larvicidal activity in third instar larvae of *Spodoptera litura* (family: noctuidae).

Materials and Methods

Plant Material

Fresh leaves of *Cassia fistula* L and *Clerodendron inerme* Gaertn were collected from the field. The leaves were air dried in the laboratory for 14 days and milled in a mixer blender to obtain fine powder. The leaf powder was passed through a fine sieve to remove the larger leaf parts. Fine leaf powders were used as raw material for extraction in methanol and ethanol solvents.

Insects

Spodoptera litura were obtained from the laboratory stock culture from the insectary. The larvae were reared on artificial diet. The entire insect culture was maintained in the insectary at 25ºC, 70% RH and 16:8 (L: D) photoperiod.

Soxhlet Extraction

Powdered leaf of *cassia fistula and clerodendron inerme* was weighed and was wrapped in Whatmann No. 1 filter paper. 40 grams of the enveloped leaf powder was introduced into the soxhlet chamber at one time for the extraction. In the round bottom flask 350 ml of methanol was taken as solvent. The apparatus was run till the solvent in the soxhlet chamber became transparent Then the extract along with the solvent was removed and solvent mixture was subjected to vacuum drying and the paste was stored at 4°C till the bioassay were

performed. Paste was treated as the pure extract and for bioassay different concentrations of the leaf extract was prepared. Similar procedure was repeated with ethanol as solvent. The concentration of 0.01% w/v, 0.25% w/v, 0.05% w/v, 0.1% w/v and 0.15% w/v were used for both ethanol and methanol extracts.

Bioassay

Third instar larvae were collected from the rearing stock and were kept in ventilated plastic containers (15cm diameter and 5 cm in height) for the bioassay. Three replicates per treatment (concentration) per solvent extract and controls were kept. Each replication consisted of ten third instar larvae.

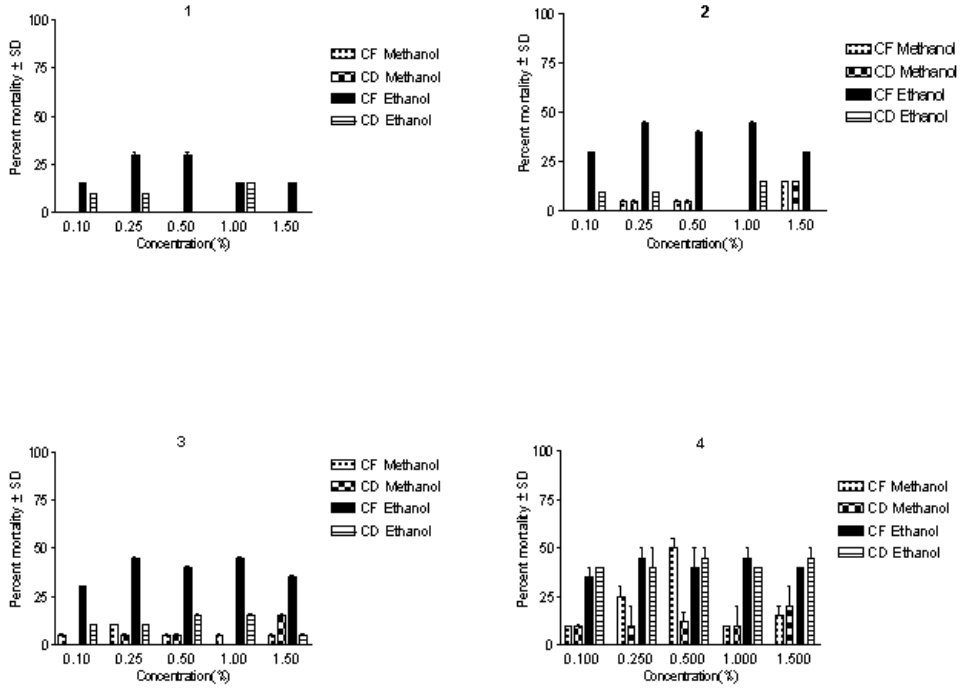
Topical application: 2µl of the solvent extract was applied topically on the thoracic dorsum of each insect with the help of a micropipette (Tarsons, T10). After the treatment the larvae were released in the container containing cabbage leaves. Controls were treated with the solvent. Insect mortality on larvae was assessed after 1, 3, 7, 14, and 21 days of treatment. The biological efficacy of the extracts on *Spodoptera* larvae were expressed using Abbott's correction [18].

Results

Solvent extracts of both methanol and ethanol of both the plants showed bioactivity on *Spodoptera* larvae. Among the extracts, ethanol extract of *Cassia fistula* (CF) (LD_{50}: 1.70389% w/v) and methanol extracts of *Clerodendron inerme* (CI) (LD_{50}: 3.8467% w/v) was highly toxic (Figures 1-4; Table 1). CD extracts show early toxic effects in both *Spodoptera litura* (Figure1-4).

Table 1. Mortality of *Spodoptera litura* after topical application with methanol and ethanol extracts from the leaves of *Cassia fistula* and *Clerodendron inerme*. Ethanol extracts of *Cassia fistula* and *Clerodendron inerme*, LD_{50} estimates and subsequent chi square tests (df=3) is highly effective as compared to the other extracts.

Solvent Extract	LD_{50} (%W/V)	Lower limit	Upper limit	Slope	Intercept	Chi^2
CF Methanol	13.747	0.203	927.589	0.5588	-0.63604	0.25515
CF Ethanol	1.704	0.044	65.378	0.2635	-0.06099	0.21518
CD Methanol	3.847	1.639	378.454	1.2684	-0.74213	0.98979
CD Ethanol	18.663	0.000	3407.849	0.1250	-0.15897	0.07186



Figures (1-4) represent the percent mortality rates (± SD) at 3,7,14 and 21 days after treatment (DAT) using various concentrations of ethanol and methanol leaf extracts of *Cassia fistula* and *Clerodendron inerme* respectively

Discussion

Noctuid moths from genera *Spodoptera* are polyphagous pests causing economic damage in several agricultural crops throughout the world [3]. Broad spectrum insecticides have been used for its control has resulted in development of resistance to many of the registered pesticides for its control [19, 4, 20]. In this scenario newer types of insecticides originating from natural products, targeting *Spodoptera litura* could be useful alternative for integrated pest management [21].

In the present study all the plant extracts showed moderate toxicity to *Spodoptera litura* larvae, while the ethanol extracts of *Cassia fistula* was highly toxic (LD_{50} = 1.704 % w/v), while the methanol extract of *Clerodendron inermis* showed slightly lesser toxicity to *Spodoptera litura* larvae (LD_{50} = 3.847 % w/v). similar studies using extracts from *Ocimum canum*, *Ocimum sanctum* and *Rhinacanthus nasutus* larvicidal activity on fourth instar larvae of *Spodoptera litura* and after 24 hours obtained methanol extracts of *O.canum and R.nasutus* showed higher toxicity (LC_{50} = 36.46 and 68.84 ppm respectively) while acetone extracts of produced toxicity (LC_{50}= 68.08 ppm) while acetone extracts of produced toxicity (LC_{50}= 68.08 ppm) [22]. [23] analysed the larvicidal activity of several plant extracts and found that ethanol extracts of *Azadirachta indica* and *Melia azedarach* produced toxicity of 22.2% and 25% respectively. [24] evaluated ethyl acetate and hexane extracts of fruit pulp of *Momordica dioica*, results indicate that both the extracts showed a concentration dependent antifeedant effect on *Spodoptera litura* larvae. Growth reduction and toxic effects of *Synedrella nodiflora* Gaertn (Asteraceae) extracts on *S. litura* was reported by [25]. [26] reported that hexane extracts of *Porteresia coarctata* produced more than 50% mortality after 24 hours of treatment on *Spodoptera litura* larvae.

Results of our study indicates that the plant extracts of *Cassia fistula* and *Clerodendron inerme* are higher larvicidal efficacy as compared several other plants. Further characterization and isolation of bioactive molecules from ethanol extracts of cassia fistula and methanol extracts of *Clerodendron inermis* will provide further clarity about the nature of these bioactive compounds. This could become alternative to the conventional insecticides used for the regulation of *Spodoptera litura*.

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