# Ovicidal activity of crude extracts of few traditional plants against *Helicoverpa armigera* (Hubner) (Noctuidae: Lepidoptera)

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**ABSTRACT**

*Cipadessa baccifera* Miq., *Melia dubia* (Cav.) (Meliaceae); *Clausena dentata* (Rutaceae) and *Dodonaea angustifolia* (Sapindaceae) are common medicinal plants found in Western Ghats and are used traditionally for various purposes. The petroleum ether, chloroform, hexane, acetone and water extracts of the leaves were investigated for their ovicidal property against *Helicoverpa armigera* (Hubner) (Lepidoptera : Noctuidae). The different extracts of the test plants differed significantly in their efficacy. Among the four plants tested for ovicidal activity, *Clausena dentata* reduced the egg hatchability and proved to be highly ovicidal compared to others. Among the age of eggs, it was clear that the early stage of the eggs namely 24 h old eggs, to be highly susceptible to all the treatments.

**Key words :** *Helicoverpa armigera*, plant extracts, ovicidal activity

## INTRODUCTION

The injudicious use of chemical fertilizers and pesticides has resulted in multiple problems such as increase in the insect resistance to insecticides, emergence of new pests, minor pests becoming major pests and pesticide pollution to the environment (Kannaiyan, 1999). To add to the serious side effects, large number of suicides among cotton farmers in the Warangal district of Andhra Pradesh has raised more serious concern (Parthasarathy Shameem, 1998). In addition to the direct economic loss caused by the insect pests (5-15%) (Banerjee *et al.,* 2000) indirect damage such as transmitting plant diseases is also observed.

Among the different pests, *Helicoverpa armigera* Hubner (Lepidoptera: Noctuidae) is considered to be important, since they are capable of wiping out the entire crop (Pawar, 1998). An annual loss of about Rs. 2,000 crores in India by *Helicoverpa armigera* (Ignacimuthu and Jayaraj, 2003) is observed. *H. armigera* is a polyphagous migratory noctuid attacking 96 cultivated and 61 uncultivated plants. The major susceptibles include cotton, pulses, oilseeds, millets and vegetables (Reed and Pawar, 1982). Insecticides valued at US $ 660 million are used annually on all crops in India and more than half of it is used on cotton (Manjunath, 2004). Cost of the 21,500 metric tonnes (active ingredient) of insecticides used on cotton in India in 2001 was US $ 340 million. Further, the most destructive pest, *Helicoverpa armigera* is known to have developed resistance against most of the recommended insecticides (Kranthi *et al.,* 2001; Ramasubramanyam, 2004). However, to promote ecological balance and to minimize the insect resistance to insecticides, it is necessary to shift to integrated management of pests (Reddy and Manjunath, 1999; Ravi *et al*., 2008).

Interest in the application of natural products in Integrated Pest Management (IPM) remains high. Certain plants by nature possess secondary metabolites, which act as antifeedants, oviposition deterrents, larvicidal and insect growth regulators (Vasantharaj David, 2008) as well as effective against plant pathogens (Harborne, 1988). Moreover, botanicals are preferred over other methods since they are easily available, biodegradable and least toxic to non-target organisms (Wink and Guo, 1995).

The antifeedant and growth-inhibitory activities of *Melia dubia* methanol extract and the allelochemical toosendanin isolated from this fraction to *Helicoverpa armigera* revealed reduction in relative growth and consumption rates after oral administration of toosendanin, with a concomitant reduction in efficiency of conversion of ingested food (ECI) at higher concentration only (Opender Koul *et al*., 2002). Extracts of *Ocimum basilicum, Gynandropsis gynandra, Acorus calamus, Lantana camara* caused significant mortality in *H. armigera* (Pandey *et al*., 1983), while 0.1% neem seed kernel caused larval-pupal intermediaries and abnormal adults (Jotwani and Srivastava, 1984). Neem seed kernel extract along with pepper fruits caused deterrent/antifeedant activity against *H. armigera* (Hongo and Karel, 1986). The same when applied with neem oil and *Pongamia glabra* oil (3 and 5%) on chickpea, it reduced the pod damage by the insect

(Kumar and Sangappa, 1984). The combined application with fenvalerate resulted in less damage to pigeon pea (Kotikal, 1998). Sahayaraj *et al.* (2006) reported the ovicidal activity of *Pedalium murex* Linn. on *Dysdercus cingulatus* (Fab.).

Suresh *et al.* (2002) reported that higher concentration of *Hyptis suaveolens* and neem reduced the egg laying capacity of *S. litura*. Maximum oviposition deterrent activity was recorded in *Acorus calamus* leaf extract with hexane followed by *Wedelia calendulacea*, *A. malaba ricus* and also in *A. calamus* root. Generally, *W. calendu lacea* in all the concentrations showed significant deter rent activity. Oviposition deterrence may be due to the presence of deterrence compounds present in the plants dissolved in various solvent extracts that may repel the adult moth to lay egg on treated leaves (Raja *et al*., 2003). Naumann and Isman (1995) reported that 1 % crude oil emulsion significantly reduced the proportion of eggs laid by S. *litura* on treated plants. Significant increases in larval mortality, antifeedant and ovipositional repellency were found in radish terminal leaves treated with *Azadirachta indica* (Kumar *et al*., 1997). *Azadirachta indica* exhibited maximum repellency with 41.01 per cent reduction in egg laying in *Plutella xylostella* (Roopa Patil and Basavana Goud, 2003). There was no oviposition in an area treated with the methanol extract of neem at 0.01% for 5 days (Ayyangar and Rao, 1989). Dichloromethane and methanol extract of *A. calamus* reduced oviposition reduction from 86.5 to 65.0% at higher concentration (Jayakumar *et al*., 2005).

*Cipadessa baccifera* Miq. (Meliaceae) is a bushy shrub, distributed in North Circas, Deccan and Western Ghats. It has multiple uses such as fuel, fodder and fish poison. In addition, it is used against piles problem, diabetes, diarrhoea and headache. The active constituents isolated from the seeds of *C. baccifera* include cipadesin, 17a, 20R-dihydroxypregnan-3, 16-dione, 1, 4-epoxy-16-hydroxyheneicos- 1, 3, 12, 14, 18-pentaene etc (Luo *et al.*, 2000). *Clausena dentata* (Rutaceae) is a small tree distributed in North Circas, Hills of Ganjam and Vizagapatam to 5,000 ft; Western Ghats in Wayanad, Malabar, Anamalais, Pulneys and Travancore at 3,000 to 5,000 ft. It is mainly used against kidney pains (Armando, 2003). â-sitosterol (Prakash *et al.*, 1980), amides (Mingh *et al.*, 1988) and terpenoids (Ito *et al.*, 2000) have been reported from the leaves. Estragole, leaf oil of *C. anisata* was toxic to third nymphal instar of the grasshopper *Zonocerus variegata* (Okunade and Olaifa, 1987).

*D. angustifolia*, commonly known as hop bush (Sapin daceae) is a perennial shrub dominates at Pachaimalai foot hills 250 m, Kolli hills (200- 1300 m), Servarayans, Yercaud (1400 m), Ulundurpettai (75 m), (sub coastal), Chitteris, Oomathi (1000 m) and at Western Ghats, in Shola forests (up to 2000 m) (Gamble, 1987). It has a wide application from ancient time till date in modern medicines all over the world. Traditionally the farmers in various places use these plants widely as fencing material, broom, thatching the roof, firewood, apart from the medicinal use such as cuts, open wounds, snake bite and rheumatism (Subashini *et al*., 2004). The plant possesses biochemical compounds such as essential oil, fatty oils, flavonoids, terpenoids, resin, phenols, coumarins, sterols and unidentified alcohols (Sachdev and Kulshreshtha, 1986; Van *et al.*, 2000; Abdelmogib *et al.*, 2001). Ethanolic extract of the leaves of *D. viscosa* showed antibacterial activities against *Micrococcus, Bacillus, Salmonella* species and *Coryne bacterium diphtheriae, Sarcina lutea* and *Escherichia coli* (Sukkawala and Desai, 1962). Hexane extract of *D. viscosa* resulted in reductions in fecundity and hatcha bility, highest larval mortality and reduction in pupation against *H. armigera* (Subashini *et al*., 2004) and chloro form and aqueous extracts against *Spodoptera littoralis* in the laboratory (Abdelaziz and Omer, 1995).

*Melia dubia* (Cav.) (Meliaceae) is a large deciduous tree/ shrub distributed in N. Circas, Ganjam and Nallamalai Hills and S. Canara to Tinnevelly at low elevations. Its timber is mainly used for furniture and agricultural implements while its leaves are highly nutritious and used for fodder (Amarasekara, 1995). Monoterpene camphene (21.68%), alpha- and beta-pinene (3.12% and 5.13%), sabinene (2.75%) were the major constituents of leaf essential oil of *M. dubia* (Nagalakshmi *et al.*, 2001). Two new tetranor terpenoids, compositin and compositolide have been isolated from leaves and seeds of *M. dubia* (Porusho thaman *et al.*, 1984). Dichloroethane and methanol extracts of *M. dubia* were more toxic to *Spodoptera litura* and *Helicoverpa armigera* larvae 72h after topical application (Opender *et al.*, 2000). The environmental hazards posed by synthetic pesticides provide an impetus for investigations into some ecofriendly and biorational alter natives. The critical literature search indicates that *C. bac cifera*, *C. dentata*, *D. angustifolia* and *M. dubia* have not been tested for its ovicidal activity against *H. armigera*.

## MATERIALS AND METHODS

### Insect source and mass rearing

*H. armigera* larvae were collected from the fields of lady's finger, cotton and chickpea from Kannivadi in Dindigul district. The larvae collected from various hosts were maintained in the laboratory at 22 ± 2°C and 70 – 75 % RH. The larvae were reared on semi-synthetic diet (Shorey

**Table 1.** Ovicidal activity of *Cipadessa baccifera crude* extracts (in %) against *Helicoverpa armigera*

| Treatments | | Concentrations | Egg hatchability (in %) | | |
|---|---|---|---|---|---|
| | | | 24 h | 48 h | 72 h |
| *C.baccifera* extracted with | Hexane | 0.5 | 43.33 | 46.66 | 30.00 |
| | | 1.0 | 20.00 | 13.33 | 6.66 |
| | | 1.5 | 10.00 | 0.0 | 6.66 |
| | Petroleum ether | 0.5 | 3.33 | 46.66 | 23.33 |
| | | 1.0 | 3.33 | 6.66 | 0.0 |
| | | 1.5 | 33.33 | 56.66 | 0.0 |
| | Chloroform | 0.5 | 3.33 | 33.33 | 10.0 |
| | | 1.0 | 3.33 | 33.33 | 0.0 |
| | | 1.5 | 3.33 | 16.66 | 0.0 |
| | Acetone | 0.5 | 0.0 | 16.66 | 6.66 |
| | | 1.0 | 3.33 | 46.66 | 0.0 |
| | | 1.5 | 20.00 | 13.33 | 36.66 |
| | Water | 0.5 | 23.33 | 23.33 | 73.33 |
| | | 1.0 | 23.33 | 23.33 | 6.66 |
| | | 1.5 | 43.33 | 13.33 | 6.66 |
| | Hexane | | 0.0 | 53.33 | 6.66 |
| | Petroleum ether | | 0.0 | 46.66 | 16.66 |
| | Chloroform | | 26.66 | 0.0 | 6.66 |
| | Acetone | | 13.33 | 36.66 | 6.66 |
| | Untreated control | | 86.66 | 96.66 | 86.66 |
| | CD (P = 0.05) | | 9.82 | 9.19 | 8.20 |

**Table 2.** Ovicidal activity of *Clausena dentata* crude extracts (in %) against *Helicoverpa armigera*

| Treatments | | Concentrations | Egg hatchability (in %) | | |
|---|---|---|---|---|---|
| | | | 24 h | 48 h | 72 h |
| *C. dentata* extracted with | Hexane | 0.5 | 0.0 | 0.0 | 0.0 |
| | | 1.0 | 0.0 | 0.0 | 0.0 |
| | | 1.5 | 0.0 | 0.0 | 0.0 |
| | Petroleum ether | 0.5 | 0.0 | 0.0 | 16.66 |
| | | 1.0 | 10.0 | 0.0 | 13.33 |
| | | 1.5 | 0.0 | 16.66 | 0.0 |
| | Chloroform | 0.5 | 3.33 | 0.0 | 0.0 |
| | | 1.0 | 20.00 | 0.0 | 16.66 |
| | | 1.5 | 0.0 | 0.0 | 0.0 |
| | Acetone | 0.5 | 3.33 | 0.0 | 0.0 |
| | | 1.0 | 0.0 | 23.33 | 26.66 |
| | | 1.5 | 10.0 | 16.66 | 0.0 |
| | Water | 0.5 | 56.66 | 6.66 | 23.33 |
| | | 1.0 | 10.0 | 13.33 | 46.66 |
| | | 1.5 | 33.33 | 13.33 | 6.66 |
| | Hexane | | 0.0 | 20.0 | 13.33 |
| | Petroleum ether | | 0.0 | 13.33 | 0.0 |
| | Chloroform | | 26.66 | 0.0 | 0.0 |
| | Acetone | | 13.33 | 36.66 | 33.33 |
| | Untreated control | | 83.33 | 83.33 | 86.66 |
| | CD (P= 0.05) | | 8.63 | 6.21 | 6.80 |

and Hale, 1965; Sathiah, 1987) in individual containers to prevent cannibalism and contamination.

**Extraction method**

The leaves of *Cipadessa baccifera, Clausena dentata, Dodonaea angustifolia* and *Melia dubia* were collected from different parts of Kolli Hills, Tamil Nadu. The leaves of *C. baccifera, C. dentata, D. angustifolia and M. dubia* were shade dried and powdered. One kg of powdered leaves was extracted successively using non-polar to polar solvents *viz.*, hexane, petroleum ether, chloroform and acetone. In each solvent, the plant material was soaked for 24h at 30 ± 2°C, filtered and to the residue the same solvent was added. This procedure was repeated thrice to obtain maximum extractables. All the filtrates were pooled and evaporated under vacuum in a rotary evaporator (Harborne, 1998) at 190 rpm/min (the temperature varies between extracts *viz.*, 40-60°C for petroleum ether, 60-62°C for chloroform and acetone and 66-70°C for hexane). Aqueous extraction was also done. One kg of fresh leaves was macerated in a blender by adding 5 litres of autoclaved distilled water and squeezed it with cheese cloth. It was filtered with 595-Bogen sheets twice and later with vacuum filter (15 Hgs vac). The extracts were lyophilized in 100 ml slots at 55° C (26 lbs) for 18 hours per slot. The containers with water extract were immersed in liquid nitrogen. The crude extracts were measured and used in desired concentrations for bioassay. The experiment was conducted at M. S. Swaminathan Research Foundation, Chennai during 2008-09.

**Ovicidal activity**

Crude extracts of *C. baccifera, C. dentata, D. angustifolia* and *M. dubia* were chosen for the study. 1 ml of the extracts at three doses *viz.*, 0.5, 1.0 and 1.5% were topically applied by micropipette on *H. armigera* eggs of different age (24, 48 and 72 hours old). For each treatment 10 eggs with triplicate were maintained and the inhibition percentage of egg hatchability due to various treatments at various doses was recorded. The data were analyzed statistically using Agres package version 4.

**RESULTS**

The hatchability of eggs of *H. armigera* at different ages viz., 24, 48 and 72 h old were tested with topical application of crude extracts of botanicals at three different concentrations namely 0.5, 1.0 and 1.5%. The results obtai ned from various treatments are enlisted in Tables 1-4.

***Cipadessa baccifera***

Among the different extracts tested, acetone extract at 0.5% on 24 h old eggs resulted in zero hatchability. Similar arrest of egg hatchability was observed in hexane and petroleum ether solvents. 48 h old egg was unable to hatch in hexane extract at 1.5% concentration and 72 h old was suppressed by 1% concentration of petroleum ether, chloro form and acetone extracts. The earlier age was more susce ptible to the crude extract at minimum concentration itself (Table 1).

***Clausena dentata***

The results revealed that hexane extract at all the three concentrations were able to arrest the egg hatchability completely irrespective of the age (Table 2). This was followed by chloroform extract at all the 3 doses against 48 h old eggs.

***Dodonaea angustifolia***

Table 3 revealed that at the minimum dose (0.5%) petroleum ether and acetone extracts curtailed the egg hatchability of 24 h old egg (10%), whereas at 1.0% concentration chloroform extract was effective against 24 h old egg, which resulted in zero hatchability.

***Melia dubia***

Eggs of 24, 48 and 72 h old eggs were prevented from hatching by 0.5% of hexane extract. Petroleum ether extract at 0.5% dose resulted in zero hatchability of 72 h old eggs. The medium dose of 1.0% was effective in case of acetone extract against 48 h old eggs. Contrastingly, at higher doses the egg hatchability was not completely arrested (Table 4).

**DISCUSSION**

Ovicidal action on hatching (32.77%) was higher in 10% methanolic extract of *Melia azedarach* (Mukesh Mahla *et al.*, 2002). Extracts from calyxes of *Hibiscus sabdariffa* exhibited antifeedant as well as oviposition-deterrent activities against *Earias vitella* (Dongre and Rahalkar, 1992). The effects of methanolic extracts of *A. indica* and *Melia azedarach* seeds on the oviposition behaviour and hatchability of eggs of *Earias vitella* under laboratory conditions proved that eggs laid on extract-treated oviposition substrate exhibited reduced hatching and marked adverse effects on hatching were noticed when the eggs were dipped in different concentrations of extracts (Gajmer *et al.*, 2002).

Among the four plants tested for ovicidal activity, *Clausena dentata* reduced the egg hatchability and proved to be highly ovicidal compared to others. It was clear that the early stage of the eggs namely 24 h old

**Table 3.** Ovicidal activity of *Dodonaea angustifolia* crude extracts (in %) against *Helicoverpa armigera*

| Treatments | | Concentrations | Egg hatchability (in %) | | |
|---|---|---|---|---|---|
| | | | 24 h | 48 h | 72 h |
| *D.angustifolia* extracted with | Hexane | 0.5 | 20.00 | 13.33 | 36.66 |
| | | 1.0 | 3.33 | 23.33 | 0.0 |
| | | 1.5 | 10.00 | 26.66 | 0.0 |
| | Petroleum ether | 0.5 | 10.00 | 0.0 | 36.66 |
| | | 1.0 | 10.00 | 23.33 | 6.66 |
| | | 1.5 | 10.00 | 43.33 | 0.0 |
| | Chloroform | 0.5 | 20.00 | 3.33 | 26.66 |
| | | 1.0 | 0.0 | 56.66 | 16.66 |
| | | 1.5 | 0.0 | 13.33 | 0.0 |
| | Acetone | 0.5 | 10.0 | 46.66 | 3.33 |
| | | 1.0 | 3.33 | 56.66 | 6.66 |
| | | 1.5 | 3.33 | 26.66 | 0.0 |
| | Water | 0.5 | 33.33 | 10.00 | 6.66 |
| | | 1.0 | 33.33 | 13.33 | 10.00 |
| | | 1.5 | 53.33 | 23.33 | 36.66 |
| | Hexane | | 0.0 | 53.33 | 13.33 |
| | Petroleum ether | | 0.0 | 46.66 | 0.0 |
| | Chloroform | | 26.66 | 0.0 | 0.0 |
| | Acetone | | 13.33 | 36.66 | 26.66 |
| | Untreated control | | 86.66 | 96.66 | 90.00 |
| | CD (P= 0.05) | | 11.61 | 8.35 | 7.44 |

**Table 4.** Ovicidal activity of *Melia dubia* crude extracts (in %) against *Helicoverpa armigera*

| Treatments | | Concentrations | Egg hatchability (in %) | | |
|---|---|---|---|---|---|
| | | | 24 h | 48 h | 72 h |
| *M. dubia* extracted with | Hexane | 0.5 | 0.0 | 0.0 | 0.0 |
| | | 1.0 | 20.00 | 0.0 | 0.0 |
| | | 1.5 | 20.00 | 63.33 | 16.66 |
| | | 0.5 | 63.33 | 13.33 | 0.0 |
| | Petroleum ether | 1.0 | 33.33 | 6.66 | 13.33 |
| | | 1.5 | 53.33 | 0.0 | 6.66 |
| | | 0.5 | 23.33 | 13.33 | 3.33 |
| | Chloroform | 1.0 | 16.66 | 30.00 | 16.66 |
| | | 1.5 | 3.33 | 30.00 | 13.33 |
| | | 0.5 | 33.33 | 3.33 | 26.66 |
| | Acetone | 1.0 | 20.00 | 0.0 | 13.33 |
| | | 1.5 | 10.00 | 33.33 | 46.66 |
| | | 0.5 | 33.33 | 13.33 | 56.66 |
| | Water | 1.0 | 23.33 | 36.66 | 43.33 |
| | | 1.5 | 23.33 | 13.33 | 56.66 |
| | Hexane | | 0.0 | 36.66 | 10.00 |
| | Petroleum ether | | 0.0 | 26.66 | 13.33 |
| | Chloroform | | 26.66 | 23.33 | 6.66 |
| | Acetone | | 13.33 | 16.66 | 6.66 |
| | Untreated control | | 83.33 | 83.33 | 90.00 |
| | CD (P= 0.05) | | 10.64 | 9.55 | 8.59 |

eggs, to be highly susceptible to all the treatments. This opined with Kuppusamy and Murugan (2008) who noted that exposure of freshly laid *Culex quinquefasciatus* eggs to ethanolic extract of *Euphorbia heterophylla* was more effective than older eggs. The reason may be that the embryonic development would not have started or probably in its threshold. Hence, killing may be easier in causing the death of the egg. This was in confirmation with Satyanarayana Rao and Gujar (1995), wherein the toxicity of two plumbaginoids *viz*., plumbagin and juglone to the eggs of the cotton stainer, *Dysdercus koenigii* show ed less susceptibility in the middle of embryogenesis.

All the four plants efficiently inhibited the egg hatchability. The ovicidal activity was concentration dependent factor as observed by Mahmoud and Shoeib (2008) in *Bactrocera zonata* (Sunders). The failure to hatch and develop into the next stage was taken into account as one of the effects of juvenile hormone (JH) activity. Earlier works indicate, cessation of embryogenesis occurred in *S. litura* when their eggs were treated with juvenoids (Abo-El-Ghar *et al*., 1996). Enslee and Riddiford (1977) suggested that the juvenoids when treated on eggs resulted in incomplete blastokinesis in embryo and abnormal breakage of extra embryonic membranes. This may result in the failure to hatch in the treated eggs.

The experimental results proved that the biopesticides derived from less explored plants play a major role in combating the insect pests at the egg stage itself, and thereby prevent the damage caused by the larval stages. In addition, if applied at the right dosage and time it would certainly be an alternative to chemical pesticides at the field level.

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