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AZADIRACHTA INDICA EXTRACTS FOR CONTROLLING COTTON PEST MYLABRIS CICHORII LARVAE

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ABSTRACT
The important medicinal plant extract *Azadirachta indica* leaves aqueous extract against the fourth instars larvae of cotton Pest, During the experimental period insects' weight were significantly decreased from 24, 48, 72 and 96 hrs of exposure time of leaves aqueous extract of *A. indica* treated against *M. cichorii*. The high percent mortality presents in the 800-ppm concentration of *A. indica* at 96 hrs exposure time of leaves extract followed by 600, 400, and 200 ppm, respectively. The high percentage mortality was observed in the 800-ppm concentration of *A. indica* at 96 hrs exposure time of leaves extract followed by 600, 400, and 200 ppm, respectively. These results have been suggested that the *A. indica* plant leaves aqueous extracts were used as the integral pest management studies of the cotton pest, *M. cichorii*.

Key words: Biocidal activity, cotton pest management, *M. cichorii*, *A. indica*, mortality rate, weight of the insect.

INTRODUCTION
India is gifted with varied agro-climatic conditions for growing an array of vegetable crops to combat the present ill-balanced diet. The eggplant/brinjal belongs to the family Solanaceous and to the genus Solanum, species melongena. The eggplant, Guinea squash are some synonyms to brinjal. It is a hardy crop and does best in worm and moist climate.

The crop is generally grown twice or thrice in a year. The edible part, fruit is of high nutritive value and can well be compared with tomatoes. Insect pest constitutes the major limiting factors in the successful production of eggplant. In the sub-Himalayan region of north east India eggplant is cultivated at a commercial scale but insect and mite pest damage constitute a limiting factor for its successful production (Ghosh 1999).

In the subHimalayan region of north east India major vegetables like eggplant, ladysfinger cabbage, cauliflower etc are cultivated at a commercial scale but insect and mite pest damage constitute a limiting factor for its successful production (Ghosh et al., 1999; Ghosh et al., 2000; Ghosh and Senapati 2001a; Ghosh and Senapati 2001b; Chaudhury et al., 2001). Hadda/ spotted beetle (Epilachne vigintioctopunctata), aphid (Aphis gossypii), leafhopper (Amrasca biguttula biguttula), thrips (Thrips tabaci), spider mite (Tetranychus spp.), blister beetle (Mylabris pustulata Thunberg) and white fly (Bemisia tabaci) are the important pests of eggplant causes heavy damage (Ghosh, 1999). Pest complex of eggplant is very high and blister beetle (Mylabris pustulata Thunberg) (Coleoptera: Meloidae) causes heavy 118 damage to the crop. These beetles are brightly colored insects that secrete a compound containing cantharidin when disturbed. Cantharidin, a terpenoid,

produces blisters on human skin upon contact and hence these beetles are called "blister" beetles. In general, this is a minor pest, although occasional outbreaks can occur.

MATERIALS AND METHODS

Collection and preparation of leaves of *A. indica*

The leaves of *A. indica* were collected from our college campus. The leaves of the plant were washed thoroughly with distilled water to remove dust and other particles. The washed plant part is then dried at room temperature. The collected *A. indica* barks and leaves were cut into small pieces. The plant parts were dried in an incubator for 7 days at 40^0C, crushed in an electrical grinder and then the powder was separated.

Extraction Methods

For extraction of secondary metabolites cold percolation methods (Thillairajasekar *et al*., 2009; Das *et al*., 2010) was followed.

Cold percolation method

For cold percolation method, 250 gm of the powdered leaves of *A. indica* material was soaked with 750 ml (1:3 w/v) of selected aqueous in an aspirator bottle for 48 hrs at room temperature. The extract was filtered through a funnel with Whatman number 1 filter paper. After filtration, the filtered extract was poured into the distillation unit at 20º C for separation of the solvent and the secondary metabolites residue (10 mL) was evaporated and dried over sodium sulphate in a desiccator under vacuum. The crude extracts were stored in the refrigerator (LG, India) for further use. The extraction rate was calculated by weighing the crude extracts obtained from 100 g of dry plant material after extraction with respective solvent.

Pest collection and rearing

Nymphs and adults of *M. cichorii*, were collected from cotton fields. The collected pests were maintained in the insectory under laboratory conditions (temperature 28 ± 2°C, 70 ± 5 % RH and a photoperiod of 11L: 13D hrs) in transparent plastic containers (8cm height × 6.5cm diameter) containing a layer of sterile coarse sand (4cm thick). Insects were fed with its natural host cotton flower and also cotton bolls. Insects were maintained at least for 2 generations. The laboratory emerged 6-12 hrs old third stadium *M. cichorii* were used for this experiment.

Nymphicidal activity bioassay

Bioassay studies were carried out using uniform sized (24.7±0.4 mg weight), 6-12 hrs old third stadium *M. cichorii.* which was selected randomly from the stock culture. Five insects were placed in a transparent plastic container (8 cm height × 6.5 cm diameter). Different concentrations of *A. indica* leaf aqueous extracts [200, 400, 600 and 800ppm (4mg extract in 5ml diet- 800 ppm)] mixed in aqueous. Experimental animals were allowed to feed for 96 hrs continuously. 200µL of aqueous extracts of leaves was pour into the small cotton flowers and provided to the insects. The food was changed every day. Three replications were maintained for each concentration. Mortality was recorded every 24 hrs, till 96 hrs. The mortality was corrected using Abbott's formula (Abbott, 1925), if more than 10% mortality was observed in control category. The corrected mortality data was subjected to probit analysis (Finney, 1971) to find out the LC_{50}, Chi square, df and p values. After 96 hrs, live nymphs were provided with water-soaked cotton flowers till their death.

RESULTS

During the experimental time the weight of the insect was significantly decreased in 400 ppm (t=2.5; df= 5; p=0.057), 200 ppm (t=9.07; df=5; p=0.000) and 600 ppm (t=15.31; df=5; p=0.000) at

24 hrs of exposure time of leaf extract compare to control at 800 ppm the weight was increased when compare to control (Figure 1). During the experimental time the weight of the insect was significantly decreased in 400 ppm (t=3.5; df= 5; p=0.047), 800 ppm (t=9.00; df=5; p=0.000), 200 ppm (t=10.31; df=5; p=0.000) and 600 ppm at 48 hrs of exposure time of leaf extract when compare to control (Figure 2). During the experimental time the weight of the insect was significantly decreased in 600 ppm (t=22.55; df=5; p=0.000), 800 ppm (t=18.20; df=5; p=0.000), 200 ppm (t=12.10; df=5; p=0.000) and 400 ppm (t=11.10; df=5; p=0.000) at 72 hrs of exposure time of leaf extract when compare to control (Figure 3). During the experimental time the weight of the insect was significantly decreased in 200 ppm (t=12.55; df=5; p=0.000), 600 ppm (t=16.00; df=5; p=0.000), 400 ppm (t=9.10; df=5; p=0.000) and 800 ppm all the animals are dead at 96 hrs of exposure time of leaf extract when compare to control. The high percentage mortality was observed in the 800-ppm concentration of *A. indica* at 96 hrs exposure time of leaves extract followed by 600, 400, and 200 ppm, respectively (Figure 4).

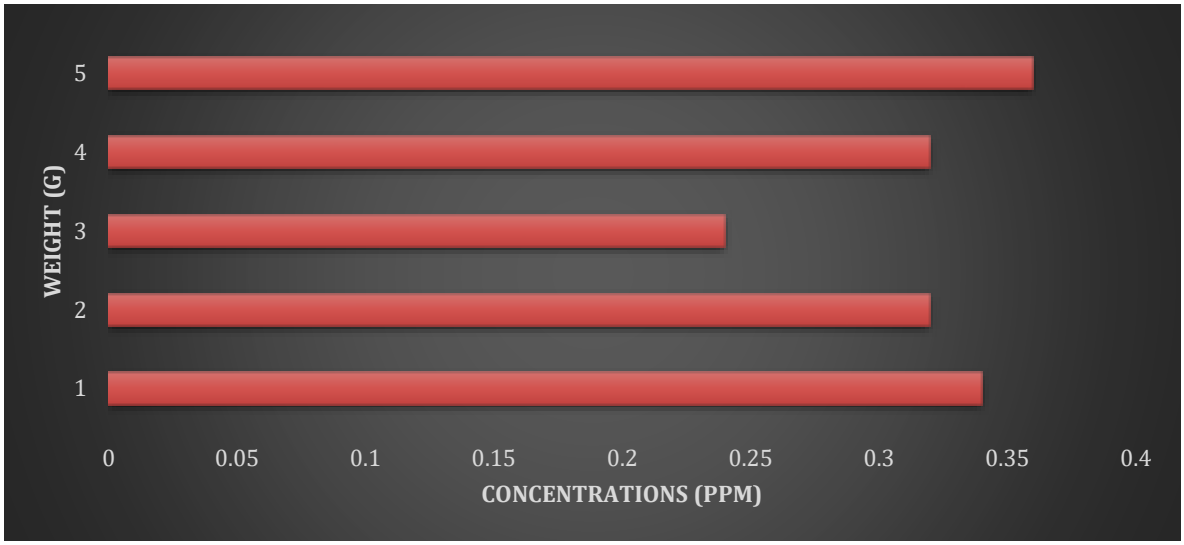

Figure 1. Effect of *A. indica* leaf aqueous extract on whole body wet weight (g) of *M. cichorii* nymphs at 24 hrs

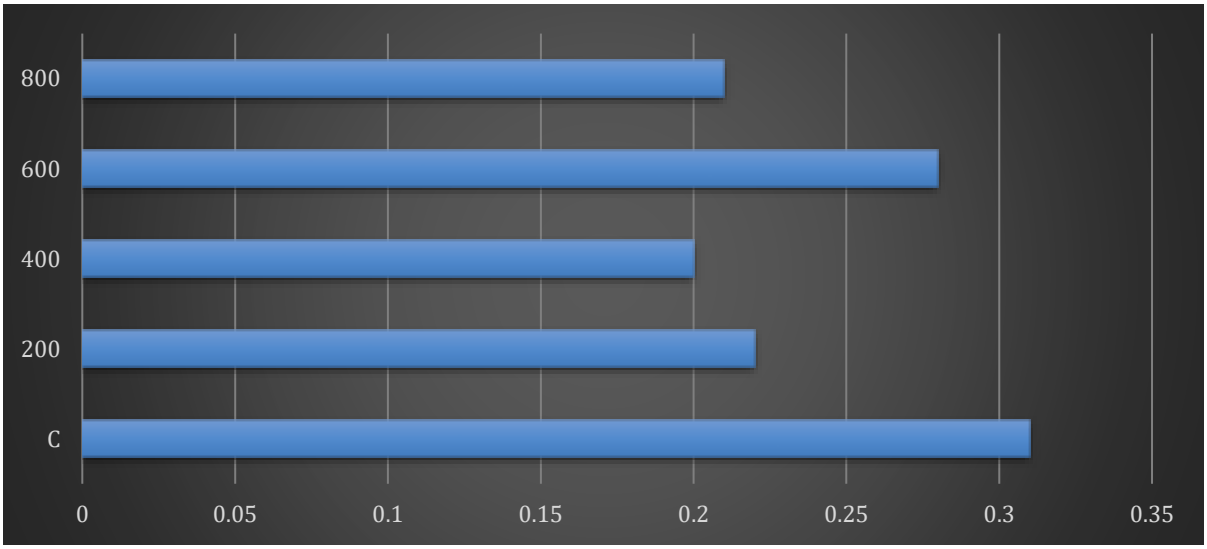

Figure 2. Effect of *A. indica* leaf aqueous extract on whole body wet weight (g) of *M. cichorii* nymphs at 48 hrs

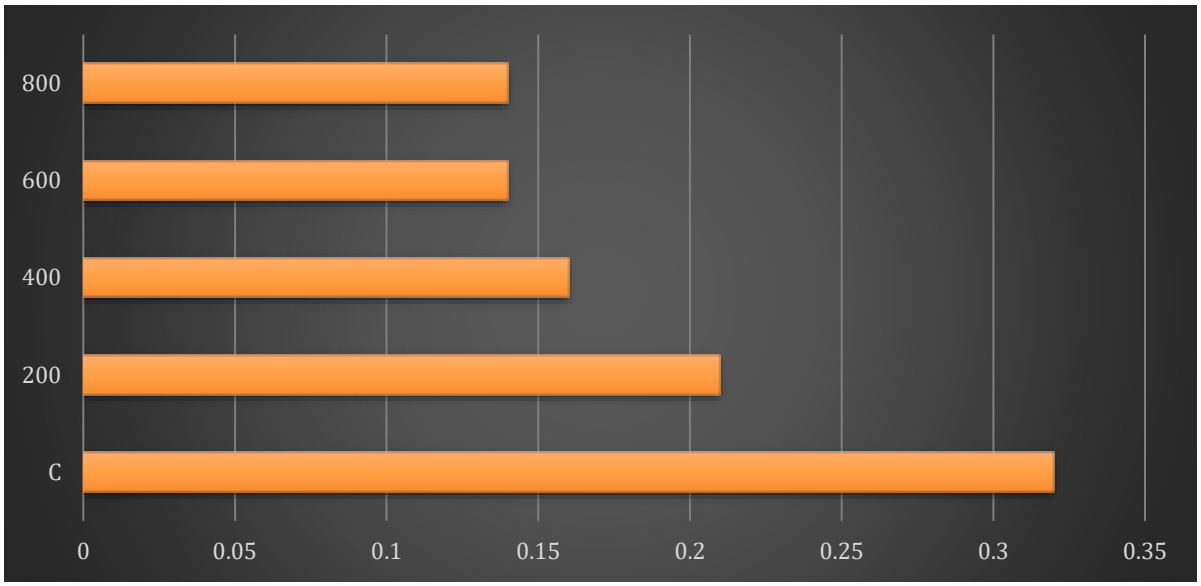

Figure 3. Effect of *A. indica* leaf aqueous extract on whole body wet weight (g) of *M. cichorii* nymphs at 72 hrs

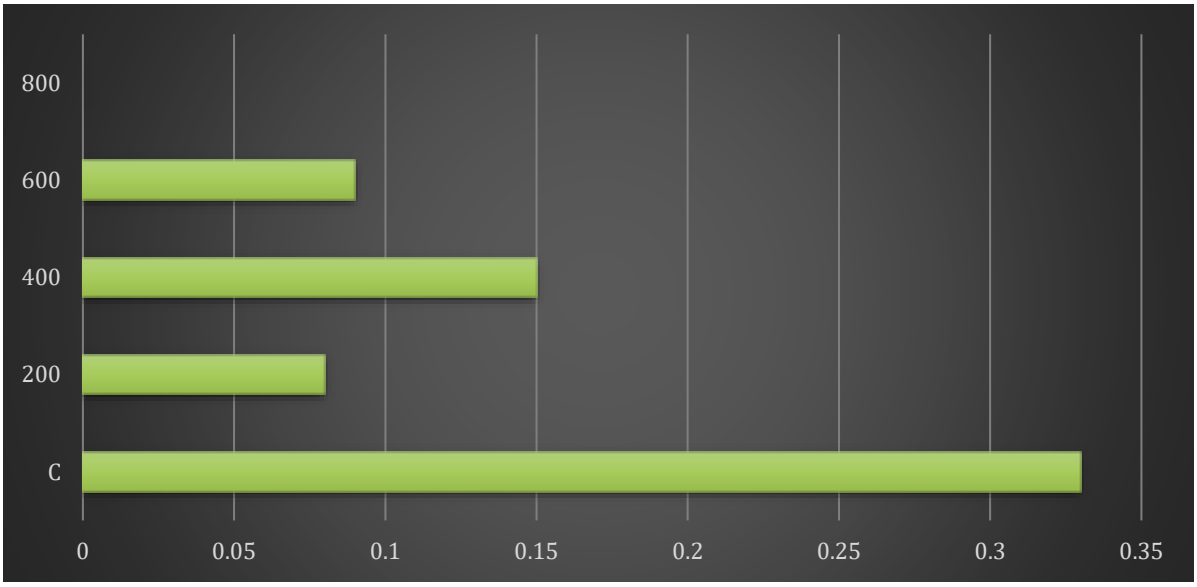

Figure 4. Effect of *A. indica* leaf aqueous extract on whole body wet weight (g) of *M. cichorii* nymphs at 96 hrs

DISCUSSION

In our result showed that *Tinosora cardifolia* plant extracts have significant antifeedant and ovicidal activity against choosed important agricultural lepidopteran field pest *Spodoptera litura* and *H. armigera*. The results are comparable with an earlier data by Pavela, 2010. investigated that the significant differences in antifeedant activity were found in the highest tested dose also between both pest species tested. *Spodoptera littoralis* larvae were less sensitive to the extracts when plant 43 extracts noticed antifeedant activity lower than the 50%, and an effective in the range of 50-95% was found in 13 plant extracts. Out of all checked extracts, only the extracts obtained from the plants *Imperatoria ostruthium*, *Angelica archangelica*, *Psorolea bituminosa*, and *Vincetoxicum hirundinaria* showed antifeedant activity loft than 95%, and their effective doses (ED50) were assessed of 44, 34, 72, and 11 µg/cm2, respectively (Krishnappa et al., 2010). They have been reported that *Taget spatula* volatile oil contained 10 compounds, and they were tested against the IV

instar larvae of *Spodoptera litura* for their antifeedant action by leaf disc experiment. Among the compounds tested, terpinolene was the most effectual feeding deterrent agent again *S. litura*. Zoubiri and Baaliuaomer (Zoubiri and Baaliuaomer, 2011), who observed that the alternatives to conventional pesticides, crucial oils extracted from aromatic plants have been widely investigated. Hamshou et al. 2010 reported that the effects of the Rhizoctonia solani lectin on the growth, development, and survival of an insect economically vital caterpillar in agriculture and horticulture, the cotton leafworm, S. littoralis were studied. Munoz et al. 2013 have reported that the extracts from the *Calceolario talcana* exhibited strong bio-insecticidal effects against *Drosophila melanogaster* and *Spodoptera frigiperda*. The most extract was ethyl acetate and its majority compound verbascoside. The loftest lethal concentration to the larvae of *S. frugiperda* and *D. melanogaster* was 20.0 µg/ml of the ethyl acetate extract with 95.8% and 67.0% of passed, respectively. Isman and Seffrin (Isman MB, Saffrin R, 2014) investigated that the crude extracts from seeds, leaves, bark, twigs, and fruits obtained from the plant species of custard-apple family are extensively tested in recent years for bioactivity to gadfly insects and connected arthropods worldwide.

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