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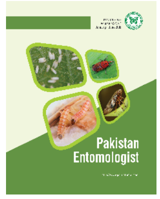

TOXICITY OF *NERIUM ODORUM* (A.) AND *PARTHENIUM HYSTEROPHORUS* (L.) BOTANICALS AGAINST *SPODOPTERA LITURA* (L.)

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ABSTRACT

Herbivory in insects has conferred remarkable fitness success and suitable host plants pose consequential effects on their biological traits. *Spodoptera litura* is a serious pest of numerous crops due to its higher fecundity, migrating efficiencies and polyphagous nature. Botanical like *Nerium odorum aiton* and *Parthenium hysterophorus* L. (leaf and flower) extracts were used to evaluate their effects on various biological activities of *S. litura*. 70-75% larval mortality was recorded when both the plant extracts were applied at 15% concentration in addition to significant antifeeding effect. Insect antifeeding and mortality has direct relation with concentration of plant extracts tested up-to a certain limit and can be used as alternate tool for the control of insect pests.

Keywords: Antifeeding index, *Nerium odorum*, *Parthenium hysterophorus*, *Spodoptera litura*, toxic botanicals

INTRODUCTION

Leaf worm, *Spodoptera litura,* is an important insect pest attacking and consuming various crops including fruits and vegetables(Summarwar and Pandy, 2013). It is found on more than 180 cultivated and wild host plant species from 45 families all over the world(Qin *et al*., 2003).
Wide range of synthetic insecticides are efficiently used to kill the insect pests in almost all cultivated crops worldwide. Injudicious exploitation of pesticides has created hazardous effects on human health that are difficult to tackle with in addition to insecticide resistance development in target organisms. Additionally, excessive use of pesticide is alarming for the insect biodiversity in term of species richness (Pimentel, 2005) and beneficial fauna of insects (Desneux *et al*., 2007). *S. litura* has also developed high resistance to most of the frequently used insecticides groups especially organophosphates, pyrethroids, carbamates, avermectins and insect growth regulators (IGR) resulting in failure of effective controls (Kranthi *et al*., 2002; Huang and Han, 2007; Ahmad *et al*., 2008). Insect pest control methods like use of plants extracts are providing a good alternative of chemical pesticides being relatively very less toxic and environment friendly. Therefore, use of plant extracts is greatly increasing in organic farming and home gardening with respect to past because of increasing awareness Regnault-Roger and Philogène, 2008).
The extracts of locally available plants can be effective as crop and stored grains protectants, either used alone or in combination with conventional insecticides at reduced rates (EL-Kamali, 2009; Baskar and Ignacimuthu, 2012;). Indigenous knowledge and traditional practice can make valuable contributions to domestic food production as well as protection in countries where strict enforcement of pesticide regulations is impractical (Mekonnen and Agonafir, 2002). Botanicals and essential oils of many plant species i.e., *Azadirectin indica*, *Blumea mollis*, *Hygrophila auriculata, Artemisia annua*, *Allium sativum, Echinacea* spp, *Panax ginseng*, *Cymbopogon* spp., *Ocimum* spp., *Eucalyptus* spp., *Nerium* spp., *Parthenium hysterophorus*, *Cassia fistula* and *Clerodendron inerme* are being used for effective control of different insect pests at lower level of farming and kitchen gardening (Sahayaraj, 1998; El-Shazly, 2000; Javaid and Anjum, 2006; Baskar and Ignacimuthu, 2012; Summarwar and Pandy, 2013).
Nerium odorum L. is a bulky glabrous evergreen horticultural

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shrub belongs to family Apocynaceae. Its leaves, fruits and flowers excretes milky juice when plucked (Kumar and Haripriya, 2010). It possess good antimicrobial activity and it is very effective against many bacterial microbes (Hussain and Gorsi, 2004; Jastaniah, 2014). *Nerium* leaf extract also possess insecticidal properties and can effectively control the blow fly maggots (El-Shazly *et al*., 2000). *Parthenium hysterophorus* is a fast-growing annual weed, which spreads rapidly. It is very common around the agricultural fields, along the roadsides and wastelands. It is an allopathic plant which usually suppress the growth of other plants around it (Javaid and Anjum, 2006). It is biologically effective to control many insect pests and its leaf extract found very effective for the control of *Aedes aegypti* L. (Kumar *et al*., 2011). Its leaf extract is also known for their antimicrobial activity against *Escherichia coli, Staphylococcus aureus, Xanthomonas vesicatoria and Ralstonia solanacearum* (Sukanya *et al*., 2009).

Botanicals are of great value but, at the moment, they occupy a minute contribution in pest management strategies including crop protection programs (Isman, 2006). Based on these lines, the current study was designed to evaluate the biological toxicity of *Nerium odorum* Aiton and *Parthenium hysterophorus* (L.) extracts on *Spodoptera litura* feeding activity on cabbage leaves treated with different concentrations of these botanicals.

MATERIALS AND METHODS

Insect collection and rearing

This research was conducted in the Entomological research laboratory, University of Sargodha in 2017. Egg masses of *S. litura* were collected from chemically untreated cabbage fields of research area of University. To get a homogenous larval culture, egg masses were allowed to hatch at 25±2 ºC and 65±5% relative humidity in a sterilized ventilated glass chamber (1.5 cubic feet). Hatched larvae were offered fresh cabbage leaf cuts and culture was maintained in climatic chamber for next progeny under the same conditions of temperature and humidity. After the hatching of second-generation larvae freely fed for three days, homogenous 2^{nd} instar larvae were selected for further study.

Preparation of plant extracts

Two kg insecticide free leaves and flowers of *N. odorum* and *P. hysterophorus* (L.) were collected from university field area, washed with distilled water and sun dried. After drying, they were grinded into very fine powder with an electric grinder (Mortar Grinder RM 200, RETSCH GmbH, Germany). Then, it was sieved through a 60-mesh sieve (Fieldmaster, USA). Ten grams of leaf and flower powder from both plants were mixed separately with 100 ml of 95% methanol in a 250 ml flask on a rotary shaker (MK1171T45, Thomas Scientific, USA) for 48 hours at 120 rpm. Afterwards, the extract solutions were filtered through filter paper (Whatman No. 40) into 250 ml beakers (Ali *et al*., 2017). These filtrates were evaporated under fume hood till a constant volume of 20 ml. These stock extracts were kept under 4 ºC till further use(;).

Bioassays

Second instar *S. litura* larvae were exposed to five different concentrations of each plant extract separately (2.5, 5, 7.5, 10 and 15%) on hit and trial basis. methanol, water and an insecticide (deltamethrin, 2.5% EC, UDL Pakistan LTD.) at recommended dose through leaf dip method in 90 mm Petri dishes provided with treated and non-treated fresh cabbage leaves on filter papers. Larval feeding and mortality data were recorded after every 12 hours for 3 days. Larval feeding was assessed by measuring the weights of feed offered and consumed after the said time interval. The antifeeding index (AI) was calculated using the following formula():

Antifeeding Index (AI) = [1-(Feed consumed in T/ Feed consumed in Co)] × 100

Where T = treated, Co = control

Ten replications were maintained for each treatment with 20 larvae in each treatment. Different extract concentrations were made form stock solution by diluting stock solution with methanol().

Statistical Analysis

The data were analyzed through factorial analysis of variance (ANOVA) using Statistix 8.1 software and the means of treatments were compared by Tuckey's HSD test (at 5% alpha) to check effect of treatments on mortality and feeding performance of larvae.

RESULTS

Effect of botanicals on feeding activity of *S. litura*

N. odorum leaf and flower extracts caused significant anti-feeding in exposed larvae which was increased with the increase in the concentrations of plant extracts. Maximum antifeeding index (AI) (75% and 79%) was observed at a 15% concentration from both of the plant parts while it was recorded 25% in methanol control treatment and was found zero in water treated leaf feeding as negative control (Fig. 1). Even, the antifeeding index of the botanicals at higher concentrations was greater than the insecticide impact. It was also noted that the antifeeding index of flower extracts was slightly greater than the leaf extracts. Similarly, maximum antifeeding index was also observed with *P. hysterophorus* leaf and flower extracts treatments at higher concentrations (Fig. 2).

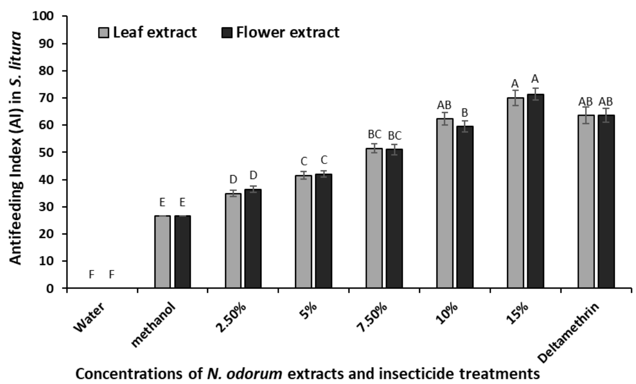

Fig. 1.
Antifeeding index for *N. odorum* leaf and flower extracts against *S. litura* (L.)

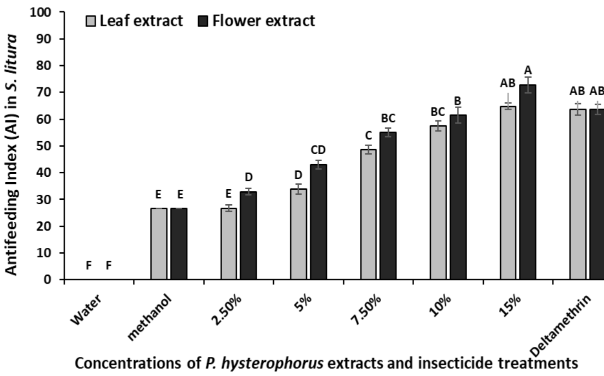

Fig. 2.
Antifeeding index for *P. hysterophorus* leaf and flower extracts against *S. litura* (L.)

Effect of botanicals on mortality of *S. litura*

Maximum mortality (85%) was observed at the highest botanical concentration (15%) for flower extracts of *N. odorum* while leaf extracts caused 75% mortality during the same time period. Overall, flower extracts caused higher mortalities in all the treatments as compared to the leaf extracts. Similarly, *P. hysterophorus* extracts caused the maximum mortalities (75% and 65%) for flower and leaf extracts at 15% botanical concentrations. Flower extracts showed more mortality at 15% concentration as compare to the leaf extracts from both of the plants. The lowest larval mortality was observed in case of methanol treatment which was 24% and 0% mortality observed in water control treatment. At 15% botanical concentration level, larval mortality was even greater than the mortality caused by the insecticide as shown in Fig. 3 and 4. Flower extracts have shown higher toxicity by causing slightly higher mortality as compared to the leaf extracts in all the treatments with higher concentrations of plant extracts but the effect of flowers and leaves extracts was non-significantly different at lower concentrations as shown in Fig. 3 and 4. The flower extracts from *P. hysterophorus* caused significantly higher mortality (Fig. 4). It was also revealed that mortality was significantly increased with increasing concentrations of leaf and flower extracts up-to 10% concentrations.

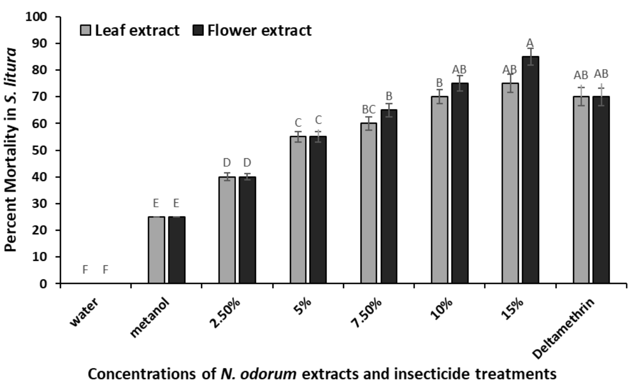

Fig.3.
Larval mortality in *S. litura* at different concentrations of *N. odorum* leaf and flower extracts

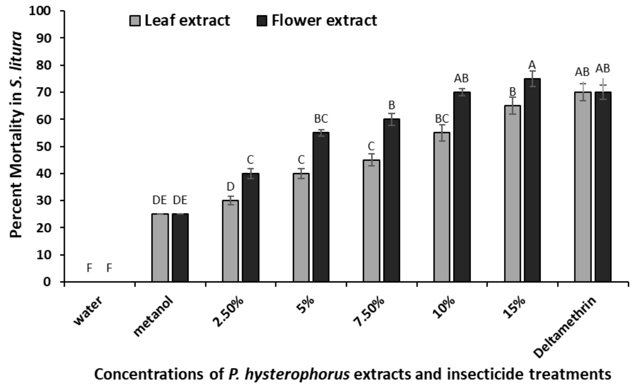

Fig.3.
Larval mortality in *S. litura* at different concentrations of *N. odorum* leaf and flower extracts

DISCUSSION

Assessment of anti-feedant effect of botanicals against insects has been considered staggering in various countries for insect control (Yasui et al., 1998; Pavunraj *et al*., 2012). From a natural point of view, antifeedants may not affect the target insects adversely and allow them to be available for their typical enemies (Jeyasankar *et al*., 2010). The use of such insect antifeedants as yield protectants is actually appealing Isman, 2002). For most antifeedants, the techniques for movement are composed at the taste cells. A standard gustatory sensillum in insects contains receptors specific for impairments and others for stimulants (sugars and amino acids). Antifeedants offer first line plant protection against different insect pests. In nutshell, antifeedants have adversarial impacts on insects (Hummelbrunner and Isman, 2001).

Anti-feedants produce phagodeterrent or phagorepellent effects and can play a crucial role in managing insect pests below economic damages (Gokulakrishnan *et al*., 2012). Plant substances used as antifeedants are found in all the compounds of discretionary plant absorption (Koul, 2008). A couple of plant discretionary metabolites are known as antifeedants, for instance, triterpenes (Van Beek and De Groot, 1986), sesquiterpenes, lactones and alkaloids (Nawrot *et al*., 1986), cucurbitacines, quinines and phenols.

The studied plant extracts possessed significant toxicity indicated by the mortality and anti-feeding response among the treated larvae. Toxicity of both extracts was concentration dependent for both of the plant species used i.e., *P. hysterophorus* and *N. odorum* but the flower extracts possess higher toxicity as compared to the extracts from leaves of the tested plants. So, antifeeding has direct association with centralization of these concentrates being antifeedant against the larvae of *S. litura* (Datta and Saxena, 1997). It might be due to the differences in the chemical compositions of the constituents of leave and flowers. Many plant extracts like *Azadirachta indica*, *Vitex negundo, Citrus sinensis* and *Zingiber officinale* caused restriction of *S. litura* by acting as a feed deterrent in terms of very low food consumption, approximate digestibility, fecal pellets production, and reduction in body weight of *S. litura* larvae *(Sahayaraj, 1998)*. Similarly, leaf extract of *Porteresia coarctata* exhibited significant anti-feeding and toxic effects to the larvae of *S. litura* and cause biochemical changes in its body. *P. coarctata* contains phytochemicals and act as a potential botanical insecticide (Ulrichs *et al*., 2008; Sreelatha *et al*., 2010). *P. hysterophorus* extracts have allergic and antifeeding properties that showed good antifeedant effect on *Tribolium casteneum* adults and sixth instar larvae of *S. litura*. It was also noted that the aqueous extract of *N. odorum* leaves showed repellent activities to the slug, *Sarasinula plebeia* (Howard *et al*., 1991).

Parthenin is the active compound present in *P. hysterophorus.* Its whole plant extract showed negative effect on insect growth when applied against the fifth instar larvae of *S. litura* (Balasubramanian, 1982). Parthenin is the major sesquiterpene lactone and exhibit a wide spectrum of biological activities, which include cytotoxic, antitumour, allergen, antimicrobial, antifeedant, phytotoxic and insecticidal properties (Rodriguez *et al*., 1976). The phytochemical analysis of various parts of *N. odorum* identified the presence of a wide range of phytochemicals such as phenolics, glycosides, alkaloids, tannin, flavonoid, etc., which show the toxic effects to the plants (Dey et al., 2012). The extracts of *N. odorum* are lethal to larvae of western-banded blow fly (El-Shazly *et al*., 2000). The insecticidal property of *N. odorum* has also been reported on larvae of *Cydia critica and Plutella xylostella* (Satphathi and Ghatak, 1990). Extracts of *P. hysterophorus* act as insect growth regulators against different insects such as *S. litura*. It is assumed that the botanical pesticides are the potential alternatives to minimize or replace the use of synthetic pesticides as they possess an array of properties including toxicity to the pest, repellency, antifinance, insect growth regulatory activities against insect pests of agricultural importance (Isman, 2002). The higher mortalities caused by these botanicals are due to their interference with the principal metabolic, biochemical, behavioral and physiological components of insects and their high obsession can cause insect mortality by affecting the nerve cells in them (Enan, 2005) as it was indicated at higher concentrations of the extracts.

CONCLUSION

Present study shows that insect antifeeding and mortality has direct relation with concentration of plant extracts tested up-to a certain limit. They can be used as alternate tool for the control of insect pests under controlled conditions especially in green houses. The different plant parts of the tested species must be chemically explored in future to identify the real target components to be used for insect pest management.

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AUTHORS CONTRIBUTION

SA conceived the experiment, MA, FF, FR, JA and MS performed experiments, MIU and SK provided technical assistance in experimentation, MAF performed statistical analyses and ZUS helped in proof reading of manuscript. All authors read the manuscript and proved for the publication.

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