



Toxicity and Behavioural Modification on Blackgram Leaf folder, *Anticarsia irrotata* Fab., (Noctuidae; Lepidoptera) Due to Wild Indigo, *Tephrosia purpurea* L.

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Bio efficacy of ethanol and n-hexane leaf extracts of wild indigo, *Tephrosia purpurea* L. was evaluated against second instar larvae of bengal gram leaf folder, *Anticarsia irrotata* under laboratory condition using leaf disc and topical method. The results of *in-vitro* study was validated

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under field conditions too. In leaf disc method, the second instar larvae were allowed to feed on the treated leaves while in topical application, the treatments were sprayed directly over the insects. Observations were recorded up to 3 days at 12 h interval. Observation on weight reduction of surviving larvae over control was analyzed. In leaf disc method, the mean mortality ranged from 8.33 per cent to 49.8 per cent with the highest mean mortality (41.1%) at 5% ethanol extract. Maximum reduction in weight was seen at 5 % conc. with 70.4 per cent followed by 64.7 per cent and followed by 55.3 per cent with 3 and 1% respectively. Similarly, when the effect of n-hexane extract was considered, the highest mean mortality of 41.6% against the second instar was seen at 5% conc. however, ethanol extract showed maximum mortality at 5% than n-hexane extract. Similarly, the maximum reduction in weight was seen at 5 % conc. with 65.4 per cent followed by 53.8 and 45.5 per cent at 3 and 1 % conc. Respectively. Similarly, when topical method was considered, 5% n-hexane extract of *T. purpurea* effected highest mean mortality (24.9%) however, the mortality rate was reduced compared to leaf disc method. Minimum mean mortality was recorded in control (8.33%) irrespective of the bioassay methods. Maximum reduction in weight was seen in at 5 % conc. with 83.5 % followed by 74 and 68.2 % with 3 and 1% conc. respectively. The maximum reduction in weight was seen at 5 % conc. with 80.4 per cent followed by 71.8 and 56.5 per cent at 3 and 1 % conc. respectively.

Keywords: Wild indigo; *Tephrosia purpurea*; blackgram leaf folder; *anticarsia irrotata*; mortality; weight reduction.

1. INTRODUCTION

“Black gram (*Vigna Mungo* L.), an important pulse, grown in India, is grown at an area of 4.02 lakh ha, production of 2.25 lakh tonnes and productivity of 559 kg/ha in Tamil Nadu. Indian blackgram ecosystem in general is a rich source of biodiversity of beneficial arthropods and insect pests. More than 50 insect pests are reported in blackgram. Moderate to heavy infestation by aphids, leaf hoppers, thrips, whiteflies, pod bugs, stink bugs, gram pod borer, spotted pod borer, field bean pod borer, pod fly and pulse beetle were the major pests reported to cause 25 to 60 per cent yield loss. Farmers since mostly preferred insecticides for the pests management, it resulted in heavy insecticide residues, secondary pests outbreak, insecticide resistance and contamination of food chain. Therefore, a study was undertaken using wild indigo plant which are eco-friendly, locally available, easily processed, inexpensive, target specific, leaving nil residue” (Chang et al., 1997). “Wild indigo, *T. purpurea* L. is a deep rooted bush-like perennial herb, up to 60–90 cm high, remaining green throughout the dry season. It is highly branched, suberect, herbaceous perennial with thin firm glabrous branches leaves narrow, green and glabrescent, glaucous and obscurely silky beneath flower fascicled, pedicels short, bracteoles minute. Colour is brown with characteristic odour and aromatic taste. Tephrosin, deguelin and quercetin are the most common metabolites of *Tephrosia*. The roots also contain isotephrosin and rotenone. Around

2.5% rutin is found in roots and leaves. Rotenoids are formed in tissue cultured plants grown under *in vitro* condition. Purpurin, a flavonone has been isolated from the seed. *T. purpurea* has a high concentration of phytochemicals that have anti-inflammatory and anti-cancer properties. Phytochemical analysis also revealed the presence of tannin, saponin, flavonoid and alkaloids in wild indigo. Studies have shown that high dose of flavonoid altered the normal body functioning of insects. Also *T. purpurea* oil was found to be effective against both larval and adult stages of mosquitoes due to flavonoid, tannin, saponin in leaf extract” (Rajamma et al., 2011). Saponin are a class of steroidal or triterpenoid secondary plant metabolite with diversified biological properties, such as antifeeding and growth inhibitory activities. *A. irrorata*, the owl moth, is a serious pest of Tamil Nadu, hence, an ecofriendly strategy was planned and executed.

2. MATERIALS AND METHODS

Homogenous culture of the host insect, *A. irrorata* was maintained under *in-vitro* condition as per the methodology of Smith et al. (1966) at 25±2°C and 70% relative humidity. In leaf disc method, second instar larvae were allowed to feed on treated leaves while in topical application, the treatments were sprayed directly over the thorax of the larvae. Observations were recorded up to 72 h at 12 h interval. Leaves of *T. purpurea* was used for the extract preparation as per the procedure of NRCS (National Research

Conservation Service, 2011). *Tephrosia* leaves collected were rinsed thoroughly with tap water to remove the inert particles. The washed plant samples were dried in hot air oven and pulverized using an electric homogenizer. The powder was refrigerated for testing its efficacy against the host insect. The powdered leaves were soaked in ethanol and hexane solvents for 72 h (3 days). After 3 days, it was filtered with Whatman filter paper no. 41. Collected extract was evaporated using rotary evaporator. The crude extract collected was used for testing its efficacy. Crude extract was weighed and required concentrations viz., 1, 3 and 5% were prepared. Two methods such as leaf disc and topical methods were employed for testing the efficacy of *T. purpurea* against *A. irrorata*. Both the bioassays were conducted with five treatments such as 1, 3 and 5 % conc. of ethanol / n-hexane extract of *T. purpurea*, solvent alone and control were tested against second instar larvae of *A. irrorata* with three replications using two methods of bioassay. Blackgram leaf discs were punctured at 2 cm dia. and dipped in test solutions for 30 sec. Treated leaf discs were dried and released with four larvae per replication placed on a petri dish. Mortality was recorded for three days at 12 h interval.

2.1 Statistical Analysis

The experimental data collected in per cent damage were transformed to arc-sine values and values of weight reduction in numbers were transformed into square root of $x+0.5$ values. After transformation, the data were analyzed using Agdata-Agres software using LSD design.

3. RESULTS AND DISCUSSION

The data obtained due to *T. purpurea* against second instar of blackgram leaf folder were analyzed, interpreted and evaluated comparatively between different solvents, different bioassay methods are given below.

3.1 Effect of Ethanol Extract of *T. purpurea* against *Anticarsia irrorata*

Bioassay was carried out with five treatments such as 1, 3 and 5 per cent ethanol extract of *T. purpurea*, ethanol (solvent) alone including a control against second instar larvae of *A. irrorata* using leaf disc method. The mean mortality ranged from minimum of 8.33 to a maximum of 41.1 per cent. The highest mean mortality (41.1%) against the second instar larva was seen at maximum conc. of 5 per cent

concentration of *T. purpurea*. The weight reduction over the control larvae were analyzed over control for the surviving larvae in different treatments. The maximum reduction in weight was seen at 5 per cent conc. with 68.8 per cent followed by 62.3 per cent and followed by 54.11 per cent at 3 and 1 per cent conc. over control respectively.

Topical bioassay was carried out with five treatments such as 1, 3 and 5 per cent ethanol extract of *T. purpurea*, ethanol (solvent) alone including a control against second instar larvae of *A. irrorata*. The mean mortality ranged from a minimum of 8.33 per cent to a maximum of 24.9 per cent. The highest mean mortality (24.9%) was seen with 5% conc. of *T. purpurea* however, the mean mortality at topical method was reduced. Minimum mean mortality was recorded at control (8.33%). The maximum reduction in weight was seen at the 5 % conc. with 61.7 per cent followed by 50.28 and 46.28 per cent with 3 and 1 per cent conc. over control respectively.

3.2 Effect of n-Hexane Extract of *T. purpurea* against *A. irrorata*

Leaf disc bioassay was conducted with different concentrations such as 1, 3 and 5 per cent n-hexane extract of *T. purpurea*, n-hexane alone and control were treated against second instar larvae of *A. irrorata*. The highest mean mortality (49.8%) against the second instar was seen at 5 % conc. Of ethanol extract of *T. purpurea* while 41.48 per cent was seen with the maximum concentration of 5% of *T. purpurea* using topical method. The maximum weight reduction in the surviving larvae was also seen at the 5 % conc. with 80 per cent followed by 71.11 and 59.4 per cent with 3 and 1 per cent conc. respectively. Solvent, n-hexane extract efficacy was higher than ethanol extract. The maximum reduction in weight of the surviving larvae was seen at % conc. with 83.33 per cent followed by 74.59 and 65.94 per cent at 3 and 1 per cent over control respectively.

According to Zhang et al. (1994) "the insecticidal activities of leaf extracts derived from 13 strains of *Tephrosia*, after being diluted to 1:500, 1:1000, and 1:1500 with water, were evaluated for their toxicity against nymph and adult aphids. Marked variation in the mortality of nymphs and adults were observed among the *Tephrosia* strains and dilution rates. The mortality rates of aphids after 24 h of treatment with 1:500 diluted extracts derived from 13 strains ranged from 8.00 to

100.00 per cent and from 9.33 to 100.00 after 48 h of treatment. At the dilution ratio of 1:1000, the mortality percentages varied from 6.00% to 88.00 % after 24 h of treatment as well as from 6.67 % to 100.00 % after 48 h of treatment with the increased dilution, the mortality rates decreased, as the 1:1500 dilution resulted in mortality percentages that differed from 2.67 % to 34.00 % at 24 h as well as from 3.33% to 48.67%. Toxins after 48 h of treatment. Regardless of the dilution, the insecticidal activities of the leaf extracts derived from *T.vogelii* collected from the U.S and Kenya were the highest among the 13 strains due to the high contents of rotenone and deguelin. The insecticidal activities of the extracts from *T. adunca* collected from Venezuela and *T. vogelii* from Bolivia and Puerto Rico were significantly lower than those of PI 257533 and PI 305346, but the extracts derived from the remaining eight strains had even lower insecticidal activities”.

Ebadollahi et al. (1995) reported that, “due to this several studies have been conducted that evaluate the pesticidal potency of plant-derived essential oils. This review presents the pesticidal efficiency of essential oils isolated from different genera of the Lamiaceae family including *Agastache Gronovius*, *Hyptis Jacquin*, *Lavandula L.*, *Lepechinia Willdenow*, *Mentha L.*, *Melissa L.*, *Ocimum L.*, *Origanum L.*, *Perilla L.*, *Perovskia Kar.*, *Phlomis L.*, *Rosmarinus L.*, *Salvia L.*, *Satureja L.*, *Teucrium L.*, *Thymus L.*, *Zataria Boissier* and *Zhumeria Rech*. Along with acute toxicity, the sub-lethal effects were illustrated such as repellency, antifeedant activity, adverse effects on the protein, lipid, carbohydrate contents on the esterase and glutathione S-transferase enzymes. Chemical profiles of the introduced essential oils and the pesticidal effects of their main components have also been documented including terpenes (hydrocarbon monoterpene, monoterpene, hydrocarbon sesquiterpene, and sesquiterpenoid) and aliphatic phenyl propanoid”.

According to Anees (2008), “One gram of crude extract was first dissolved in 100 ml of respective solvent (stock solution). From the stock solution, 1,000 ppm was prepared with dechlorinated tap water. Polysorbate 80 (Qualigens) was used as an emulsifier at the concentration of 0.05% in the final test solution. A total of 50 larvae were exposed in five replicates of ten larvae each. Experiments were maintained at $28 \pm 1^\circ\text{C}$, $65 \pm 2\%$ relative humidity. Mortality was determined 24 h after larvae were placed on

disks. Among the crude extracts tested are the chloroform extracts of acetone leaf of *O. sanctum* (LC₅₀ = 425.76). The LC₅₀ values of *O. sanctum* against the larvae of *A. aegypti* were 425.94, 150.40, 350.78, 575.26, and 175.67 and against the larvae of *C. quinquefasciatus* were 592.60, 93.92, 212.36, 76.61, and 82.12 ppm, respectively”.

Baskar et al. (2012) were studied “hexane, chloroform and ethyl acetate crude extracts of *Atalantia monophylla* leaf for ovicidal activity against *Helicoverpa armigera* at 0.5, 1.0, 2.5 and 5.0% concentrations. The ovicidal activity of 67.10% was statistically significant noticed in hexane extract at 5.0% concentration. The chloroform and ethyl acetate extracts manifested ovicidal activity of 47.49 and 43.36% respectively at 5.0% concentration. Active crude hexane extract was fractionated using silica gel column chromatography. Twelve fractions were collected and evaluated for their ovicidal activity at 125, 250, 500 and 1000 ppm concentrations. Among them, fraction 9 showed maximum ovicidal activity of 72.21% at 1000 ppm concentration with least LC₅₀ value of 435.92 ppm”.

Sahayaraj et al. (2012) reported that “toxic effect of *Tephrosia purpurea* (Linn.) and *Acalypha indica* (Linn.) Aqueous Extracts Impact on the Mortality, Macromolecules, Intestinal Electrolytes and Detoxication Enzymes of *Dysdercus cingulatus* (Fab.). Here, the toxicological effects of *Tephrosia purpurea*(Linn.) (Fabaceae) crude extract on the mortality, reproductive organs macromolecules, mineral level in alimentary canal and detoxication enzyme level in the fat body and intestine of *Dysdercus cingulatus* (Fab.) (Pyrrhocoridae) was studied”. For the past 30 years, plant extracts and their bioactive molecules have been intensively utilized as an alternative pest management component because they are safe to our health and also to our environment (Seffrin et al., 2010). “Available sources of indigenous plant material can possibly be used to control plant pests under field condition after screening them in the laboratory. Most of the novel bioactive principles of plants constituted by secondary metabolites like alkaloses, terpenoids, flavonoids, phenolic compounds, organic acids and lipids” (Harborne, 1998).

Jasmine et al. (2012) reported that “biosafety of *Tephrosia purpurea* Stem-based Formulation (Telp 3% EC) against three *Rhynocoris* species. Asian Journal of Biological Sciences, *Tephrosia*

purpurea (Dil.) Pers, (Fabaceae), is used traditionally for digestible, anthelmintic, alexiteric, leprosy, ulcers, antipyretic, alternative, cures diseases of liver, spleen, heart, blood, tumours, asthma, dyspepsia, diarrhoea, rheumatism, asthma and urinary disorders". Species of the Genus *Tephrosia* has been used as insecticide for instance *T. candida*, *T. purpurea* Pers., *T. vogelii* and *T. noctiflora* Bojer ex. Baker (Klocke, 1989). In addition, the bark of *Tephrosia purpurea* has insecticidal activity against the third instar larvae of *Plutella xylostella* (You-Zhi et al., 2011) and *Corcyra cephalonica* (Jadhav, 2009). "The roots and seeds of this plant reported to have insecticidal properties" (Hegazy et al.,

2009). "A laboratory trial was conducted to investigate the biosafety of *Tephrosia purpurea* stem-based formulation (Telp 3% EC) against three reduviid predators such as, *Rhynocoris fuscipes*, *Rhynocoris marginatus* and *Rhynocoris longifrons* adults using Y-shaped olfactometer considering olfactory response as a tool. Telp 3% EC was impregnated in Whatman No. 1 filter paper, Bt cotton leaves (BT bunny) and groundnut leaves (TMV 4). The Access Proportion Index (API) was calculated in different time intervals like 20, 40 and 60 min. It has been concluded from the results that Telp 3% EC can be incorporated along with reduviid predators in BT cotton pest management" (Jadhav, 2009).

Table 1. Effect of ethanol extract of *T. purpurea* against *A.irrorata*

Treatment	Percent Mortality after				Percent Mean Mortality	Percent weight Reduction Over Control
	12h	24h	48h	72h		
T1- <i>T. purpurea</i> 1% extract	0 (0.82)	8.3 (16.74)	0 (0.82)	8.3 (16.74)	4.15 ^b (8.78)	54.11
T2- <i>T. purpurea</i> 3% extract	0 (0.82)	8.3 (16.74)	8.3 (16.74)	8.3 (16.74)	6.22 ^c (11.63)	62.3
T3- <i>T. purpurea</i> 5% extract	8.3 (16.74)	16.6 (24.04)	8.3 (16.74)	16.6 (24.04)	12.45 ^d (20.39)	68.8
T4 -Ethanol alone	0 (0.82)	0 (0.82)	16.6 (24.04)	8.3 (16.74)	6.22 ^b (11.63)	-
T5 - Control	0 (0.82)	0 (0.82)	8.3 (16.74)	0 (0.82)	2.07 ^a (4.80)	-
CD at (0.05)	T = 1.52		O = 1.32		T*O =2.91	

Each value is a mean of five replications.

Value within parenthesis are arc-sine transformed values.

Data was analysed using Agres-Agdata Software by LSD.

In a column means followed by the same alphabets are not significantly different p=0.05% by LSD

Table 2. Effect of ethanol extract of *T. purpurea* on *A. irrorata* using topical application

Treatments	Percent Mortality after				Percent Mean Mortality	Percent weight reduction over control
	12h	24h	48h	72h		
T1- <i>T. purpurea</i> 1% extract	0 (0.82)	8.3 (16.74)	0 (0.82)	0 (0.82)	2.07 ^b (4.80)	46.28
T2- <i>T. purpurea</i> 3% extract	0 (0.82)	8.3 (16.74)	0 (0.82)	8.3 (16.74)	4.15 ^c (8.78)	50.28
T3- <i>T. purpurea</i> 5% extract	8.3 (16.74)	0 (0.82)	16.6 (24.04)	8.3 (16.74)	8.30 ^d (16.75)	61.7
T4 -Ethanol alone	0 (0.82)	0 (0.82)	8.3 (16.74)	8.3 (16.74)	4.15 ^b (8.78)	-
T5 - Control	0 (0.82)	8.3 (16.74)	0 (0.82)	0 (0.82)	2.07 ^a (4.80)	-
CD at (0.05)	T = 1.56		O = 1.38		TxO = 3.02	

Each value is a mean of five replications.

Value within parenthesis are arc-sine transformed values.

Data was analysed using Agres-Agdata Software by LSD.

In a column means followed by the same alphabets are not significantly different p=0.05% by LSD

Table 3. Effect of n-hexane extract of *T. purpurea* against *A. irrorata* using leaf disc method

Treatments	Percent Mortality after				Percent mean mortality	Percent weight reduction over control
	12h	24h	48h	72h		
T1- <i>T. purpurea</i> 1% extract	0 (0.82)	8.3 (16.74)	8.3 (16.74)	8.3 (16.74)	6.22 ^b (11.63)	59.4
T2- <i>T. purpurea</i> 3% extract	8.3 (16.74)	0 (0.82)	8.3 (16.74)	16.6 (24.04)	8.3 ^c (16.75)	71.11
T3- <i>T. purpurea</i> 5% extract	0 (0.82)	16.6 (24.04)	16.6 (24.04)	16.6 (24.04)	12.45 ^d (20.39)	80
T4 –n-hexane alone	0 (0.82)	8.3 (16.74)	8.3 (16.74)	0 (0.82)	4.15 ^b (8.78)	-
T5 – Control	0 (0.82)	8.3 (16.74)	0 (0.82)	0 (0.82)	2.07 ^a (4.80)	-
CD at (0.05)	T = 1.42		O = 1.36		TxO = 2.98	0.01

Each value is a mean of five replications.

Value within parenthesis are arc-sine transformed values.

Data was analysed using Agres-Agdata Software by LSD.

In a column means followed by the same alphabets are not significantly different p=0.05% by LSD

Table 4. Effect of n-hexane extract of *T. purpurea* on *Anticarsia irrorata* (Topical application:

Treatments	Percent Mortality after				Percent Mean Mortality	Percent Weight Reduction over Control
	12h	24h	48h	72h		
T1- <i>T. purpurea</i> 1% extract	0 (0.82)	8.3 (16.74)	8.3 (16.74)	8.3 (16.74)	6.22 ^b (11.63)	65.94
T2- <i>T. purpurea</i> 3% extract	16.6 (24.04)	8.3 (16.74)	0 (0.82)	8.3 (16.74)	8.3 ^c (16.75)	74.59
T3- <i>T. purpurea</i> 5% extract	0 (0.82)	8.3 (16.74)	16.6 (24.04)	16.6 (24.04)	10.37 ^d (18.56)	83.33
T4 –n-hexane alone	8.3 (16.74)	0 (0.82)	0 (0.82)	8.3 (16.74)	4.15 ^b (8.78)	-
T5 – Control	0 (0.82)	0 (0.82)	0 (0.82)	8.3 (16.74)	2.07 ^a (4.80)	-
CD at (0.05)	T = 1.34		O = 1.22		T*O = 2.38	0.01

Each value is a mean of five replications.

Value within parenthesis are arc-sine transformed values.

Data was analysed using Agres-Agdata Software by LSD.

In a column means followed by the same alphabets are not significantly different p=0.05% by LSD

(Rao et al. (1936) reported “a comprehensive review on ethnomedicine, phytochemistry, pharmacology, and toxicity of *Tephrosia purpurea* (L.) Pers. The present review comprehensively summarizes the ethnomedicine, phytochemistry, pharmacology, and toxicology of *T. purpurea*. Further research on elucidation of the structure-function relationship among active compounds, understanding of multi-target network pharmacology and clinical applications will intensify its therapeutic potential”.

(Kumar et al. (2012) reported “larvicidal activity of *Tephrosia purpurea*, (L) against the larvae of *Culex quinquefasciatus*. Journal of Applied Pharmaceutical Science. The aim of this work was to study the larvicidal activity of *Tephrosia purpurea* (L) Pers. against the larvae of *Culex quinquefasciatus*. The preliminary laboratory trail was undertaken to determine the efficacy of petroleum ether and ethyl acetate extract of dried whole plant of *Tephrosia purpurea* belonging to the family Papilionaceae at various concentrations against the late third or early fourth instar larvae of *Culex quinquefasciatus* by

following the WHO guidelines. The results suggest that 100% mortality. Petroleum ether and ethyl acetate extract of *Tephrosia purpurea* (L) Pers. was observed at 250ppm and 300ppm respectively. The results suggested that use of plants in insect control as an alternative method for minimizing the noxious effect of some pesticide compound on the environment. Thus the extract of *Tephrosia purpurea* delivers promising more selective and biodegradable agent”.

(Elsheikh et al. (2014) evaluated “the toxicity effect of *Tephrosia Purpurea* extracts against filarial vector *Culex quinquefasciatus*. Excessive in using insecticides has led to damage to the environment and public health, which led to reconsideration in use the natural pesticides of plant origin in the fight against mosquitoes. Laboratory studies were conducted to evaluate the toxicity effect of methanolic leaves and stems extracts of the plant *Tephrosia purpurea* (Fabaceae) against the mosquito *Culex quinquefasciatus*. The efficacy of stems extract against 3rd instar larvae seemed to be less effective with LC₅₀ 2348ppm than leaves extract with LC₅₀ 58.3ppm. Leaves extract showed a remarkable reduction effects on adult emergence, fecundity and fertility more than stems extract. Both leaves and stems extract exerted delayed toxic effect on the pupae and adults resulted from treated larvae. Furthermore, some morphological deformities (pupal-adult intermediate) were observed after treatment with leaves extract. The current results considered promising to move forward in studying the bioactive plants which form an environmentally sound alternative for the synthetic insecticides”.

(Wafaa et al. (2017) reported “botanical insecticide as simple extractives for pest control, Cogent Biology. According to this article, one of the most important global problems is protecting crops from insects. In this, we review the use of plant compounds (essential oils, flavonoids, alkaloids, glycosides, esters and fatty acids) having anti-insect effects and their importance as an alternative to the chemical compounds used in the elimination of insects in different ways, namely repellents, feeding deterrents/antifeedants, toxicants, growth retardants, chemosterilants, and attractants. Botanical insecticides affect only target insects, not destroy beneficial natural enemies and provide residue-free food and safe environment. We, therefore, recommend using botanical insecticides as an

integrated insect management program which can greatly reduce the use of synthetic insecticides”.

(Sangavi et al. (2017) have reported that “the experiments were conducted under laboratory conditions to study the anti-insect activities of locally available plants against lepidopteran insect where the highest anti-feedant activity against *P. xylostella* was observed in *P. juliflora* (58.41 %) and the oviposition deterrence percentage by free choice test was found to be the highest in *S. grandiflora* (85.95 %) and by no choice test was found to be the highest in *P. juliflora* (61.26 %)”.

(Hanem et al. (2013) reported that “*Hyptis brevipes* (Lamiaceae) extracts are shown to exhibit strong insecticidal activity against the 3rd instar larva of the cotton leaf worm *Spodoptera littoralis* (Lepidoptera: Noctuidae), inducing complete larval mortality due to the arrest and/or disruption of metamorphosis. This disruption induced a wide range of abnormalities. Two active compounds were isolated from the extract following bioassay-guided fractionation and were identified as 5-hydroxy-7,4'-dimethoxy-flavon-3-ol and 5-hydroxy-7-methoxy-2-(4'-methoxyphenyl)-chromen-4-one based on spectroscopic data. This is the first report of these secondary metabolites in *H. brevipes*”.

A study was conducted by Sushilkumar et al (2018) to determine “the preference and host range of a polyphagous lepidopteron, *Spodoptera litura*, collected during a search for biocontrol agent of *Trianthema portulacastrum* L. culture of *S. litura* was maintained on *T. portulacastrum* leaves at 26 ± 2 °C and 70 ± 5% RH. The experiment was done using 10 days old larvae of *S. litura* obtained from the laboratory reared nucleus culture. Forty five plant species of crops and weeds belonging to 21 families were used for host preference study. In each replication, 10 larvae were placed on bouquet of various crop and weed plants in well aerated large size containers (2x3x2 ft.). Out of the 45 crop and weed plants tested, larvae of *S. litura* showed high, moderate, low and nil preference for 15,12,7 and 9 plant species, respectively. Among the crop plants, maximum preference was observed on *Lycopersicon esculentum* Mill., *Spinacea oleracea* L., *Brassica oleracea* L. var. *capitata* and *Trifolium alexandrinum* L. Among the 25 weed plants tested, high feeding preference was observed on *Alternanthera philoxeroides*

Mart., *Euphorbia hirta* L., *Eichhornia crassipes* Mart., *Trianthema portulacastrum* L., *Parthenium hysterophorus* L., *Cichorium intybus* L., *Rumex obtusifolius* L., *Chenopodium album* L., and *Ipomoea fistulosa* Mart”.

(Mogili Ramaiah et al. (2018) reported “the biology, morphometrics and geometrical progression of *Spodoptera litura* by rearing *S. litura* under laboratory conditions during 2017. The study was mainly focused on observing morphology of different stages i.e., egg, larva, pupa and adult along with the duration, *S. litura* had five instars. Measurements of all the stages of insect life cycle were recorded. The width of head capsule was recorded at each moult 0.25, 0.36, 0.50, 0.70 and 1.11 mm respectively. The mean values of head capsule width observed (0.36 to 1.11 mm) and estimated (0.36 to 1.10 mm) and the progression factor in the growth of *S. litura* was observed as 1.45, which indicated that an increase in head width during successive instars was very slightly varying from Dyar’s law but followed a geometric progressive in growth. Due to the several studies by Ebadollahi et al (2020), they evaluate the pesticidal potency of plant-derived essential oils. This review presents the pesticidal efficiency of essential oils isolated from different genera of the Lamiaceae family including *Agastache Gronovius*, *Hyptis Jacquin*, *Lavandula* L., *Lepechinia Willdenow*, *Mentha* L., *Melissa* L., *Ocimum* L., *Origanum* L., *Perilla* L., *Perovskia Kar.*, *Phlomis* L., *Rosmarinus* L., *Salvia* L., *Satureja* L., *Teucrium* L., *Thymus* L., *Zataria Boissier*, and *Zhumeria Rech.* Along with acute toxicity, the sub lethal effects were illustrated such as repellency, anti feedant activity, and adverse effects on the protein, lipid, and carbohydrate contents, and on the esterase and glutathione S transferase enzymes. Chemical profiles of the introduced essential oils and the pesticidal effects of their main components have also been documented including terpenes (hydrocarbon monoterpene, monoterpene, hydrocarbon sesquiterpene, and sesquiterpene)”).

According to (Sashnka Sekhar Dash et al. (2020), Bioassay studies were conducted to determine the relative toxicity of selected novel insecticides viz., chlorfluazuron, chlorantraniliprole, spinetoram, indoxacarb, emamectin benzoate and spinosad against second instar larvae of *S. Litura* by topical application, residue method and surface diet method.

4. CONCLUSION

Studies have shown that high dose of flavonoid altered the normal body functioning of insects. Also T. purpurea oil was found to be effective against both larval and adult stages of mosquitoes due to flavonoid, tannin, saponin in leaf extract. The data on the toxicity of insecticides to *S. litura*, clearly indicated that among the six insecticides tested against the second instar, emamectin benzoate, spinetoram, indoxacarb, chlorfluazuron, chlorantraniliprole showed greater toxicity against *S. litura* compared to spinosad in all the three methods of application.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of this manuscript.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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