

**BIOEFFICACY OF *Quisqualis indica* L. AND *Samadera indica*
GAETRN. AGAINST TOBACCO CATERPILLAR,
Spodoptera litura FABRICIUS (LEPIDOPTERA: NOCTUIDAE)
IN POLYHOUSE CONDITION**

ANUSREE S. S.

(2014-11-120)

**DEPARTMENT OF AGRICULTURAL ENTOMOLOGY
COLLEGE OF AGRICULTURE
VELLAYANI, THIRUVANANTHAPURAM-695522
KERALA, INDIA**

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by

ANUSREE S. S.

(2014-11-120)

THESIS

**Submitted in partial fulfillment of the
requirements for the degree of**

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DEPARTMENT OF AGRICULTURAL ENTOMOLOGY

COLLEGE OF AGRICULTURE

VELLAYANI, THIRUVANANTHAPURAM-695522

KERALA, INDIA

2016

DECLARATION

I, hereby declare that this thesis entitled “**BIOEFFICACY OF *Quisqualis indica* L. AND *Samadera indica* GAETR. AGAINST TOBACCO CATERPILLAR, *Spodoptera litura* FABRICIUS (LEPIDOPTERA: NOCTUIDAE) IN POLYHOUSE CONDITION**” is a bonafide record of research work done by me during the course of research and the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title, of any other University or Society.

Vellayani,
Date: 04-10-2016

Anusree S. S.
(2014 - 11-120)

CERTIFICATE

Certified that this thesis entitled “**BIOEFFICACY OF *Quisqualis indica* L. AND *Samadera indica* GAETR. AGAINST TOBACCO CATERPILLAR, *Spodoptera litura* FABRICIUS (LEPIDOPTERA: NOCTUIDAE) IN POLYHOUSE CONDITION**” is a record of bonafide research work done independently by Ms. Anusree S. S. (2014-11-120) under my guidance and supervision and that it has not previously formed the basis for the award of any degree, diploma, fellowship or associateship to her.

Vellayani,

Date: 04-10-2016

Dr. Nisha M. S.

(Major Advisor, Advisory Committee)

Assistant Professor and Head

Department of Nematology

College of Agriculture, Vellayani

Thiruvananthapuram- 695 522

CERTIFICATE

We, the undersigned members of the advisory committee of Ms. Anusree S. S. (2014-11-120), a candidate for the degree of **Master of Science in Agriculture** with major in Agricultural Entomology, agree that the thesis entitled **“BIOEFFICACY OF *Quisqualis indica* L. AND *Samadera indica* GAETRN. AGAINST TOBACCO CATERPILLAR, *Spodoptera litura* FABRICIUS (LEPIDOPTERA: NOCTUIDAE) IN POLYHOUSE CONDITION”** may be submitted by Ms. Anusree S. S., in partial fulfilment of the requirement for the degree.

Dr. Nisha M. S.
(Chairman, Advisory Committee)
Assistant Professor and Head
Department of Nematology
College of Agriculture, Vellayani

Dr. Narayana R.
(Member, Advisory Committee)
Assistant Professor
Department of Nematology
College of Agriculture, Vellayani

Dr. K. Sudharma
(Member, Advisory Committee)
Professor and Head
Department of Agricultural Entomology
College of Agriculture, Vellayani

Dr. Thomas George
(Member, Advisory Committee)
Professor
Department of SS & AC
AINP on Pesticide Residues
College of Agriculture, Vellayani

EXTERNAL EXAMINER

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LIST OF ABBREVIATIONS AND SYMBOLS USED

°C	Degree Celsius
%	Per cent
ANOVA	Analysis of variance
ai	Active ingredient
CD	Critical difference
cm	Centimeter
EC	Emulsifiable concentrate
<i>et al.</i>	and other co workers
Fig.	Figure
Kg	Kilogram
LD ₅₀	Lethal dose required for killing 50 per cent of test insect
LT ₅₀	Lethal time taken for killing 50 per cent of test insect
L.	Litre
ml	Millilitre
µg	Microgram
µl	Microlitre
NS	Not significant
NSKE	Neem Seed Kernel Extract
No.	Number
ppm	Parts per million
sp.	Species
<i>viz.</i>	Namely

Introduction

1. INTRODUCTION

The world has faced a new revolutionary change when polyhouses were introduced into agriculture. The polyhouse cultivation enhanced crop yield four to eight times than that of open field cultivation and ensures year round production of quality produce. This could be attributed to the peculiar structure of polyhouse, which keep the pest and pathogens away. But the pits and falls in the construction of polyhouses led to pest incidence. The major pests encountered under polyhouse cultivation include *Spodoptera litura* (Fabricius), *Helicoverpa armigera* (Hubner), *Plutella xylostella* (L), *Trialeurodes vaporariorum* (Westwood), *Myzus persicae* (Sulzer), *Liriomyza trifolii* (Burgess) and *Thrips tabaci* (Lindeman) (Vashisth *et al.*, 2013). The favourable environment within the polyhouses causes rapid multiplication of the pest resulting in significant crop loss. Capsicum infested by mites and thrips under protected condition recorded around 20-35 per cent crop loss (Nandini *et al.*, 2012). Most of the polyhouse farmers rely upon synthetic chemicals to manage the insect pests.

The extensive and indiscriminate use of chemical pesticides resulted in food contamination, environmental pollution and resistance build up in insect pests (Bami, 1997; Tong *et al.*, 2004). The persistence of insecticides is more severe under polyhouses than open field conditions resulting in residue problems. Nowadays people are more concerned about safe and quality food and the pesticide residue problems in agricultural produce became a matter of concern. This demands a search for safer alternatives, which are environmental friendly and economically viable.

In many parts of the world, locally available plants were used to manage insect pests as an ancient technology (Roy *et al.*, 2005). According to Jayaraj (1991), around 1005 species of plants exhibit insecticidal properties; 384 antifeedant, 297 repellent, 27 insect attractant and 31 growth inhibitory activities. Plant based pesticides do not promote drug resistance and are easily degradable and do not cause

negative impact on non-target pests. Usage of plant products as pesticide keeps the agricultural produce safe and protects the environment from getting polluted.

The locally available plants such as *Azadirachta indica* Juss, *Lantana camara* L., *Tegetes erecta* L. and *Nerium indicum* Mill. were found effective to manage potato tuber moth, *Phthorimaea operculella* (Zeller) under polyhouse condition (Thakur and Chandla, 2013). Among the various plants, Neem (*A. indica*) has gained worldwide recognition and it was commercially exploited as a substitute for synthetic chemicals, but the less availability of this botanical with growing demand and increased pesticide resistance warrant a thorough search for newer molecules of plant origin with different mode of action. Most of the studies to manage pests using botanicals have been conducted in open field conditions and information on its eco-friendly management under protected cultivation is limited.

Considering the above perspectives, the present study entitled “Bioefficacy of *Quisqualis indica* L. and *Samadera indica* Gaetrn. against tobacco caterpillar, *Spodoptera litura* Fabricius (Lepidoptera: Noctuidae) in polyhouse condition” was undertaken with the following objectives:

- To evaluate the antifeedant effect, insecticidal effect, effect on adult emergence, fecundity and egg hatching of aqueous and solvent extracts of *Q. indica* flower and *S. indica* leaf against *S. litura* under *in vitro* condition.
- To standardize appropriate methods of extraction and application of botanicals.
- Evaluation of promising botanicals in comparison with bioagent and chemical by pot culture study under polyhouse condition.

Review of Literature

2. REVIEW OF LITERATURE

A comprehensive review of the present study on the bioefficacy of two botanicals, *Quisqualis indica* L. and *Samadera indica* Gaetrn against tobacco caterpillar, *Spodoptera litura* Fabricius by virtue of their antifeedant effect, insecticidal effect and effect on adult emergence, fecundity and egg hatching has been presented here.

2.1. BIOEFFICACY OF AQUEOUS EXTRACTS OF PLANTS

2.1.1. Antifeedant Effect

Ladhari *et al.* (2012) evaluated the antifeedant activity exhibited by the aqueous extracts of spiderflower, *Cleome arabica* L. against third instar larvae of cotton leaf worm, *Spodoptera littoralis* Boisduval. Highest antifeedant activity was observed in stem and leaf extracts (21.75 %) followed by siliquae extracts (8.82 %), while aqueous extract of seeds exhibited phagostimulant activity at 10000 ppm.

The aqueous extracts of *Allium sativum* L. bulb, *Azadirachta indica* Juss. leaf and *Ricinus communis* L. leaf at 1 per cent concentration exhibited 72, 53 and 48 per cent antifeedant activity against *Papilio polytes* L. respectively (Elayidam, 2014).

2.1.2. Insecticidal Effect

Nathala and Dhingra (2006) reported that the aqueous extracts of *Caesalpinia crista* L. seeds exhibited insecticidal action against third instar larvae of *Helicoverpa armigera* (Hubner) with LC 50 value of 0.0891 per cent. Arora *et al.* (2011) reported the insecticidal effect of aqueous extracts of *Withania somnifera* L. against mature late instar larvae of *Tribolium castenum* Herbst. The stem, leaf and root aqueous extracts at 10 per cent concentration exhibited mortality of 35.86, 39.99 and 46.20 per cent respectively.

Onunkun (2012) reported that the aqueous extracts of *Jatropha curcas* L., *Vernonia amygdalina* Delile and *Annona squamosa* L. at 10 per cent concentration reduced the population of *Podagrira sjostedti* Jacq. in okra at 63, 58, and 55 per cent

respectively. Ravi (2013) reported cent per cent mortality in neem seed kernel extract 5 per cent, garlic emulsion 2 and 20 per cent and sweet flag emulsion 10 per cent against third instar larvae of *S. litura*. Silva *et al.* (2013) reported that the water extract of *Peumus boldus* Molina leaves showed 75 and 30 per cent mortality against third instar larvae of *Spodoptera frugiperda* J.E. Smith and *Helicoverpa zea* Boddie with LC 50 values of 2.31 and 16.05 ml/kg respectively at seven days after treatment.

2.1.3. Effect on Adult Emergence

The bioefficacy of crude aqueous extracts of *Eupatorium triplinerve* M. Vahl leaves against *S. litura* was reported by Kandagal and Khetagouder (2012). Maximum larval mortality of 92 per cent was recorded in 15 per cent concentration of the extract from fourth instar to adult stage with 46.66 per cent mortality in prepupae, 20 per cent deformity in pupae and 25.66 per cent deformity in adults. An increase in mortality was observed with increased dose of the extract. The EI 50 and EI 90 values of *E. triplinerve* were 4.07 and 14.10 per cent respectively.

Elayidam (2014) reported the growth inhibitory effect of aqueous plant extracts against *P. polytes*. The aqueous extracts of *A. sativum* bulb, *A. indica* leaf, *R. communis* leaf at one per cent concentration exhibited 32, 59 and 45 per cent normal adult emergence respectively.

Samia *et al.* (2016) reported the adverse effect of aqueous extracts of *Copaifera langsdorffii* F. leaves and bark on the biology of *S. frugiperda*. Treatment of second instar larvae with bark extract 5 per cent caused significant reduction in larval weight (342 mg) compared to control (686 mg). They also reported that the aqueous extracts did not have any influence on larval and pupal survival, duration of the pupal stage, sex ratio, duration of pre-oviposition period, and female fecundity.

2.1.4. Effect on Fecundity and Egg Hatching

Elayidam (2014) reported that the aqueous extracts of *A. sativum* bulb, *A. indica* leaf and *R. communis* leaf at one per cent concentration exhibited 88, 51 and 79 per cent ovicidal activity against *P. polytes* respectively. The oviposition deterrent

activity of 58 and 45 per cent was recorded in *A. sativum* bulb and *A. indica* leaf aqueous extracts at one per cent concentration.

2.2. BIOEFFICACY OF SOLVENT BASED PLANT EXTRACTS

2.2.1. Antifeedant Activity

Bai (1996) reported that the crude methanolic extracts of *Thevetia neriifolia* Juss. seeds at one per cent concentration could impart 90 per cent feeding inhibition to third instar grubs of *Henosepilachna vigintioctopunctata* Fabricius. Abdel-Rahman and Al-Mozini (2007) reported the antifeedant action exhibited by crude petroleum ether extracts of *Calotropis procera* (Aiton) W. T. Aiton (18.00 %), *Rhazya stricta* Decne (52.96 %) and *Solenostemma argel* (Delile) Hayne (26.76 %) against fourth instar larvae of cotton leaf worm, *S. littoralis*. Rani and Rajasekharreddy (2009) reported the antifeedant action of wild Indian almond, *Sterculia foetida* L. seed extracts against castor semilooper, *Achaea janata* L. Acetone extract of *S. foetida* seed at 4 mg/cm² exhibited cent per cent feeding deterrent activity to third instar larvae of *A. janata*.

Baskar *et al.* (2010) reported the feeding deterrent action of *Couroupita guianensis* (Aubl.) against third instar larvae of *H. armigera*. Maximum antifeedant activity (81.67 %) was recorded in hexane leaf extract at 5 per cent concentration, followed by chloroform and ethyl acetate extracts (73.68 and 69.70 % respectively).

Jeyasankar *et al.* (2010) conducted studies on the antifeedant activity of *Syzygium lineare* Wall against fourth instar larvae of *S. litura*. They observed that ethyl acetate crude extract of *S. lineare* leaves at 5 per cent concentration exhibited 79.4 per cent antifeedant activity. Further fractionation of bioactive ethyl acetate extract revealed that maximum antifeedant activity of 53.98, 60.54 and 91.58 per cent was exhibited by fraction 1, 3 and 6 respectively at 1000 ppm concentration. Least antifeedant activity was shown by fraction 2 and 7 at 125 ppm concentration (7.15

and 10.6 % respectively). The phytochemical analysis of fraction 6 confirmed the presence of coumarin, quinone, alkaloids and terpenoids.

Vendan *et al.* (2010) reported the antifeedant activity of ethyl acetate crude extract of *Hydnocarpus alpina* Wt. leaves (72.2 %) against third instar larvae of *S. litura*. Fractionation of ethyl acetate extract confirmed maximum antifeedant action of 68.5 per cent in eighth fraction. Krishnappa *et al.* (2010) studied the feeding deterrent action of compounds isolated from the volatile oil of *Tagetes patula* L. against fourth instar larvae of *S. litura*. It has been found that terpinolene exhibited appreciable antifeedant activity of 89 per cent at 5 mg/cm².

Pavunraj *et al.* (2011) reported the effect of crude extracts of *Pergularia daemia* (Forssk) Choiv. on the feeding behaviour of *S. litura* and *H. armigera*. Ethyl acetate crude extract of *P. daemia* leaves showed maximum antifeedant activity of 71.82 and 70.30 per cent against fourth instar larvae of *S. litura* and *H. armigera* respectively. Fractions collected from ethyl acetate extract at hexane and ethyl acetate (80:20) yielded a compound, 6-(4,7-hydroxy-heptyl) quinone, which exhibited 80.22 per cent (*H. armigera*) and 68.31 per cent (*S. litura*) antifeedant activity at 2000 ppm.

Baskar *et al.* (2011a) reported the antifeedant property of leaf and root extracts of *Aristolochia tagala* Cham. against third instar larvae of *S. litura*. Highest antifeedant activity of 56.06 and 49.86 per cent was recorded in ethyl acetate and hexane leaf extracts of *A. tagala* respectively at 5 per cent concentration. Minimum antifeedant activity (31.71 %) was exhibited by ethyl acetate extract at 5 per cent concentration.

The antifeedant activity of *C. procera* leaf extract on third instar larvae of *S. litura* was reported by Bakavathiappan *et al.* (2012). Chloroform extract at 5 per cent concentration recorded 63.42 % feeding inhibition, followed by hexane, ethanol, acetone, ethyl acetate and methanol extracts. Ladhari *et al.* (2012) reported that the methanol extract of *C. arabica* siliquae showed 37.89 % antifeedant activity followed

by seed (32.15 %), stem and leaf (7.8 %) extracts at 10000 ppm against third instar larvae of *S. littoralis*.

Yogesh *et al.* (2013) reported that the crude methanol extracts of *Parthenium hysterophorus* L. and *Ageratina adenophora* (Spreng.) King & H. Rob exhibited 69.92 and 63.47 per cent antifeedant activity at 10 per cent concentration respectively against fifth instar larvae of *S. frugiperda*.

Arivoli and Tennyson (2013) reported that the ethyl acetate extract of *Strychnos nux-vomica* L. (88.98 %), hexane extract of *Vitex negundo* L. (86.41 %) and *Murraya koeingii* (L.) Sprengel (81.46 %), ethyl acetate extract of *Zanthoxylum limonella* Alston (80.58 %) and hexane extract of *Abrus precatorius* L. (78.61 %) exhibited significant antifeedant activity against third instar larvae of *S. litura*. The acetone extract of *Dodonaea viscosa* Jacq. and water extract of *Adathoda vasica* Nees at 10 per cent concentration were reported to possess antifeedant activity against third instar larvae of European corn borer, *Ostrinia nubilalis* Hubner with antifeedant indices of 33.58 and 30.50 respectively (El-Hefny and Amany, 2013).

Sivaraman *et al.* (2014a) screened *S. nux-vomica* and *Semicarpus anacardium* L. seed extracts against third instar larvae of *H. armigera* for their antifeedant action. Hexane, chloroform, ethyl acetate and methanol extracts at 0.5, 1.0, 1.5 and 2 per cent concentrations were tested. The chloroform extract of *S. anacardium* seeds at 2 per cent concentration exhibited 74.27 per cent antifeedant activity. The hexane extract of *S. nux-vomica* seeds recorded maximum antifeedant activity of 70.57 per cent at 2 per cent concentration.

Sivaraman *et al.* (2014b) reported that the methanol extract of *Sinapis alba* L. seeds showed 71.42 per cent antifeedant activity against third instar larvae of *H. armigera*. The phytochemical analysis of methanol extract confirmed the presence of alkaloids, quinones and saponins. Jeyasankar *et al.* (2014) reported the

antifeedant activity of *Barleria buxifolia* L. leaf extracts against insect pests. The maximum antifeedant activity was exhibited by ethyl acetate extract at 5 per cent concentration against fourth instar larvae of *S. litura* (78.5 %) and *H. armigera* (75.4%). The phytochemical analysis of ethyl acetate extract showed the presence of terpenoids, flavanoids, phenols and quinones.

Pavunraj *et al.* (2014) reported the antifeedant activity of different solvent extracts of *Spilanthes acmella* (L.) Murr. leaves against third instar larvae of three economically important pests, *S. litura*, *H. armigera* and *Earias vitella* Sherborn. Maximum antifeedant activity was observed in dichloromethane extract at 5 per cent concentration against *H. armigera* (53.22 %), followed by dimethyl sulfoxide, acetone and aqueous extracts. Dichloromethane extract at 5 per cent concentration recorded 65.43 and 56.72 per cent feeding inhibition against *S. litura* and *E. vitella* respectively, followed by acetone, dimethylsulfoxide and aqueous extracts.

Vattikonda (2015) reported that 'Andrographolide' isolated from *Andrographis paniculata* (Burm. F.) Wall. at 200 ppm concentration exhibited 83.60 and 80.05 per cent feeding inhibition to fourth instar larvae of *Papilio demoleus* L. after 24 and 48 hours of exposure respectively.

Elanchezhiyan *et al.* (2015) reported that the methanol extract of *Tinospora crispa* (L.) Hook. F. & Thomson and *Psidium guajava* L. leaves at 500 ppm concentration recorded 100 and 98.38 per cent antifeedant activity against fourth instar larvae of *S. litura*. Thangarasu *et al.* (2015) reported that the methanol extract of *A. precatarius* leaves exhibited antifeedant activity of 67.41, 78.73 and 95.28 per cent at 100, 200 and 300 ppm concentration respectively against fourth instar larvae of *S. litura*.

Rahmat *et al.* (2015) reported the possibility of utilizing the wood waste of mahogany, *Swietenia mahagoni* (L.) Jacq. as botanical against *S. litura*. The wood waste at 3 per cent concentration exhibited 21.30 per cent antifeedant activity against third instar larvae of *S. litura*.

Thushimanan *et al.* (2016) screened three plants, *Punica granatum* L., *Cassia fistula* L. and *Erythrina variegata* L. against fourth instar larvae of *S. litura*. Maximum antifeedant activity was exhibited by the methanol extracts of *P. granatum* (83.80 %), *C. fistula* (73.20 %) and *E. variegata* (56.40 %) at 5 per cent concentration. The ethyl acetate extract of *Duranta erecta* L. leaves at 5 per cent concentration exhibited significant antifeedant activity against fourth instar larvae of *S. litura* (80.37 %) and *H. armigera* (78.18 %) (Chennaiyan *et al.*, 2016a).

Chennaiyan *et al.* (2016b) studied the effect of petroleum ether, chloroform and ethyl acetate extracts of *Barleria longiflora* (L.) F. against fourth instar larvae of *S. litura* and *H. armigera*. Ethyl acetate extract at 5 per cent concentration exhibited maximum antifeedant activity of 79.40 and 77.36 per cent against *S. litura* and *H. armigera* respectively.

2.2.2. Insecticidal Activity

Rathi and Gopalakrishnan (2005) reported the insecticidal activity of aerial parts of *Synedrella nodiflora* Gaertn. against *S. litura*. Methanol extract at 0.08 per cent concentration recorded 100 per cent mortality against fourth instar larvae of *S. litura*. Water extract was found to be least toxic with LD 50 value of 0.061 per cent, followed by benzene and chloroform extracts. The LD 50 value of water extract was about 20 times higher than its methanol extract. The phytochemical analysis of methanol extract confirmed the presence of steroids, reducing sugars, phenolic compounds, saponins and tannins.

Nathala and Dhingra (2006) reported the effect of different solvent extracts of *C. crista* seeds against third instar larvae of *H. armigera*. The methanol, hexane, ethyl acetate and butanol extracts showed significant larvicidal action with LC 50 values of 0.0236, 0.0555, 0.0762 and 0.1247 per cent respectively.

Kamaraj *et al.* (2008) reported that the methanol extracts of *Ocimum canum* L., *Rhinacanthus nasutus* (L.) Kurz and acetone extract of *Ocimum sanctum* L.

exhibited significant larvicidal action against third instar larvae of *S. litura* with LC 50 values of 36.46, 68.08 and 68.84 ppm respectively. The acetone seed extract of *S. foetida* exhibited cent per cent mortality to third instar larvae of *S. litura* with LC 50 value of 0.67 mg/cm² (Rani and Rajashekarreddy, 2009).

Baskar *et al.* (2010) reported the effect of *C. guianensis* leaf extracts on mortality of third instar larvae of *H. armigera*. Maximum larval mortality (81.67 %) was exhibited by hexane extract at 5 per cent concentration. Further fractionation of hexane extract showed that the fraction 8 at 1000 mg/kg showed 80.88 per cent larval mortality with LC 50 and LC 90 values of 413.30 and 1181.68 mg/kg respectively.

Rathi and Gopalakrishnan (2010) evaluated the insecticidal action of *Lantana wightiana* Wall. against fourth instar larvae of *S. litura* and reported that the crude methanol extract of *L. wightiana* aerial parts at 0.08 per cent concentration recorded 80 per cent mortality within twenty four hours of treatment. Deshmukhe *et al.* (2010) reported that the topical application of cold ethyl alcohol extract of *A. squamosa* seeds at 25 per cent concentration showed 61.66 per cent mortality to fourth instar larvae of *S. litura*. The extract when incorporated in diet caused 76.66 per cent mortality.

The hexane, chloroform and methanol extracts of *Ipomoea pauciflora* M. Martens & Galeotti at 4 mg/ml when incorporated in diet for 7 days showed mortality of 96.88, 93.75 and 57.29 per cent against neonate larvae of *S. frugiperda* (Elisa *et al.*, 2010).

Chauhan *et al.* (2011) reported that the ethanol extracts of *Cassia fistula* L. and methanol extracts of *Clerodendron inerme* Gaertn was found to be highly toxic to third instar larvae of *S. litura* with LD 50 values of 1.704 and 3.847 respectively. Kumar *et al.* (2011) reported the larvicidal activity (76.67 %) of acetone extract of *Catharanthus roseus* (L.) G. roots at 5 per cent concentration against third instar larvae of *S. litura*.

Rhein (1,8-dihydroxyanthraquinone-3-carboxylic acid), a compound isolated from the ethyl acetate extract of *C. fistula* flower at 1000 ppm showed significant mortality against fourth instar larvae of *H. armigera* (67.5 %) and *S. litura* (36.25 %) with LC 50 values of 606.50 and 1192.55 ppm respectively (Duraipandiyan *et al.*, 2011).

Bhagath and Kulkarni (2012) studied the larvicidal activity of three *Jatropha* species viz., *J. nana* Dalz. & Gibs., *J. gossypifolia* L. and *J. glandulifera* Roxb. against third instar larvae of *S. litura*. Maximum insecticidal action was exhibited by seed oils of *J. nana* and *J. gossypifolia* with LC 50 value of 10.92 and 10.95 respectively followed by methanol extract of *J. gossypifolia* leaves with LC 50 value of 19.75 and 27.87 respectively.

Risco *et al.* (2012) reported the pesticidal property of leaves, stems, spikes, fruits and seeds of *Piper tuberculatum* Jacq. against third instar larvae of *S. frugiperda*. The chloroform : methanol extract (2:1) from mature spikes produced maximum mortality of 90 per cent at 0.1850 mg/ μ L after 24 hours of treatment. The ethanol extract from mature spikes at 0.1850 mg/ μ L caused 100 % mortality at 72 hours after treatment. Bakavathiappan *et al.* (2012) reported the insecticidal activity of *C. procera* leaf extracts against third instar larvae of *S. litura*. Maximum larval mortality (67 %) was recorded in chloroform extract at 5 per cent concentration, with LC 50 and LC 90 values of 2.85 and 17.94 per cent respectively.

The insecticidal activity of weed plants, *P. hysterophorus* and *A. adenophora* was reported by Yogesh *et al.* (2013). The methanol extract of *P. hysterophorus* and *A. adenophora* recorded LC 50 values of 5.92 and 7.82 per cent respectively and LC 90 values of 8.14 and 8.96 per cent respectively against fifth instar larvae of *S. frugiperda* at 72 hours after exposure. Syahputra (2013) reported that the crude ethanol extract of *Barringtonia sarcostachys* (Blume) Miq. bark possessed strong lethal effect to fourth instar larvae of cabbage head caterpillar,

Crocidolomia pavonana F. with LC 50 and LC 99 values of 0.14 and 0.87 per cent respectively.

Pavunraj *et al.* (2014) reported the insecticidal action of different solvent extracts of *S. acmella* leaves against lepidopteran pests. The dichloromethane extract of *S. acmella* leaves at 5 per cent concentration recorded 48.88, 44.88 and 75.11 per cent larval mortality against third instar larvae of *H. armigera*, *S. litura* and *E. vitella* respectively, followed by acetone, dimethylsulfoxide and aqueous extracts. The crude methanol extract of *Trichilia silvatica* C. DC. leaves, bark and flowers showed 24, 32 and 54 per cent viability of second instar larvae of *S. frugiperda* respectively at 1 per cent concentration (Freitas *et al.*, 2014).

Pavela *et al.* (2014) reported the acute and chronic toxicity of *Ailanthus altissima* Swingle, commonly known as tree of heaven, against third instar larvae of *S. littoralis*. Methanol extract fraction 1 and 2 at 200 µg/cm² exhibited significant chronic toxicity of 83.3 and 93.3 per cent respectively. Fraction 5 and 6 of methanol extract exhibited acute toxicity of 81.7 and 68.5 per cent respectively at 100 µg/larvae. Phytochemical study of methanol extract fraction 5 revealed the presence of five major quassinoids *viz.*, Ailanthone, Chaparrinone, Glaucarubinone, 13(18)-Dehydroglaucarubinone and Shinjulactone H.

Thangarasu *et al.* (2015) studied the pesticidal effect of hexane, diethyl ether, dichloromethane, ethyl acetate and methanol extracts of *A. precatorius* leaves against fourth instar larvae of *S. litura*. Highest larvicidal activity of 100 per cent was recorded in methanol extract at 500 ppm concentration with LC 50 value of 225.76 ppm. Least larvicidal activity of 18.8 per cent was recorded in hexane extract at 100 ppm concentration with LC 50 value of 255.91 ppm.

The topical application of ethanol crude extract of *Clerodendron inerme* (L.) Gaertn and *Clerodendron viscosum* L. at 2.5 per cent concentration exhibited significant larvicidal action of 76.66 and 73.33 per cent against third instar larvae of *S. litura* respectively (Jadhav *et al.*, 2016).

2.2.3. Effect on Adult Emergence

Jeyasankar *et al.* (2010) reported the growth inhibitory activities of ethyl acetate extract of *S. lineare* leaves against fourth instar larvae of *S. litura*. Highest larval deformity (9.4 %) was observed in sixth fraction of ethyl acetate extract at 500 ppm concentration. Ethyl acetate extract fraction 3 at 500 ppm concentration showed 11.8 per cent pupal deformity and 15.5 per cent adult deformity.

The bioactivity of leaf extracts of *A. tagala* against *S. litura* was reported by Baskar *et al.* (2011a). Maximum mortality of 68.66 per cent (40.66 % larval mortality and 28 % pupal mortality) was recorded in ethyl acetate leaf extract at 5 per cent concentration. Prolonged larval - pupal duration of 12.04-13.08 days was also observed. Sumathy and Sanjayan (2011) reported that plumbagin, a secondary metabolite of *Plumbago zeylanica* L. at 500 ppm significantly reduced the weight gain of the third instar larva to 0.44 mg, compared to control (1.25 mg/larva).

Lethal and sub lethal effects of saponin extracted from *Passiflora alata* Dryander on *S. frugiperda* was studied by D’Incao *et al.* (2012). Saponin extract at 20000 ppm exhibited cumulative mortality of 58 % and deformity of 68.3 per cent. Deformities observed were larvae with abdominal distension, larvae with necrosis in integument, larvae with incomplete molting, pupae with globular evagination, pupae with failure in sclerotization, larval-pupal intermediate and adult with unfolded wings.

Lingampally *et al.* (2012) reported the effect of ‘Betulinic acid’, a terpenoid compound derived from the bark of *Ziziphus jujube* Mill. against *Tribolium confusum* Duval. Fifth instar larvae, treated with betulinic acid at 1µg/µl produced 33.3 per cent larval-pupal intermediates. Sixth instar larvae (30%) treated with betulinic acid at 1µg/µl failed to pupate. Ray *et al.* (2012) reported the growth inhibitory effect of *Thevetia nerifolia* Juss. against third instar larvae of *S. litura*. The methanol extract of leaves of *T. nerifolia* recorded 53.8 % larval mortality and exhibited 29.6 per cent pupation and 22.3 per cent adult emergence. The methanol extract when subjected to

sub-fractionation showed that the chloroform extract recorded maximum bioactivity with 27.5-61.5 per cent larval mortality, 28.4-60.2 per cent pupation and 19.8-52.8 per cent adult emergence.

Yogesh *et al.* (2013) reported that the crude methanol extracts of *P. hysterophorus* and *A. adenophora* at 10 per cent concentration exhibited significant larval, pupal and adult deformities like larva with abdominal distension, necrosis in integument and incomplete molting, pupae with globular evagination and adults with rolled wings against *S. frugiperda*.

The growth regulatory activity of *S. nux-vomica* and *S. anacardium* seed extracts was reported by Sivaraman *et al.* (2014a). Methanol extracts of *S. anacardium* seeds showed 28.57 per cent pupicidal activity at 2 per cent concentration. The hexane extract of *S. nux-vomica* at 2 per cent concentration recorded 31.43 per cent adult deformity. Sivaraman *et al.* (2014b) reported that the hexane extract of *Cleome viscosa* L. and *S. alba* at 2 per cent concentration exhibited 33.93 and 32.86 per cent adult deformities respectively. The deformities include relatively poor body size, highly curled wings and undergrown wings.

Magdum and Gupta (2014) reported the effect of tetra hydroxyl-p-benzoquinone on metamorphosis of fifth instar larvae of *S. litura*. The compound produced 89 per cent mortality, 10 per cent pupal abnormality and 23 per cent emergence of female moths at 10 µg concentration. Delayed pupal period of 18-25 days was also observed. Ghaly *et al.* (2014) reported that the methanol extract of *K. beharensis* and *Kalanchoe longiflora* leaves exhibited 73.3 and 93.3 per cent inhibition for adult emergence of *S. littoralis* respectively.

Bhatt *et al.* (2014) reported the bioactivity of medicinal plants against first and third instar larvae of *S. litura*. The methanol extract of *Dendrophloe falcata* (L. F.) Ettingsh leaves at 5 mg/ml concentration exhibited 98.58 per cent reduction in weight gain of first instar larvae. The methanol extracts of *Lantana camara* L. leaf and fruit caused 99.43 per cent reduction in weight gain of third instar larvae.

Methanol extracts of *L. camara* leaf and fruit at 10 mg/ml concentration showed 100 per cent mortality to first and third instar larvae of *S. litura*.

Pandy and Summarwar (2015) reported that the acetone extract of *O. sanctum* leaves at 0.5 per cent concentration exhibited significant larval (11.66 %), pre-pupal (6.66 %), pupal (2.86 %) and adult (2.16 %) mortality against *S. litura*.

2.2.4. Effect on Fecundity and Egg Hatching

Pavunraj *et al.* (2006) reported that the hexane extract of *Excoecaria agallocha* L. leaf at 5 per cent concentration showed 83.71 per cent oviposition deterrent and 65 per cent ovicidal activities against *S. litura*. The acetone extract of *C. roseus* roots at 5 per cent concentration exhibited reduced fecundity of 381 numbers compared to control (510.8 numbers) and 60.83 per cent egg hatching in *S. litura* (Kumar *et al.*, 2011).

The ovicidal activity of botanical oil formulations against lepidopteran pests was reported by Packiam *et al.* (2012). The highest ovicidal activity of 76.74 and 69.36 per cent was shown by PONNEEM at 20 µl/L against *H. armigera* and *S. litura* respectively. Pongam oil at 20 µl/L exhibited lower percentage egg mortality of 31.34 and 24.76 per cent against *H. armigera* and *S. litura* respectively.

Gokulakrishnan *et al.* (2012) reported the oviposition deterrent activity of plant oils against *E. vitella*, *H. armigera* and *S. litura*. Lime (0.15 ml), Calamus (0.45 ml), Lemon (0.15 ml) and Tagetus (0.15 ml) oils at 100 ppm concentration exhibited repellency of 76.55, 79.90 and 84.75 % against *E. vitella*, *S. litura* and *H. armigera* respectively.

Baskar *et al.* (2012a) studied the ovicidal action of hexane, ethyl acetate and chloroform extracts of *Atalantia monophylla* (L.) Correa against *S. litura*. Highest ovicidal activity of 61.94 per cent was recorded in hexane leaf extract at 5 per cent concentration with LC 50 value of 3.06 per cent. Further fractionation of hexane extract showed that 75.61 per cent egg mortality was recorded in fraction 9 at 1000

mg/kg concentration with LC 50 and LC 90 values of 318.65 and 1473.31 mg/kg respectively.

Jeyasankar *et al.* (2013) reported the oviposition deterrent and ovicidal activities of *S. lineare* leaves against *S. litura*. Maximum oviposition deterrent activity (49.97 %) and ovicidal activity (45.22 %) was recorded in ethyl acetate crude extract at 5 per cent concentration. Further fractionation of bioactive ethyl acetate extract showed that fraction 6 and 3 at 0.1 per cent concentration recorded maximum oviposition deterrent (60.41 %) and ovicidal (66.74 %) activities respectively. The compounds A and B isolated from fractions 3 and 6 in the ratio of hexane:ethyl acetate (10:90 and 30:70) exhibited significant oviposition deterrent (73.25%) and ovicidal (69.05 %) activities respectively at 100 ppm concentration.

Arivoli and Tennyson (2013) screened hexane, diethyl ether, dichloromethane and ethyl acetate extract of twenty five plants at 1 per cent concentration for ovicidal activity against *S. litura*. The hexane extract of *Cleistanthus collinus* (Roxb) Benth at 0.05 per cent exhibited maximum ovicidal activity of 85.16 per cent, followed by diethyl ether extract of *M. koeingii* (83.60 %) and ethyl acetate extract of *Aegle marmelos* (L.) Corr (76.14 %).

The ovicidal action of *Couroupita guianensis* Aubl. against *S. litura* was reported by Baskar *et al.* (2014). The hexane extract of *C. guianensis* leaves at 50 mg/ml exhibited 67.33 per cent ovicidal activity, while 47 and 42 per cent egg mortality was recorded in chloroform and ethyl acetate extracts respectively at 50 mg/ml concentration. Among the 8 fractions obtained from hexane extract, fraction 8 recorded maximum ovicidal activity of 30.46 per cent at 125 g/ml with LC 50 and LC 90 values of 384.43 and 1576.55 g/ml respectively.

Elanchezhiyan *et al.* (2015) reported the ovicidal activity of *T. crispa* and *P. guajava* against fourth instar larvae of *S. litura*. Methanol extracts of *T. crispa* and *P. guajava* leaves exhibited 100 per cent egg mortality at 160 and 200 ppm concentrations respectively. Thangarasu *et al.* (2015) reported that the methanol

extract of *A. precatorius* leaves exhibited 100 per cent ovicidal and oviposition deterrent activity against *S. litura* at 300 ppm concentration.

2.3. EVALUATION OF PLANT EXTRACTS UNDER *in vivo* CONDITION

2.3.1. Plant Extracts

A study conducted by Govindachari *et al.* (2001) reported that four quassinoids, indaquassin C, samaderins C, B and A isolated from the seeds and bark of *S. indica* exhibited significant antifeedant and growth regulatory activities against the tobacco caterpillar, *S. litura*. Indaquassin C was found to be the most effective antifeedant (62.5 %) at 5 µg/cm², while Samaderin C increased pupal duration (9.7 days) and pupal mortality (54.1 %) at 0.5 µg/cm².

Methanolic extract of *Q. indica* flower exhibited acetylcholinesterase inhibition activity in electric eel (Wetwitayaklung *et al.*, 2007). The toxic effects of *Q. indica* on scale insect was reported by Song *et al.* (2013), who recorded 95.70 per cent nymphal mortality in black pine bast scale, *Matsucoccus thunbergianae* Miller and Park. They observed that *Q. indica* was equally effective to the insecticide, fenitrothion 50 per cent EC against *M. thunbergianae* in field trials. This was in accordance with the findings of Song *et al.* (2014). They reported that the methanol extract obtained from the fruits of *Q. indica* possessed significant insecticidal activity against four Coccoidea species (*Eriococcus lagerstroemiae* Kuwana, *Ceroplastes japonicas* Green., *Crisicoccus pini* Kuwana and *Planococcus citri* Risso).

The field evaluation of plant extracts *viz.*, neem seed extract (2.5%), turmeric extract (5%), henge extract (1.25%), garlic extract (5%) and an insecticide, emamectin benzoate (0.07%) against *H. armigera* on tomato was done by Shah *et al.* (2013). The minimum percentage fruit damage of 11.30 and 10.10 per cent was recorded in neem seed extract (2.5 %) and emamectin benzoate (0.07 %) respectively.

Mishra and Singh (2014) studied the effect of plant extracts against *Plutella xylostella* L. on cabbage. A combination of dhatura, neem and mehandi leaf extracts at one per cent concentration recorded a mean population of 21.02 larvae per 5 plants ten days after treatment, while 45.17 larvae per 5 plants was observed in control.

Akbar *et al.* (2014) compared the efficacy of plant extracts with chemical pesticides against diamond moth, *P. xylostella* on cauliflower. Larval mortality of 90.8, 75.8, 60.0, 50.6 and 36.8 per cent was recorded in emamectin benzoate (50 ml/10 L), methomyl (50 ml/10 L), lufenuron (200 ml/10 L), *Melia azedarach* L. (500 g/10 L) and *A. indica* (500 g/10 L) respectively under laboratory condition. Further field evaluation revealed that significant reduction in mean population of larvae with 2.2 and 3.1 larvae per plant recorded in *A. indica* (500 g/10 L) and *M. azedarach* (500 g/10 L) 7 days after spraying. Minimum mean population of larvae (0.5 larva/plant) was observed in emamectin benzoate (50 ml/10 L) treated plants.

Rahman *et al.* (2014) evaluated the effectiveness of plant extracts in comparison with chemical pesticides against *H. armigera* in Tomato. Minimum fruit damage was recorded in neem seed kernel extract 5 per cent (27.15 %) and tobacco leaf extract 2 per cent (27.71 %) compared to cypermethrin 10 EC 0.01 per cent (28.87 %).

2.3.2. Biocontrol Agent

2.3.2.1. *Beauveria bassiana* (Bals.-Criv.) Vuill.

Moorthi *et al.* (2011) studied the effect of local isolates of *B. bassiana* against third instar larvae of *S. litura*. The isolates, Bb 02, Bb 09 and Bb 10 at 10^8 spore/ml recorded 66.67, 73.33 and 80 per cent larval mortality with LC 50 values of 2.1×10^6 , 3.6×10^7 and 1.2×10^7 conidia/ml and LT 50 values of 4.8, 4.8 and 4.0 days respectively.

Baskar *et al.* (2012b) reported that *B. bassiana* (Bb 10) isolate exhibited maximum mortality of 68.06 per cent against third instar larvae of *S. litura* at 10^8 spore/ml five days after treatment. It also caused minimum pupal weight of 183 mg, 22.91 per cent adult emergence and 100 per cent abnormalities. *B. bassiana* strains viz., BNBCRC, BPMC, B14841, B16041 and B14532 at 10^8 conidia/ml exhibited significant mortality of 100, 100, 90, 86.67 and 83.33 per cent respectively against third instar larvae of *S. litura* at 5 days after treatment (Petlamul and Prasertsan, 2012).

Gupta and Kumar (2014) reported the toxic effect of *B. bassiana* against third instar larvae of *S. litura*. The spore suspension of *B. bassiana* at 0.2×10^8 spore/ml exhibited maximum mortality of 80 per cent at 15 days after treatment. The larval mortality enhanced as the concentration of spores increased.

Ummidi and Vadlamani (2014) evaluated the oil formulations of *B. bassiana* isolates against third instar larvae of *S. litura*. The oil formulations of *B. bassiana* isolate ARSEF 654 (B55) with almond oil, olive oil, gingelly oil and castor oil at 2 per cent concentration recorded 94.3, 89.4, 93.8 and 92.5 % mortality with LT 50 values of 4.69, 5.63, 5.08 and 4.69 days respectively. The oil formulations of *B. bassiana* isolate ARSEF 1725 (B51) with almond oil, olive oil, gingelly oil and castor oil at 2 per cent concentration showed 93.1, 88.2, 91.4 and 90.3 per cent mortality with LT 50 values of 4.95, 5.72, 5.18 and 4.89 days respectively.

2.3.3. Chemical Pesticide

2.3.3.1. Quinalphos 25 EC 0.05 %

Basavaraju *et al.* (2010) evaluated the effectiveness of the conventional pesticide, quinalphos 25 EC (Organophosphate) as poison bait in potato against *S. litura*. He reported that quinalphos 2ml/L when given as bait could cause 100 per cent mortality of larvae one week after the first spray.

A study conducted by Saini *et al.* (2010) on the efficacy of different pesticides to manage *S. litura* under open field condition revealed the potential of quinalphos as an effective insecticide against *S. litura*. The chemical at 2ml/L recorded a mean population of 3.66 larvae per plant at 10 days after spraying.

Suganthy and Sakthivel (2013) conducted studies on the efficacy of quinalphos 25 EC in managing *S. litura* under open field condition and reported that quinalphos 2 ml/L could cause cent per cent mortality one week after the first spray. They also compared quinalphos with two other botanicals, Azadirachtin 1 per cent and Pungam oil 3 per cent and the mean pest population recorded was 0.90 and 1.63 larvae/plant respectively one week after the first spray.

Materials and Methods

3. MATERIALS AND METHODS

The present study entitled “Bioefficacy of *Quisqualis indica* L. and *Samadera indica* Gaetrn. against tobacco caterpillar, *Spodoptera litura* Fabricius (Lepidoptera: Noctuidae) in polyhouse condition” was carried out in the Department of Entomology, College of Agriculture, Vellayani during 2014-16. The study was conducted to assess the bioactivity of *Q. indica* flower and *S. indica* leaf against *S. litura* in terms of antifeedant effect, insecticidal effect, effect on adult emergence, fecundity and egg hatching.

3.1. BIOEFFICACY OF DIFFERENT AQUEOUS PLANT EXTRACTS AGAINST *S. litura*

3.1.1. Collection of Plant Materials

Q. indica flowers and *S. indica* leaves were selected to check the bioefficacy of their extracts against *S. litura*. Fresh flowers of *Q. indica* at full bloom were collected during morning hours from in and around the Instructional farm, College of Agriculture, Vellayani, Thiruvananthapuram district, Kerala. Fresh mature leaves of *S. indica* were collected from the forest areas of Kulathuppuzha, Kollam district, Kerala. *Q. indica* and *S. indica* were identified by Dr.Valsaladevi from Department of Botany, Kerala University, Karyavattom and the voucher specimens (KUBH – 6010 and KUBH – 6011 respectively) were deposited at the herbarium of the Institute.

3.1.2. Preparation of Aqueous Extracts

The fresh plant materials collected were separately washed with clean water thoroughly and chopped into small pieces. 100 g of fresh chopped plant materials were macerated in an electric grinder and immersed in 100 ml of distilled water taken in a reagent bottle. The bottle was shaken well and kept overnight at room temperature. The mixture was then filtered with cheese cloth and Whatman No.1 filter paper in succession. The volume was made upto 100 ml to prepare the stock extract and stored in refrigerator at 4°C till usage. Different concentrations *viz.* 5, 10

and 15 per cent were prepared from the stock extract via dilution using distilled water.

3.1.3. Preparation of Neem Seed Kernel Extract

Neem seeds were collected and the kernels were separated out. The seed kernel was ground to a coarse powder using pestle and motor. 50 g powder was taken in a muslin cloth bag and kept soaked in 500 ml of water overnight. The cloth bag was squeezed well until the extract turned light brown color. This was further made upto 1 litre to get 5 per cent neem seed kernel extract.

3.1.4. Rearing of Test Insect

The tobacco caterpillar, *S. litura*, was the test insect used for evaluating the bioefficacy of plant extracts. The insect culture was maintained at Insect rearing lab, Dept. of Entomology, College of Agriculture, Vellayani under standard conditions of temperature ($28\pm 2^{\circ}\text{C}$) and relative humidity ($70\pm 5^{\circ}\text{C}$).

Egg masses of *S. litura* were collected from Instructional farm, College of Agriculture, Vellayani. The eggs were surface sterilized with 0.02 per cent sodium hypochlorite solution, dried and allowed to hatch. The neonate larvae were reared in plastic troughs of 30 cm diameter. Fresh castor leaves, after sterilization, were provided as feed on alternate days. Hygienic condition was maintained by regular cleaning of rearing troughs. Sterilized soil was provided for pupation. After pupation, the pupae were collected from the soil and placed inside a rearing bottle of 10cm diameter and 20 cm height for emergence of adult moths. The bottle was provided with chart paper folded in step like pattern to facilitate mating and egg laying of *S. litura*. Adults were maintained by providing 5 per cent honey solution. The eggs laid in chart paper were collected separately and kept in plastic troughs with castor leaves for hatching. The laboratory reared larvae were used for different experiments (Plate 1A, B, C, D and E).



A. Egg masses of *S. litura* on amaranthus leaves



B. Rearing of *S. litura* larvae on castor leaves

Plate 1. Rearing of *S. litura*



C. Pupation of *S. litura* in sterilized soil



D. Oviposition chamber of adult *S. litura*



E. Egg masses of *S. litura* collected on chart paper

Plate 1. Rearing of *S. litura*

3.1.5. Evaluation of Bioactivities

Aqueous extracts of *Q. indica* flower and *S. indica* leaf were prepared as mentioned in 3.1.2. Second instar caterpillars of *S. litura* reared as described in para 3.1.4. were used as test insect. *In vitro* screening experiments were conducted to assess the bioactivity of aqueous plant extracts *viz.*, antifeedant activity, insecticidal activity, effect on adult emergence, effect on fecundity and egg hatching as detailed below.

Design : CRD Treatments : 8 Replication : 3
T1 : *Q. indica* flower extract (5%)
T2 : *Q. indica* flower extract (10%)
T3 : *Q. indica* flower extract (15%)
T4 : *S. indica* leaf extract (5%)
T5 : *S. indica* leaf extract (10%)
T6 : *S. indica* leaf extract (15%)
T7 : Check

T8 : Untreated

3.1.5.1. Antifeedant Effect

Antifeedant activity of plant extracts were tested by no-choice method (Bentley *et al.*, 1984). Fresh castor leaves of uniform thickness were taken and dipped in plant extracts for one minute and air dried. Each leaf was placed in rearing boxes of 15 cm height and 10 cm diameter. Each leaf petiole was wrapped with cotton and kept in water taken in a vial to avoid early drying of leaves. One 3rd instar larvae pre starved for four hours was exposed to leaf in each rearing box. Three such replications were maintained for each treatment. Leaves treated with distilled water served as untreated. Neem seed kernel extract 5 per cent was used as check. The larvae were allowed to feed for 72 hours. The leaf area consumed at 24, 48 and 72 hours after treatment was observed and percentage mean leaf protection was calculated according to the formula given by Baskar *et al.* (2011b).

Percentage mean leaf protection

$$= \frac{\text{Leaf area consumed in control} - \text{leaf area consumed in treated leaf}}{\text{Leaf area consumed in control}} \times 100$$

3.1.5.2. Insecticidal Effect

The plant extracts were tested against 2nd instar larvae of *S. litura* for mortality via spraying method.

3.1.5.2.1. Spraying Method

One millilitre of plant extracts were sprayed separately with different dosages on second instar larvae of *S. litura* (20 no's) using TLC sprayer. Larvae sprayed with one milliliter of the distilled water alone were maintained as untreated. Quinalphos 25 EC (0.05%) was used as check. Each treatment was replicated thrice. The sprayed larvae were transferred to rearing bottle and fed with sufficient quantity of fresh castor leaves. Observations were recorded at 24 h interval for 3 days after treatment. Percentage mortality was calculated using the following formula

$$\text{Percentage larval mortality} = \frac{\text{Number of larvae dead}}{\text{Total number of larvae treated}} \times 100$$

3.1.5.3. Inhibition of Adult Emergence

Aqueous extracts of both plant materials were studied for inhibition effect on adult emergence. The treated larvae were provided with fresh untreated castor leaves to evaluate inhibition effect on adult emergence. Observations were taken on number of deformed larvae, pupae and adults, mortality of larvae, pupae and adults, time taken for pupation, pupal duration, pupal weight and adult longevity.

3.1.5.3.1. Effect on Larvae

Percentage of larval mortality and deformity was calculated from the total number of larvae treated.

3.1.5.3.2. Effect on Pupae

The survived larvae after treatment were continuously supplied with fresh castor leaves as feed until they became pupae. Pupal mortality was calculated by subtracting the number of emerging adults from the total number of pupae.

3.1.5.3.3. Effect on Adults

Percentage of abnormalities in adults was calculated from the number of emerged adults.

3.1.5.4. Effect on Fecundity and Egg Hatching

The emerged adults from the larvae treated with aqueous plant extracts were transferred to rearing bottle separately. Each treated female was allowed to mate. The females were allowed to lay eggs and egg masses were collected separately (Plate 2A, B and C). Five replications were maintained. Observations were recorded on the number of eggs laid by treated females and the number of eggs hatched from the total eggs laid.

3.2. EVALUATION OF BIOEFFICACY OF PLANT EXTRACTS IN DIFFERENT SOLVENTS AGAINST *S. litura*

3.2.1. *Q. indica* Flower

3.2.1.1. Preparation of Solvent Extracts

Fresh flowers of *Q. indica* collected as described in 3.1.1 were shade dried under room temperature ($28\pm 2^{\circ}\text{C}$). When they attained a constant weight, the plant materials were ground finely in an electric grinder. The powdered plant materials were kept in refrigerator till usage.



A. Egg mass of *S. litura*



B. Egg mass of *S. litura* two hours prior to hatching



C. Larvae emerging from the egg mass of *S. litura*

Plate 2. Different stages of egg hatching of *S. litura*

The powdered plant materials were subjected to extraction of bioactive components present within it. Two different solvent extraction methods *viz.*, cold extraction and soxhlet extraction were followed to yield bioactive components from plant materials. Ethyl acetate and methanol were used as solvents in the extraction process (Plate 3A, B, C and D).

3.2.1.1.1. Cold Extraction

Powdered samples of dried plant material (18 g) were separately immersed in 250 ml of the respective solvents *viz.*, ethyl acetate and methanol in reagent bottles (500 ml). These bottles were shaken in a reciprocating shaker at room temperature for 72 hours (Wetwitayaklung *et al.*, 2007). Solutions obtained were filtered through cheese cloth and Whatman No.1 filter paper in succession. The filtrate was concentrated to air dryness. The extract was further made into different concentrations *viz.*, 1.25%, 2.5% and 5% using distilled water. Tween 20 (Polysorbate) at 0.05% was used as emulsifier.

3.2.1.1.2. Soxhlet Extraction

100 g of powdered plant materials were packed in soxhlet apparatus and extracted with ethyl acetate and methanol individually (Kombiah and Sahayaraj, 2012). These solutions were then filtered through Whatman No.1 filter paper and the filtrate was concentrated using rotary evaporator. The extract was further concentrated and made into different concentrations as described in 3.2.1.1.1

3.2.1.2 Evaluation of Bioactivities

An *in vitro* study was conducted to evaluate various bioactivities *viz.*, antifeedant effect, insecticidal effect, effect on adult emergence, fecundity and egg hatchability by the solvent extracts of *Q. indica* flower against *S. litura*.

Design : CRD Replications : 3 Treatments : 10

The following were the treatments used for the study.

- T1 : Ethyl acetate extract (1.25%)
- T2 : Ethyl acetate extract (2.5%)
- T3 : Ethyl acetate extract (5%)



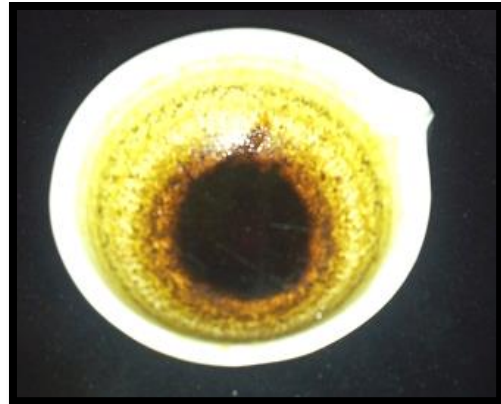
A. Fresh flowers of *Q. indica*



B. Shade drying of fresh flowers of *Q. indica*



C. Powder of dried flowers of *Q. indica*



D. Solvent extract of *Q. indica* flower

Plate 3. Solvent extraction from *Q. indica* flower

T4 : Ethyl acetate alone (control)
T5 : Methanol extract (1.25%)
T6 : Methanol extract (2.5%)
T7 : Methanol extract (5%)
T8 : Methanol alone (Control)
T9 : Check

T10: Distilled water (Untreated)

3.2.1.2.1. Antifeedant Effect

3.2.1.2.1.1. Cold Extracts of *Q. indica* Flower

The antifeedant effect of cold methanol and ethyl acetate extracts of *Q. indica* flower at 1.25, 2.5 and 5 per cent concentrations was evaluated as mentioned in 3.1.5.1. Methanol and ethyl acetate alone served as control, while distilled water served as untreated. Neem seed kernel extract 5% was used as check. Observations were taken on leaf area consumed at 24, 48 and 72 hours after treatment and percentage mean leaf protection was calculated as mentioned in 3.1.5.1.

3.2.1.2.1.2. Soxhlet Extracts of *Q. indica* Flower

The antifeedant effect of soxhlet extracts (methanol and ethyl acetate) of *Q. indica* flower at 1.25, 2.5 and 5 per cent concentrations was evaluated as mentioned in 3.2.1.2.1.1.

3.2.1.2.2. Insecticidal Effect

3.2.1.2.2.1. Cold Extracts of *Q. indica* Flower

Cold methanol and ethyl acetate extracts of *Q. indica* flower at 1.25, 2.5 and 5 per cent concentrations were evaluated for their insecticidal activity against 2nd instar larvae of *S. litura* via spraying, leaf dip and dry film methods.

3.2.1.2.2.1.1. Spraying Method

The insecticidal activity of cold extracts of *Q. indica* flower was assessed through spraying method as mentioned in 3.1.5.2.1. Methanol and ethyl acetate alone

served as control, while distilled water served as untreated. Quinalphos 0.05 % was used as check.

3.2.1.2.2.1.2. Leaf Dip Method

The fresh castor leaves of uniform thickness were cut into leaf discs of 7 cm diameter and treated with plant extracts of different concentrations separately. Both untreated and control was maintained by treating leaf discs with distilled water and solvents alone. Quinalphos 0.05% was kept as check. The treated leaf discs were kept over wet padding of cotton in a petri dish and second instar larvae of *S. litura* (20 no's) were allowed to feed on each treated leaf disc after 24 h of treatment. The diet was changed every 24 hour. Three replicates were maintained for each treatment. Larval mortality was recorded at 24 h interval for 3 days after treatment. Percentage mortality was calculated using the formula as mentioned in 3.1.5.2.1 (Bakavathiappan *et al.*, 2012).

3.2.1.2.2.1.3. Dry Film Method

One millilitre of the extract was poured in each rimless glass test tube and rotated gently. The angles of the test tube was adjusted in such a way that the extract covered three fourth of the inner surface of the test tube. The process was continued till the extract dried up leaving behind a uniform film of extract on the inner surface of glass tube. Second instar larvae of *S. litura* (20 no's) were kept in the glass tube and the mouth of the glass tube was plugged with cotton. The larvae were allowed to remain in contact with the dry film for 4 h. Both untreated and control was maintained using distilled water and solvents alone respectively. Quinalphos (0.05%) was kept as check. Each treatment was replicated thrice. Larval mortality was recorded after 4 h. Moribund larvae were also counted as dead one (Ray *et al.*, 2012). Percentage mortality was calculated using the formula as mentioned in 3.1.5.2.1.

3.2.1.2.2.2. Soxhlet Extracts of *Q. indica* Flower

Methanol and ethyl acetate extracts (1.25, 2.5 and 5 per cent concentrations) of *Q. indica* flower obtained via soxhlet extraction method were evaluated for insecticidal effect against 2nd instar larvae of *S. litura* via spraying and leaf dip methods.

3.2.1.2.2.2.1. Spraying Method

The insecticidal effect of soxhlet extracts of *Q. indica* flower through spraying method was evaluated as mentioned in 3.2.1.2.2.1.1.

3.2.1.2.2.2.2. Leaf Dip Method

Soxhlet extract of *Q. indica* flower was evaluated for insecticidal effect through leaf dip method as mentioned in 3.2.1.2.2.1.2.

3.2.1.2.2.3. Comparison of Method of Extraction

Effective method of extraction of plant materials was determined by comparing the results of antifeedant and insecticidal bioassay of *Q. indica* flower. Cold extraction method, which was found to be superior over soxhlet extraction method, was selected for extracting plant materials to carry out further studies.

3.2.1.2.3. Effect on Adult Emergence

The cold extracts of *Q. indica* flower were evaluated for adult emergence inhibition effect on *S. litura* as mentioned in 3.1.5.3. Methanol and ethyl acetate alone served as control, while distilled water served as untreated. Neem seed kernel extract 5 per cent was used as check.

3.2.1.2.4. Effect on Fecundity and Egg Hatching

Effect of cold extracts of *Q. indica* flower on fecundity and egg hatchability of *S. litura* was evaluated as mentioned in 3.1.5.4. Methanol and ethyl acetate alone

served as control, while distilled water served as untreated. Neem seed kernel extract 5% was used as check.

3.2.2. *S. indica* Leaf

3.2.2.1. Preparation of Solvent Extracts

The cold and soxhlet extracts of *S. indica* leaf were prepared as mentioned in 3.2.1.1 (Plate 4A, B, C and D).

3.2.2.2. Evaluation of Bioactivities

Evaluation of various bioactivities such as antifeedant effect, insecticidal effect, effect on adult emergence, fecundity and egg hatchability exhibited by the solvent extracts of *S. indica* leaf against *S. litura* was conducted *in vitro* as mentioned in 3.2.1.2.

Design : CRD Replications : 3 Treatments : 10

The following were the treatments used for the study.

- T1 : Ethyl acetate extract (1.25%)
- T2 : Ethyl acetate extract (2.5%)
- T3 : Ethyl acetate extract (5%)
- T4 : Ethyl acetate alone (control)
- T5 : Methanol extract (1.25%)
- T6 : Methanol extract (2.5%)
- T7 : Methanol extract (5%)
- T8 : Methanol alone (Control)
- T9 : Check
- T10: Distilled water (Untreated)

3.2.3. Comparison of Plant Parts

The cold extracts of *Q. indica* flower and *S. indica* leaf were evaluated for their potential bioactivity by comparing the results of antifeedant and insecticidal bioassay.



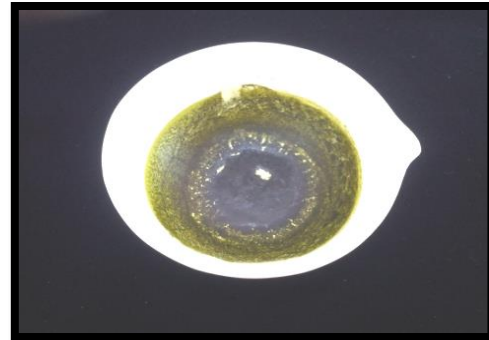
A. Fresh leaves of *S. indica*



B. Dried leaves of *S. indica*



C. Powder of dried leaves of *S. indica*



D. Solvent extract of *S. indica* leaf

Plate 4. Solvent extraction from *S. indica* leaf

3.3. TESTING THE POTENTIAL OF THE ISOLATED PLANT EXTRACTS UNDER POLYHOUSE CONDITION

The effective treatments identified for *Q. indica* flower extract and *S. indica* leaf extract were compared with standard insecticide, Quinalphos 0.05 % and recommended biocontrol agent, *Beauveria bassiana* (Bals. - Criv.) Vuill. (Bb 5) 20 g/L via pot culture experiment in polyhouse. The pot culture experiment was conducted in the polyhouse of College of Agriculture, Vellayani. The cowpea variety Vellayani jyothika was used as test plant.

3.3.1. Raising of Cowpea

Cowpea seeds, inoculated with rhizobium, were sown in pots of 30x25 cm dimension, filled uniformly with 1:2:1 potting mixture (sand : soil : cow dung). The crop was maintained following the KAU package of practice recommendations (2011) during February to May (Plate 5).

3.3.2. Preparation of Plant Extracts

The cold methanol extracts of *Q. indica* flower and *S. indica* leaf were prepared as mentioned in 3.2.1.1.1.

3.3.3. *B. bassiana*

The effect of talc based product obtained from the biocontrol agent *B. bassiana* isolate, Bb 5 at 20 g/L was evaluated on *S. litura*.

3.3.4. Quinalphos 25 EC

The insecticidal effect of quinalphos 25 EC 0.05 per cent (Ekalux) was evaluated against *S. litura*.



Plate 5. Experimental plot



Plate 6. Infestation of *S. litura* larvae on cowpea

3.3.5. Evaluation of Bioefficacy of Selected Botanicals against *S. litura* under Polyhouse Condition

Evaluation of selected botanicals was done in cowpea plants, infested with *S. litura*, maintained under polyhouse condition. The details of the experiment are as follows.

Design : CRD Replications : 4 Treatments : 5

The following were the treatments used for the study

T1 & T2 : Effective treatments from 3.2.3
T3 : Quinalphos 25 EC (0.05%)
T4 : *B. bassiana* (20g/l)
T5 : Control

3.3.5.1. Mortality of Larvae

The bio efficacy study was conducted in cowpea plants during March 2016. As part of the study, 25 second instar larvae of *S. litura* were released on a plant on the previous day of treatment. Pre count of larvae was taken prior to giving treatments. The treatments were sprayed on to the plants covering the entire plant. *B. bassiana* (20g/L) and quinalphos 25 EC (0.05%) were also sprayed on to separate plants to compare their efficacy with selected plant extracts. Sterile water served as untreated. Treatments were given at early morning. Observations on the post count of larvae on plants were recorded at 24 h interval till death or pupation of the larvae. Percentage larval mortality was calculated by the formula as mentioned below.

$$\text{Percentage larval mortality} = \frac{\text{Pre count} - \text{Post count}}{\text{Pre count}} \times 100$$

3.3.5.2. Leaf Area Damage

The cowpea plants treated as mentioned in 3.3.5.1. were evaluated for per cent leaf area damage caused by 2nd instar larvae of *S. litura* (Plate 6). The leaf area damage was estimated at 48 hour interval for 7 days after treatment by recording observations from six leaves (two each from upper, middle and lower strata) per plant. Area of leaves was taken by plotting it in a graph paper and estimated in cm². The leaf area damaged was calculated by the formula as mentioned below.

$$\text{Per cent leaf area damaged} = \frac{\text{Total leaf area} - \text{Leaf area left undamaged}}{\text{Total leaf area}} \times 100$$

3.4. STATISTICAL ANALYSIS

The observations recorded from laboratory and pot culture experiments were converted into data and were analyzed using one way analysis of variance after subjected to angular and square root transformations respectively (Panse and Sukhatme, 1967).

Results

4. RESULTS

The results on bioefficacy studies of extracts of *Quisqualis indica* L. flower and *Samadera indica* Gaetrn leaf against feeding behaviour and biology of *Spodoptera litura* Fabricius are presented below.

Aqueous and solvent extracts of *Q. indica* flower and *S. indica* leaf were tested against laboratory reared larvae of *S. litura*. Pot culture studies were conducted to confirm the potential of selected plant extracts under polyhouse condition.

4.1. BIOEFFICACY OF AQUEOUS EXTRACTS ON *S. litura*

The aqueous extracts of *Q. indica* flower and *S. indica* leaf at 5, 10 and 15 % concentrations were tested against *S. litura* for antifeedant effect, insecticidal effect, effect on adult emergence, fecundity and egg hatching and the results are presented in Table 1.

4.1.1. Antifeedant Effect

The aqueous extracts of *Q. indica* flower and *S. indica* leaf showed statistically significant variation in antifeedant activity against *S. litura*. *S. indica* leaf extract 15 % recorded maximum leaf protection of 10.98 per cent on one day after treatment and it was found significantly inferior to NSKE 5 % (48.77 %). The lowest percentage leaf protection (3.88) was recorded in *Q. indica* flower extract 15 % and it was statistically on par with *S. indica* leaf extract 10 % (4.30). The aqueous plant extracts exhibited antifeedant activity 24 hours after treatment and it was absent from second day onwards.

4.1.2. Insecticidal Effect

The aqueous extracts of *Q. indica* flower and *S. indica* leaf were found ineffective to impart insecticidal activity on *S. litura* as there was no larval mortality.

Table 1. Effect of different aqueous plant extracts on feeding inhibition, adult emergence, fecundity and egg hatching of *S. litura*

Treatments	Mean percentage leaf protection**	Effect on adult emergence**				Effect on fecundity and egg hatchability***	
		Mean larval duration (Days)	Mean pupal duration (Days)	Mean adult longevity (Days)	Mean pupal weight (g)	No. of eggs laid per female	No. of eggs hatched
<i>Q. indica</i> flower extract (5%)	0*	13.00±0.5	8.83±0.5	7.05±0.58	0.32±0.02	671.40±40.99	652.40±44.90
<i>Q. indica</i> flower extract (10%)	0*	12.83±0.29	9.33±0.29	8.00±0.5	0.28±0.03	653.60±67.26	630.60±65.23
<i>Q. indica</i> flower extract (15%)	3.88 ± 0.27 ^c	12.17±0.29	9.17±0.29	7.33±0.58	0.26±0.03	664.60±70.74	640.00±71.19
<i>S. indica</i> leaf extract (5%)	0*	13.00±0.5	8.33±0.29	8.00±0.00	0.30±0.03	683.60±52.72	656.80±48.80
<i>S. indica</i> leaf extract (10%)	4.30 ± 0.29 ^c	12.83±0.29	8.33±0.58	7.50±0.5	0.27±0.02	627.00±73.62	598.20±51.79
<i>S. indica</i> leaf extract (15%)	10.98 ± 0.75 ^b	13.00±0.29	9.33±0.58	7.66±0.5	0.26±0.04	698.80±39.80	666.20±72.54
NSKE 5%	48.77 ± 3.33 ^a	12.17±0.58	8.00±0.5	7.00±1.00	0.29±0.03	654.60±54.59	605.00±51.03
Untreated	0*	13.67±0.5	9.17±0.5	8.00±0.58	0.33±0.04	681.60±72.60	663.00±60.22
CD (0.05)	2.18	NS	NS	NS	NS	NS	NS

* Excluded for analysis

**Mean of three replications; Mean ± SD

***Mean of five replications; Mean ± SD

4.1.3. Effect on Adult Emergence

The aqueous extracts of *Q. indica* flower and *S. indica* leaf had no effect on deformation and mortality of larvae, pupae and adults, larval and pupal duration, pupal weight and adult longevity (Table 1).

4.1.4. Effect on Fecundity and Egg Hatching

The aqueous extracts of *Q. indica* flower and *S. indica* leaf had no effect on fecundity and egg hatching of *S. litura* (Table 1).

4.2. BIOEFFICACY OF SOLVENT EXTRACTS ON *S. litura*

The cold and soxhlet extracts of *Q. indica* flower and *S. indica* leaf at 1.25, 2.5 and 5 % concentrations were tested for antifeedant effect, insecticidal effect, effect on adult emergence, fecundity and egg hatching on *S. litura*. The results are given in Table 2-8.

4.2.1. *Q. indica* Flower

4.2.1.1. Antifeedant Effect

4.2.1.1.1. Cold Extracts

The cold extracts of *Q. indica* flower showed statistically significant variation in antifeedant activity against *S. litura* (Table 2). Among the cold extracts of *Q. indica* flower, maximum percentage leaf protection (31.87) was observed in methanol extract at 5 % concentration and it was significantly different from all other treatments at twenty four hours after treatment. Ethyl acetate extract 5 % showed 27.24 per cent leaf protection and it was found to be statistically on par with methanol extract 2.5 % (24.24 %). Methanol and ethyl acetate extract 2.5 % showed percentage leaf protection of 24.24 and 20.62 respectively and the effect of these two treatments was statistically on par. Methanol extract of *Q. indica* flower at 1.25 % concentration was found to be statistically on par with ethyl acetate extract 2.5 and 1.25 % and the percentage leaf protection in these treatments ranged from 13.23 to 20.62. Minimum leaf protection was recorded in ethyl acetate extract 1.25 % (13.23) and it was statistically similar to methanol extract 1.25 % (16.95). All the treatments

Table 2. Antifeedant effect of solvent extracts of *Q. indica* flower on *S. litura*

Treatments	Mean percentage leaf protection					
	Cold extraction			Soxhlet extraction		
	Hours after treatment			Hours after treatment		
	24	48	72	24	48	72
Ethyl acetate 1.25%	13.23±1.23 ^f	9.72±0.71 ^e	6.37±0.40 ^f	10.02±0.81 ^e	7.07±0.61 ^e	3.54±0.28 ^f
Ethyl acetate 2.5%	20.62±1.92 ^{de}	16.56±1.22 ^d	10.92±0.68 ^e	17.68±1.43 ^d	11.98±1.03 ^d	8.89±0.70 ^d
Ethyl acetate 5%	27.24±2.54 ^c	20.86±1.53 ^c	17.57±1.10 ^c	22.22±1.80 ^c	18.60±1.60 ^c	11.05±0.87 ^c
Methanol 1.25%	16.95±1.58 ^{ef}	10.78±0.79 ^e	7.35±0.46 ^f	12.36±1.00 ^e	9.97±0.86 ^{de}	6.27±0.49 ^e
Methanol 2.5%	24.24±2.26 ^{cd}	18.67±1.37 ^{cd}	13.43±0.84 ^d	20.35±1.64 ^{cd}	17.11±1.47 ^c	11.19±0.88 ^c
Methanol 5%	31.87±2.97 ^b	26.99±1.98 ^b	20.64±1.29 ^b	28.80±2.33 ^b	23.36±2.01 ^b	16.83±1.32 ^b
NSKE 5%	56.41±5.26 ^a	51.80±3.81 ^a	48.77±3.06 ^a	59.15±4.78 ^a	53.87±4.64 ^a	50.27±3.95 ^a
CD (0.05)	4.45	2.86	1.96	3.45	3.06	2.12

Mean of three replications; Mean ± SD

No percentage leaf protection was recorded in untreated and control.

showed significantly inferior mean leaf protection against *S. litura* compared to NSKE 5 % (56.41 %).

Cold extracts of *Q. indica* flower showed decreasing trend in antifeedant activity over exposure time. Methanol extract of *Q. indica* flower 5 % recorded significantly higher percentage leaf protection (26.99 %) and it was significantly different from all other treatments. Ethyl acetate extract 5 % showed 20.86 per cent leaf protection and it was on par with methanol extract 2.5 % (18.67 %). Methanol extract of *Q. indica* flower 2.5 % was found to be statistically on par with ethyl acetate extract 5 and 2.5 % and the percentage leaf protection in these treatments ranged from 16.56 to 20.86. Lowest percentage of leaf protection was recorded in ethyl acetate extract 1.25 % (9.72) and it was statistically on par with methanol extract 1.25 % (10.78). The mean leaf protection exhibited by NSKE 5 % (51.80 %) was found significantly superior to all treatments.

Methanol extract of *Q. indica* flower 5 % showed 20.64 per cent leaf protection at seventy two hours after treatment. Next best treatment in the order of effectiveness were ethyl acetate extract 5 %, methanol extract 2.5 % and ethyl acetate extract 2.5 %. The effect of above three treatments was significantly different and mean leaf protection in these treatments ranged from 10.92 to 17.57 per cent. Ethyl acetate extract 1.25 % was found to be the least effective treatment with mean leaf protection of 6.37 per cent and it was statistically on par with methanol extract 1.25% (7.35 %). None of the treatments was found statistically on par with NSKE 5 % (48.77 %).

4.2.1.1.2. Soxhlet Extracts

Soxhlet extracts of *Q. indica* flower showed statistically significant variation in antifeedant activity with methanol extract 5 % (28.80 %) as the most effective treatment to impart feeding inhibition against *S. litura* at twenty four hours after treatment (Table 2). Ethyl acetate extract 5 % showed 22.22 per cent leaf protection

and it was statistically similar to methanol extract 2.5 % (20.35 %). Methanol and ethyl acetate extracts 2.5 % showed percentage leaf protection of 20.35 and 17.68 respectively and the effect of these two treatments was statistically on par. The least effect on mean leaf protection was recorded in ethyl acetate extract 1.25 % (10.02) and it was statistically on par with methanol extract 1.25 % (12.36).

A decreasing trend in antifeedant activity over exposure time was noticed for soxhlet extracts of *Q. indica* flower. Methanol extract 5 % recorded 23.36 per cent mean leaf protection against *S. litura* at forty eight hours after treatment and it showed statistically significant variation from all other treatments. Ethyl acetate extract 5 % showed 18.60 per cent leaf protection and it was on par with methanol extract 2.5 % (17.11 %). Methanol extract of *Q. indica* flower 1.25 % was found to be statistically on par with ethyl acetate extract at 2.5 and 1.25 % concentration and the percentage leaf protection in these treatments ranged from 7.07 to 11.98. Minimum percentage leaf protection was recorded in ethyl acetate extract 1.25 % (7.07) and it was statistically on par with methanol extract 1.25 % (9.97).

Methanol extract 5 % showed 16.83 per cent leaf protection at seventy two hours after treatment and it was significantly different from all other treatments. Methanol extract 2.5 % showed 11.19 per cent leaf protection and it was statistically on par with ethyl acetate extract 5 % (11.05 %). It was followed by ethyl acetate extract 2.5 %, methanol extract 1.25 % and ethyl acetate extract 1.25 %. The effect of these treatments was significantly different and mean leaf protection in these treatments ranged from 3.54 to 8.89. Lowest percentage leaf protection was recorded in ethyl acetate extract 1.25 % (3.54).

The soxhlet extracts of *Q. indica* flower was observed significantly inferior to NSKE 5 % which recorded 59.15, 53.87 and 50.27 per cent mean leaf protection at 24, 48 and 72 hours after treatment respectively.

4.2.1.2. Insecticidal Effect

4.2.1.2.1. Cold Extracts

The insecticidal effect of cold extracts of *Q. indica* flower was assessed through spraying, leaf dip and dry film methods and the results are presented in Table 3.

4.2.1.2.1.1. Spraying Method

Cold extracts of *Q. indica* flower showed statistically significant variation in insecticidal activity against *S. litura*. Among the cold extracts, methanol extract of *Q. indica* flower at 5 per cent concentration recorded significantly higher percentage larval mortality (70) at twenty four hours after treatment and it was found statistically on par with quinalphos 0.05 % (76.67 %). Methanol extract 2.5 % recorded 60 per cent larval mortality and it was on par with ethyl acetate extract 5 % (58.33 %). The effect of methanol extract 1.25 % and ethyl acetate extract 2.5 % was found statistically similar giving larval mortality of 41.67 per cent. Minimum larval mortality was recorded in ethyl acetate extract 1.25 % (25 %) and it was significantly different from all other treatments.

An increase in larval mortality over exposure time was noticed among the cold extracts of *Q. indica* flower. Methanol extract 5 % recorded 86.99 per cent larval mortality and it was found statistically on par with quinalphos 0.05 % (90.40%) and significantly superior to all other treatments at forty eight hours after treatment. Ethyl acetate extract 5 % recorded 71.70 per cent larval mortality and it was on par with methanol extract 2.5 % (70.34 %). The effect of ethyl acetate extract 2.5 % and methanol extract 1.25 % was found statistically on par showing 55.02 and 48.31 per cent larval mortality respectively. The least effect on larval mortality was recorded in ethyl acetate extract 1.25 % (33.26 %) and it was significantly different from all other treatments.

Methanol extract 5 % showed 93.51 per cent larval mortality at seventy two hours after treatment and it was found significantly inferior to quinalphos 0.05 %

Table 3. Insecticidal effect of solvent extracts of *Q. indica* flower on *S. litura*

Treatments	Mean percentage mortality at different intervals						
	Cold extraction				Soxhlet extraction		
	Spraying method			Dry film method	Spraying method		
	Hours after treatment				Hours after treatment		
24	48	72	24	48	72		
Ethyl acetate 1.25%	25.00±1.76 ^d	33.26±5.77 ^d (35.22)	39.96±5.00 ^e (39.21)	9.60±5.00 ^e (18.05)	21.67± 2.89 ^d	29.92±5.00 ^d (33.16)	34.95±5.00 ^d (36.24)
Ethyl acetate 2.5%	41.67±1.70 ^c	55.02±5.00 ^c (47.88)	61.69±2.89 ^d (51.76)	26.63±2.89 ^d (31.07)	28.33±2.89 ^d	39.96±5.00 ^d (39.21)	48.31±7.64 ^d (44.03)
Ethyl acetate 5%	58.33±1.94 ^b	71.70±2.89 ^b (57.86)	81.72±2.89 ^c (64.69)	34.95±5.00 ^{cd} (36.24)	53.33±7.64 ^{bc}	60.04±5.00 ^c (50.79)	70.08±5.00 ^c (56.84)
Methanol 1.25%	41.67±3.60 ^c	48.31±7.64 ^c (44.03)	56.73±7.64 ^d (48.87)	26.63±2.89 ^d (31.07)	46.67±7.64 ^c	56.73±7.64 ^c (48.87)	63.45±7.64 ^c (52.80)
Methanol 2.5%	60.00±1.29 ^b	70.34±10.00 ^b (57.00)	76.98±7.64 ^c (61.33)	38.31±2.89 ^c (38.24)	61.67±5.77 ^b	68.49±7.64 ^{bc} (55.85)	75.11±5.00 ^c (60.07)
Methanol 5%	70.00±1.48 ^a	86.99±5.00 ^a (68.86)	93.51±2.89 ^b (75.24)	55.02±5.00 ^b (47.88)	61.67±7.64 ^b	76.98±7.64 ^b (61.33)	89.07±7.64 ^b (70.69)
Quinalphos 0.05%	76.67±1.71 ^a	90.40±5.00 ^a (71.95)	99.38±2.89 ^a (85.50)	93.51±2.89 ^a (75.24)	73.33±7.64 ^a	89.07±7.64 ^a (70.69)	97.71±2.89 ^a (81.29)
CD (0.05)	8.96	(7.54)	(7.48)	(5.20)	11.14	(7.95)	(8.64)

Mean of three replications; Mean ± SD; Figures in parentheses are arc sin transformed values
 No percentage larval mortality was recorded in untreated and control

(99.38 %). The effect of ethyl acetate extract 5 % (81.72 %) was statistically on par with methanol extract 2.5 % (76.98 %). Ethyl acetate extract 2.5 % showed 61.69 per cent larval mortality and it was found statistically on par with methanol extract 1.25% (56.73 %). Minimum larval mortality (39.96 %) was observed in ethyl acetate extract 1.25 % and it was significantly different from all other treatments.

4.2.1.2.1.2. Leaf Dip Method

The cold extracts of *Q. indica* flower were found ineffective to exhibit insecticidal action against *S. litura* larvae by leaf dip method.

4.2.1.2.1.3. Dry Film Method

The treatments showed statistically significant variation in insecticidal activity against *S. litura* in dry film method (Table 3). Among the cold extracts of *Q. indica* flower maximum larval mortality (55.02 %) was recorded in methanol extract 5 % and it was found significantly superior to all other treatments. Methanol extract 2.5% showed 38.31 per cent larval mortality and it was statistically on par with ethyl acetate extract 5 % (34.95 %). Methanol extract 1.25 % was found to be statistically on par with ethyl acetate extract 5 and 2.5 % and the percentage larval mortality in these treatments ranged from 26.63 to 34.95. Significantly lower percentage larval mortality (9.60) was recorded in ethyl acetate extract 1.25 %. None of the treatments was found statistically on par with quinalphos 0.05 %, which recorded 93.51 per cent mortality against *S. litura* larvae.

4.2.1.2.2. Soxhlet Extracts

The insecticidal effect of soxhlet extracts of *Q. indica* flower was assessed through spraying and leaf dip method and the results are presented in Table 3.

4.2.1.2.2.1. Spraying Method

Soxhlet extracts of *Q. indica* flower showed statistically significant variation in insecticidal activity against *S. litura* with maximum larval mortality (61.67 %) recorded in methanol extract 5 and 2.5 % at twenty four hours after treatment and it was statistically on par with ethyl acetate extract 5 %. Ethyl acetate extract 5 %

showed 53.33 per cent larval mortality and it was found to be statistically on par with methanol extract 1.25 % (46.67 %). Ethyl acetate extract 1.25 % recorded minimum larval mortality (21.67%) and it was statistically on par with ethyl acetate extract 2.5% (28.33 %).

An increasing trend in insecticidal activity over exposure time was noticed for soxhlet extracts of *Q. indica* flower. Methanol extract 5 % recorded maximum larval mortality of 76.98 per cent against *S. litura* at forty eight hours after treatment and it was statistically on par with methanol extract 2.5 % (68.49 %). Ethyl acetate extract 5 % showed 60.04 per cent larval mortality and it was on par with methanol extract 2.5 and 1.25 % and percentage larval mortality in these treatments ranged from 56.73 to 68.49. Lowest percentage larval mortality (29.92) was recorded in ethyl acetate extract 1.25 % and it was statistically on par with ethyl acetate extract 2.5 % (39.96%).

Methanol extract of *Q. indica* flower at 5 per cent concentration gave 89.07 per cent larval mortality at seventy two hours after treatment and it showed statistically significant variation from all other treatments. Methanol extract 2.5 % showed 75.11 per cent larval mortality and it was statistically on par with ethyl acetate extract 5 % and methanol extract 1.25 % giving 70.08 and 63.45 per cent larval mortality respectively. Significantly lower percentage larval mortality (34.95) was recorded in ethyl acetate extract 1.25 % and it was statistically on par with ethyl acetate extract 2.5 % (48.31).

The soxhlet extracts of *Q. indica* flower exhibited significantly inferior insecticidal activity compared to quinalphos 0.05%, which recorded 73.33, 89.07 and 97.71 per cent larval mortality at 24, 48 and 72 hours after treatment respectively.

4.2.1.2.2.2. Leaf Dip Method

The soxhlet extracts of *Q. indica* flower had no effect on larval mortality of *S. litura* by leaf dip method.

4.2.1.2.3. Comparison of Method of Extraction

4.2.1.2.3.1. Antifeedant Effect

Comparison of cold and soxhlet extraction methods proved that the two methods of extraction differed significantly in their performance (Table 4). Cold extraction (12.72%) method was significantly superior to soxhlet extraction method (9.63%).

The treatments showed statistically significant variation in antifeedant activity against *S. litura*. The significantly superior treatment observed was methanol extract 5%, which recorded 18.74 per cent leaf protection. Next best treatments in the order of effectiveness were ethyl acetate extract 5%, methanol extract 2.5%, ethyl acetate extract 2.5% and methanol extract 1.25%. The effect of these treatments was found significantly different and the percentage leaf protection in these treatments ranged from 6.81 to 14.31. Ethyl acetate extract 1.25% recorded significantly lower percentage leaf protection (4.96).

4.2.1.2.3.2. Insecticidal Effect

Among the two methods of extraction, cold extraction method (70.05 %) was found significantly superior to soxhlet extraction method (64.54 %) (Table 4).

The solvent extracts of *Q. indica* flower showed statistically significant variation in larval mortality against *S. litura*. Methanol extract 5 % exhibited significantly higher percentage larval mortality (91.42) in both extraction methods. The effect of ethyl acetate extract 5% (76.15%) was found statistically on par with methanol extract 2.5 % (76.05 %). Methanol extract 1.25 % showed 60.11 per cent larval mortality and it was statistically on par with ethyl acetate extract 2.5 % (55.05%). Minimum percentage larval mortality (37.43 %) was recorded in ethyl acetate extract 1.25 % and it was significantly different from all other treatments.

The data on antifeedant and insecticidal effects exhibited by solvent extracts of *Q. indica* flower clearly showed that cold extraction was significantly superior to

Table 4. Antifeedant and insecticidal effect of cold and soxhlet extracts of *Q. indica* flower

Treatments	Antifeedant effect			Insecticidal effect (Spraying method)		
	Extraction methods		Mean percentage leaf protection	Extraction methods		Mean percentage mortality
	Cold	Soxhlet		Cold	Soxhlet	
Ethyl acetate 1.25%	6.37	3.54	4.96 ^f	39.96 (39.21)	34.95 (36.24)	37.43 ^d (37.72)
Ethyl acetate 2.5%	10.92	8.89	9.91 ^d	61.69 (51.76)	48.31 (44.03)	55.05 ^c (47.90)
Ethyl acetate 5%	17.57	11.05	14.31 ^b	81.72 (64.69)	70.08 (56.84)	76.15 ^b (60.77)
Methanol 1.25%	7.35	6.27	6.81 ^e	56.73 (48.87)	63.45 (52.80)	60.11 ^c (50.83)
Methanol 2.5%	13.43	11.19	12.31 ^c	76.98 (61.33)	75.11 (60.07)	76.05 ^b (60.70)
Methanol 5%	20.64	16.83	18.74 ^a	93.51 (75.24)	89.07 (70.69)	91.42 ^a (72.97)
Mean	12.72 ^a	9.63 ^b		70.05 ^a (56.85)	64.54 ^b (53.45)	

Mean of three replications; Figures in parentheses are arc sin transformed values

soxhlet extraction. Hence cold extraction method was accepted for further studies on the bioefficacy of *Q. indica* flower.

4.2.1.3. Effect on Adult Emergence

The cold extracts of *Q. indica* flower showed statistically significant variation in mortality of larvae and pupae of *S. litura*. However there was no effect on duration of larvae, pupae and adults and the treatments were found ineffective to cause larval, pupal and adult malformations (Table 5).

Significantly higher percentage larval mortality (30) was exhibited by methanol extract 5 %. The next best treatment was methanol extract 2.5 % (20 %) and it was statistically on par with ethyl acetate extract 5 % and NSKE 5 % showing 18.33 and 13.33 per cent larval mortality respectively. NSKE 5 % was found statistically on par with methanol extract 2.5 %, ethyl acetate extract 5 %, ethyl acetate extract 2.5 % and methanol extract 1.25 %. The larval mortality in the above four treatments ranged from 6.67 to 20 per cent. The least larval mortality (6.67) was recorded in methanol extract 1.25 %. The effect of this treatment was on par with ethyl acetate extract 2.5 % and NSKE 5 % giving 8.33 and 13.33 per cent larval mortality respectively.

Methanol extract 5 % recorded maximum pupal mortality of 18.94 per cent among the cold extracts of *Q. indica* flower and it was found significantly inferior to NSKE 5 % (32.67 %) and statistically on par with ethyl acetate extract 5 % (14.25%). Minimum percentage pupal mortality (12.53) was recorded in methanol extract 2.5 % and it was statistically on par with ethyl acetate extract 5 % (14.25).

4.2.1.4. Effect on Fecundity and Egg Hatching

The data presented in Table 5 showed that the solvent extracts of *Q. indica* flower had no influence on the number of eggs laid by *S. litura* and the number of eggs hatched.

Table 5. Effect of cold extracts of *Q. indica* flower on adult emergence, fecundity and egg hatching of *S. litura*

Treatments	Effect on adult emergence**						Effect on fecundity and egg hatchability***	
	Mean larval duration (Days)	Mean pupal duration (Days)	Mean adult longevity (Days)	Mean pupal weight (g)	Mean percentage larval mortality	Mean percentage pupal mortality	No. of eggs laid per female	No. of eggs hatched
Ethyl acetate 1.25%	12.83±0.5	9.5±0.5	8.00±0.5	0.32±0.03	0*	0*	661.40±58.59	642.00±66.57
Ethyl acetate 2.5%	13.00±0.5	9.33±0.58	7.66±0.58	0.29±0.02	8.33±2.89 ^c	0*	673.40±66.09	640.20±55.13
Ethyl acetate 5%	12.17±0.29	8.83±0.29	7.50±0.58	0.28±0.03	18.33±5.77 ^b	14.25±3.05 ^{bc}	637.80±40.02	603.60±37.67
Ethyl acetate alone	13.83±0.58	9.17±0.29	8.00±0.00	0.31±0.03	0*	0*	679.60±50.25	644.40±48.91
Methanol 1.25%	13.67±0.58	8.83±0.29	7.66±1.00	0.28±0.03	6.67±2.89 ^c	0*	707.80±51.00	679.60±46.40
Methanol 2.5%	12.83±0.37	9.00±0.58	7.33±0.58	0.27±0.02	20.00±5.00 ^b	12.53±0.79 ^c	680.00±77.27	640.80±52.16
Methanol 5%	13.67±0.29	8.5±0.5	7.00±0.00	0.25±0.04	30.00±5.00 ^a	18.94±3.16 ^b	621.60±71.98	574.80±83.12
Methanol alone	13.00±0.29	9.5±0.5	8.00±0.5	0.34±0.04	0*	0*	649.40±63.30	624.80±65.27
NSKE 5%	12.17±0.5	8.33±0.29	7.50±0.5	0.23±0.03	13.33±2.89 ^{bc}	32.67±2.97 ^a	664.60±52.11	604.40±66.32
Control	13.83±0.5	9.5±0.5	8.33±0.58	0.33±0.03	0*	0*	682.20±44.48	665.60±56.15
CD (0.05)	NS	NS	NS	NS	7.56	5.06	NS	NS

*Excluded for analysis

**Mean of three replications; Mean ± SD

***Mean of five replications; Mean ± SD

4.2.2. *S. indica* Leaf

4.2.2.1. Antifeedant Effect

4.2.2.1.1. Cold Extracts

The cold extracts of *S. indica* leaf showed statistically significant variation in antifeedant activity against *S. litura* (Table 6) with maximum leaf protection recorded in methanol extract 5 % (45.62 %) at twenty four hours after treatment. The next best treatment was ethyl acetate extract 5 %, which showed 37.71 per cent leaf protection and it was found statistically on par with methanol extract 2.5 % (36.16 %). The effect of ethyl acetate extract 2.5%, methanol extract 1.25 % and ethyl acetate extract 1.25 % was significantly different and mean leaf protection in the above three treatments ranged from 18.92 to 28.39. Lowest percentage leaf protection (18.92) was recorded in ethyl acetate extract 1.25 %.

Effect of cold extracts of *S. indica* leaf showed a decreasing trend in antifeedant activity forty eight hours after treatment. Methanol extract 5 % showed statistically significant variation from all other treatments and recorded maximum leaf protection of 42.72 per cent. Ethyl acetate extract 5 % showed 34.06 per cent leaf protection and it was found to be statistically on par with methanol extract 2.5 % (32.03 %). Methanol and ethyl acetate extracts at 2.5 % concentration showed statistically significant variation giving 32.03 and 23.45 per cent leaf protection respectively. The effect of lower concentrations of methanol and ethyl acetate extracts (1.25 %) recorded mean leaf protection of 18.98 and 14.28 per cent respectively.

Methanol extract 5 % gave maximum mean leaf protection of 34.61 per cent at seventy two hours after treatment and it was significantly superior to all other treatments. Effect of methanol extract 2.5 % and ethyl acetate extract 5 % was statistically on par showing 27.63 and 26.87 per cent leaf protection respectively. Ethyl acetate extract 2.5 % (17.35 %) was significantly inferior to the same solvent extract at 5 % concentration. The least effect on mean leaf protection (7.45 %) was

Table 6. Antifeedant effect of solvent extracts of *S. indica* leaf on *S. litura*

Treatments	Mean percentage leaf protection					
	Cold extraction			Soxhlet extraction		
	Hours after treatment			Hours after treatment		
	24	48	72	24	48	72
Ethyl acetate 1.25%	18.92±1.08 ^f	14.28±0.73 ^f	7.45±0.39 ^f	16.24±0.87 ^e	13.73±0.90 ^f	8.32±0.57 ^e
Ethyl acetate 2.5%	28.39±1.62 ^d	23.45±1.20 ^d	17.35±0.91 ^d	22.51±1.21 ^d	18.05±1.19 ^e	11.36±0.77 ^d
Ethyl acetate 5%	37.71±2.15 ^c	34.06±1.74 ^c	26.87±1.40 ^c	34.93±1.87 ^c	29.61±1.95 ^c	20.74±1.41 ^c
Methanol 1.25%	24.72±1.41 ^e	18.98±0.97 ^e	12.78±0.67 ^e	22.14±1.19 ^d	18.05±1.19 ^e	10.80±0.73 ^d
Methanol 2.5%	36.16±2.06 ^c	32.03±1.64 ^c	27.63±1.44 ^c	32.97±1.77 ^c	24.54±1.61 ^d	18.99±1.29 ^c
Methanol 5%	45.62±2.60 ^b	42.72±2.19 ^b	34.61±1.81 ^b	39.98±2.14 ^b	36.04±2.37 ^b	27.73±1.89 ^b
NSKE 5%	60.74±3.46 ^a	56.21±2.88 ^a	52.38±2.73 ^a	57.93±3.11 ^a	51.86±3.41 ^a	46.85±3.19 ^a
CD (0.05)	3.60	2.84	2.34	3.04	3.15	2.46

Mean of three replications; Mean ± SD

No percentage leaf protection was recorded in untreated and control

observed in ethyl acetate extract 1.25 % and it was significantly different from methanol extract 1.25 % (12.78 %).

NSKE 5 % recorded 60.74, 56.21 and 52.38 per cent mean leaf protection against *S. litura* at 24, 48 and 72 hours after treatment respectively and its effect was found superior to the cold extracts of *S. indica* leaf.

4.2.2.1.2. Soxhlet Extracts

Effect of Soxhlet extracts of *S. indica* leaf showed statistically significant variation in mean leaf protection against *S. litura* (Table 6). Maximum percentage leaf protection (39.98) was observed in methanol extract 5 % and it was found significantly superior to the other soxhlet extracts at twenty four hours after treatment. Ethyl acetate extract 5 % showed 34.93 per cent leaf protection and it was found statistically on par with methanol extract 2.5 % (32.97 %). The effect of ethyl acetate extract 2.5 % and methanol extract 1.25 % was statistically on par showing mean leaf protection of 22.51 and 22.14 per cent respectively. The least effective treatment observed was ethyl acetate extract 1.25 % (16.24 %) and it was significantly different from all other treatments.

Decreasing trend in antifeedant activity over exposure time was noticed for soxhlet extracts of *S. indica* leaf. Methanol extract 5 % recorded significantly higher percentage leaf protection (36.04) against *S. litura* at forty eight hours after treatment. Next best treatments in the order of effectiveness were ethyl acetate extract 5 % and methanol extract 2.5 %. The effect of these treatments was significantly different and mean leaf protection in these treatments ranged from 24.54 to 29.61 per cent. The effect of ethyl acetate extract at 2.5 % concentration was statistically on par with the lower concentration of methanol extract (1.25 %) giving mean leaf protection of 18.05 per cent. Lowest percentage leaf protection (13.73) was recorded in ethyl acetate extract 1.25 % and it was significantly different from all other treatments.

Methanol extract at 5 % concentration showed 27.73 per cent leaf protection at seventy two hours after treatment and it was found significantly superior to the

other soxhlet extracts of *S. indica* leaf. Ethyl acetate extract 5 % showed 20.74 per cent leaf protection and it was statistically on par with methanol extract 2.5 % (18.99%). The effect of ethyl acetate extract 2.5 % and methanol extract 1.25 % was statistically on par with mean leaf protection of 11.36 and 10.80 per cent respectively. Minimum leaf protection (8.32 %) was noticed in ethyl acetate extract 1.25 % and it was significantly different from all other treatments.

The mean leaf protection exhibited by the soxhlet extracts of *S. indica* leaf was significantly inferior to NSKE 5 %, which showed 57.93, 51.86 and 46.85 per cent leaf protection at 24, 48 and 72 hours after treatment respectively.

4.2.2.2. Insecticidal Effect

4.2.2.2.1. Cold Extracts

The insecticidal effect of cold extracts of *S. indica* leaf was assessed through spraying, leaf dip and dry film methods and the results are presented in Table 7.

4.2.2.2.1.1. Spraying Method

Data presented in Table 7 showed statistically significant variation among the cold extracts of *S. indica* leaf and the most effective treatment observed was methanol extract 5 % giving 38.33 per cent mortality of *S. litura* larvae at twenty four hours after treatment and the effect of this treatment was statistically on par with its lower concentration 2.5 % (30 %). Ethyl acetate extract 5 % recorded 28.33 per cent larval mortality and it was statistically on par with methanol and ethyl acetate extracts at 2.5% concentration showing 30 and 23.33 % larval mortality respectively. The effect exhibited by ethyl acetate extract 2.5 % was statistically on par with methanol extracts 2.5 and 1.25 % and ethyl acetate extracts 5 and 1.25 % concentrations. The mean leaf protection in the above five treatments ranged from 16.67 to 30 %. Ethyl acetate extract 1.25 % (16.67 %) was observed to be the least effective treatment and it was statistically on par with methanol extract 1.25 % and ethyl acetate extract 2.5% giving 18.33 and 23.33 % larval mortality respectively.

Table 7. Insecticidal effect of cold extracts of *S. indica* leaf on *S. litura*

Treatments	Mean percentage mortality at different intervals						Dry film method
	Spraying method			Leaf dip method			
	Hours after treatment			Hours after treatment			
	24	48	72	24	48	72	
Ethyl acetate 1.25%	16.67±2.89 ^d	28.11±7.64 ^e (32.02)	33.18±7.64 ^f (35.17)	6.67±2.13 ^c	11.67±2.01 ^c	15.00±2.10 ^e	0*
Ethyl acetate 2.5%	23.33±2.89 ^{cd}	34.95±5.00 ^{de} (36.24)	44.98±5.00 ^{de} (42.12)	8.33±2.65 ^c	13.33±2.30 ^c	20.00±2.80 ^{de}	6.49±2.89 ^d (14.76)
Ethyl acetate 5%	28.33±7.64 ^c	46.65±5.77 ^c (43.08)	56.68±2.89 ^c (48.84)	10.00±3.19 ^{bc}	16.67±2.88 ^c	26.67±3.73 ^{cd}	11.56±2.89 ^{cd} (19.88)
Methanol 1.25%	18.33±2.89 ^d	29.92±5.00 ^e (33.16)	38.23±7.64 ^{ef} (38.19)	8.33±2.65 ^c	16.67±2.88 ^c	23.33±3.26 ^{cd}	9.61±5.00 ^{cd} (18.05)
Methanol 2.5%	30.00±5.00 ^{bc}	43.27±7.64 ^{cd} (41.13)	55.02±5.00 ^{cd} (47.88)	13.33±4.25 ^{bc}	23.33±4.03 ^b	30.00±4.19 ^c	18.28±2.89 ^c (25.31)
Methanol 5%	38.33±2.89 ^b	61.77±7.64 ^b (51.81)	73.55±7.64 ^b (59.05)	16.67±5.31 ^b	26.67±4.60 ^b	41.67±5.83 ^b	29.92±5.00 ^b (33.16)
Quinalphos 0.05%	68.33±7.64 ^a	86.76±2.89 ^a (68.66)	95.00±0.00 ^a (77.08)	38.33±12.22 ^a	45.00±7.77 ^a	55.00±7.69 ^a	97.71±2.89 ^a (81.29)
CD (0.05)	8.76	(6.67)	(6.16)	8.11	6.62	7.40	(7.57)

Mean of three replications; Mean ± SD ; Figures in parentheses are arc sin transformed values

No percentage larval mortality was recorded in untreated and control

*Excluded for analysis

Cold extracts of *S. indica* leaf showed increasing trend in insecticidal activity over exposure time. Methanol extract 5 % recorded maximum larval mortality (61.77%) against *S. litura* at forty eight hours after treatment and showed statistically significant variation from the other cold extracts. Ethyl acetate extract 5 % showed 46.65 per cent larval mortality and it was on par with methanol extract 2.5 % (43.27%). The larval mortality recorded in ethyl acetate extract 2.5 % was statistically on par with its lower concentration 1.25 % and methanol extracts 2.5 and 1.25 % concentrations. The larval mortality in the above four treatments ranged from 28.11 to 43.27. Minimum larval mortality (28.11 %) was recorded in ethyl acetate extract 1.25 % and it was statistically on par with methanol extract 1.25 % and ethyl acetate extract 2.5 % showing 29.92 and 34.95 % larval mortality respectively.

Methanol extract 5 % of *S. indica* leaf gave 73.55 per cent larval mortality at seventy two hours after treatment. The next best treatment was ethyl acetate extract 5% (56.68 %) and it was statistically on par with methanol extract 2.5 % (55.02 %) and the latter was on par with ethyl acetate extract 2.5 % (44.98 %). The larval mortality recorded in ethyl acetate extract 2.5 % was found to be on par with methanol extract 2.5 and 1.25 % and percentage larval mortality in these treatments ranged from 38.23 to 55.02. The lowest concentration of ethyl acetate extract (1.25%) recorded minimum leaf protection of 33.18 and it was statistically on par with methanol extract 1.25 % (38.23).

The effect of cold extracts of *S. indica* leaf on larval mortality of *S. litura* by spraying method was found significantly inferior to quinalphos 0.05 %, which showed 68.33, 86.76 and 95 per cent larval mortality at 24, 48 and 72 hours after treatment respectively.

4.2.2.2.1.2. Leaf Dip Method

Effect of *S. indica* leaf cold extracts showed statistically significant variation in larval mortality of *S. litura* (Table 7). Maximum percentage larval mortality (16.67) was observed in methanol extract 5 % at twenty four hours after treatment

and the effect of this treatment was statistically on par with methanol extract 2.5 % and ethyl acetate extract 5 % giving larval mortality of 13.33 and 10 per cent respectively. The larval mortality exhibited by the least effective treatment, ethyl acetate extract 1.25 % (6.67 %) showed no statistical variation among ethyl acetate extracts 2.5 and 5 % and methanol extracts 2.5 and 1.25 %. The larval mortality in these treatments ranged from 6.67 to 13.33.

Cold extracts of *S. indica* leaf showed increasing trend in insecticidal activity over exposure time and the highest percentage larval mortality (26.67) was recorded in methanol extract 5 % against *S. litura* at forty eight hours after treatment. The effect of this treatment was statistically similar to methanol extract 2.5 % (23.33 %). Minimum mortality was recorded in ethyl acetate extract 1.25 % (11.67 %) and it was statistically on par with ethyl acetate extract 2.5 and 5 % and methanol extract 1.25%. The percentage mortality in these treatments ranged from 11.67 to 16.67.

Methanol extract 5 % gave 41.67 per cent larval mortality at seventy two hours after treatment and it showed statistically significant variation to other treatments. The treatment next to methanol extract 5 % was methanol extract 2.5 %, which showed 30 per cent larval mortality and it was statistically on par with ethyl acetate extract 5 % and methanol extract 1.25 %. The percentage larval mortality in these treatments ranged from 23.33 to 30. Methanol extract 1.25 % recorded 23.33 per cent larval mortality and it was statistically on par with ethyl acetate extract 5 % and 2.5% showing 26.67 and 20 % larval mortality respectively. Significantly lower percentage larval mortality (15) was recorded in ethyl acetate extract 1.25 % and it was statistically similar to ethyl acetate extract 2.5% (20).

Quinalphos 0.05 % recorded significantly superior larval mortality of 38.33, 45 and 55 per cent at 24, 48 and 72 hours after treatment and none of the treatments was found statistically on par with the chemical.

4.2.2.2.1.3. Dry Film Method

The data presented in Table 7 showed statistically significant variation among the treatments. Significantly higher percentage larval mortality (29.92) was recorded methanol extract 5% and it was found inferior to quinalphos 0.05 % (97.71 %). The effect of methanol extract 2.5% (18.28 %) was statistically similar to ethyl acetate extract 5% (11.56 %) and methanol extract 1.25% (9.61 %). There was no statistical variation observed among methanol extracts at 2.5 and 1.25 % and ethyl acetate extract at 5 and 2.5 % concentrations. The percentage larval mortality in these treatments ranged from 6.49 to 18.28. Least larval mortality (6.49 %) was recorded in ethyl acetate extract 2.5% and it was statistically on par with methanol extract 1.25% and ethyl acetate extract 5% giving 9.61 and 11.56 % larval mortality respectively. Ethyl acetate extract 1.25 % of *S. indica* leaf have no effect on larval mortality against *S. litura* in dry film method.

4.2.2.2.2. Soxhlet Extracts

The insecticidal effect of cold extracts of *S. indica* leaf was assessed through spraying and leaf dip methods and the results are given in Table 8.

4.2.2.2.2.1. Spraying Method

Soxhlet extracts of *S. indica* leaf showed statistically significant variation in insecticidal activity against *S. litura* (Table 8) with the most effective treatment observed was methanol extract 5 % (35 per cent larval mortality) and it was statistically similar to ethyl acetate extract 5 % (30 %). Statistically no variation was observed among methanol and ethyl acetate extracts at 2.5 % concentration giving 25 and 18.33 per cent larval mortality. Ethyl acetate extract 2.5 % showed 18.33 per cent larval mortality and it was statistically on par with methanol extracts 2.5 and 1.25 % and ethyl acetate extract 1.25 %. The percentage larval mortality in these treatments ranged from 11.67 to 25. Least larval mortality (11.67 %) was recorded in the lowest concentration of ethyl acetate extract 1.25 % and it was statistically similar to methanol extract 1.25 % (16.67 %) and ethyl acetate extract 2.5 % (18.33 %).

Table 8. Insecticidal effect of soxhlet extracts of *S. indica* leaf on *S. litura*

Treatments	Mean percentage mortality at different intervals					
	Spraying method			Leaf dip method		
	Hours after treatment			Hours after treatment		
	24	48	72	24	48	72
Ethyl acetate 1.25%	11.67±2.89 ^e	19.84±5.00 ^f (26.45)	24.89±5.00 ^e (29.93)	6.67±1.91 ^c	10.00±1.83 ^e	13.33±1.48 ^e
Ethyl acetate 2.5%	18.33±2.89 ^{de}	31.64±2.89 ^{de} (34.23)	38.31±2.89 ^{cd} (38.24)	6.67±1.91 ^c	11.67±2.13 ^{de}	16.67±1.85 ^{de}
Ethyl acetate 5%	30.00±5.00 ^{bc}	43.32±2.89 ^{bc} (41.16)	50.00±5.00 ^{bc} (45.00)	8.33±2.39 ^{bc}	18.33±3.35 ^{bc}	21.67±2.41 ^{cd}
Methanol 1.25%	16.67±2.89 ^e	24.89±5.00 ^{ef} (29.93)	31.51±7.64 ^{de} (34.15)	8.33±2.39 ^{bc}	16.67±3.04 ^{cd}	18.33±2.04 ^{cde}
Methanol 2.5%	25.00±5.00 ^{cd}	38.23±7.64 ^{cd} (38.19)	48.31±7.64 ^c (44.03)	10.00±2.87 ^{bc}	21.67±3.95 ^{bc}	23.33±2.59 ^c
Methanol 5%	35.00±5.00 ^b	51.69±7.64 ^b (45.97)	61.77±7.64 ^b (51.81)	13.33±3.82 ^b	23.33±4.26 ^b	36.67±4.08 ^b
Quinalphos 0.05%	70.00±5.00 ^a	83.39±2.89 ^a (65.95)	99.38±2.89 ^a (85.50)	35.00±10.04 ^a	43.33±7.91 ^a	51.67±5.75 ^a
CD (0.05)	7.40	(5.73)	(7.76)	6.34	6.62	5.06

Mean of three replications; Mean ± SD; Figures in parentheses are arc sin transformed values
No percentage larval mortality was recorded in untreated and control

Soxhlet extracts of *S. indica* leaf showed increasing trend in insecticidal activity over exposure time and methanol extract 5 % recorded maximum larval mortality (51.69 %) against *S. litura* at forty eight hours after treatment and it was statistically on par with ethyl acetate extract 5 % (43.32 %). The effect of methanol extract 2.5 % was statistically similar to ethyl acetate extract 5 and 2.5 %. The percentage larval mortality in the above treatments ranged from 31.64 to 43.32. Methanol extract 1.25 % recorded 24.89 per cent larval mortality and it was statistically on par with ethyl acetate extract 2.5 and 1.25 % showing 31.64 and 19.84% larval mortality respectively. Ethyl acetate extract 1.25 % (19.84) was the least effective treatment observed and it was statistically on par with methanol extract 1.25 % (24.89).

Methanol extract 5 % showed 61.77 per cent larval mortality at seventy two hours after treatment and it was statistically similar to ethyl acetate extract 5% (50%). Methanol extract 2.5 % showed 48.31 per cent larval mortality and it was statistically on par with ethyl acetate extract 5 and 2.5 % and the percentage larval mortality in these treatments ranged from 38.31 to 50. There was no significant variation observed among ethyl acetate extract 2.5 % and methanol extract 1.25 % giving 38.31 and 31.51 per cent larval mortality respectively. Minimum percentage larval mortality (24.89) was recorded in ethyl acetate extract 1.25 % and it was statistically on par with methanol extract 1.25 % (31.51).

Quinalphos 0.05 % was found significantly superior in insecticidal action compared to the soxhlet extracts of *S. indica* leaf showing 70, 83.39 and 99.38 per cent mortality of *S. litura* larvae at 24, 48 and 72 hours after treatment.

4.2.2.2.2. Leaf Dip Method

Soxhlet extracts of *S. indica* leaf showed statistically significant variation in insecticidal activity against *S. litura* (Table 8). Methanol extract 5 % gave 13.33 per cent larval mortality and it was statistically on par with methanol extract 2.5 %, ethyl acetate extract 5 % and methanol extract 1.25 %. The percentage larval mortality in

these treatments ranged from 8.33 to 13.33. Minimum larval mortality (6.67 %) was recorded in ethyl acetate extract 2.5 and 1.25 % and it was statistically on par with methanol extract 1.25 % and ethyl acetate extract 5 % (8.33 %).

Among the soxhlet extracts of *S. indica* leaf, methanol extract 5% recorded significantly higher percentage larval mortality (23.33) against *S. litura* at forty eight hours after treatment and the effect of this treatment was statistically on par with methanol extract 2.5 % (21.67 %) and ethyl acetate extract 5 % (18.33 %). Methanol extract 1.25 % showed 16.67 per cent larval mortality and it was on par with methanol extract 5 %, ethyl acetate extract 5 and 2.5 % and the percentage larval mortality in these treatments ranged from 11.67 to 21.67. Least larval mortality (10%) was recorded in ethyl acetate extract 1.25 % and it was found to be statistically on par with ethyl acetate extract 2.5 % (11.67 %).

Methanol extract 5 % showed 36.67 per cent larval mortality at seventy two hours after treatment and it was significantly superior to all other soxhlet extracts of *S. indica* leaf. Methanol extract 2.5 %, which was inferior to methanol extract 5 % showed 23.33 per cent larval mortality and it was statistically on par with ethyl acetate extract 5 % (21.67 %) and methanol extract 1.25 % (18.33 %). Methanol extract 1.25 % recorded 18.33 per cent larval mortality and it was found statistically on par with methanol extract 2.5 %, ethyl acetate extract 5, 2.5 and 1.25 %. The percentage larval mortality in the above four treatments ranged from 13.33 to 23.33. Ethyl acetate extract 1.25 % (13.33 %) was the least effective treatment observed and it was statistically on par with ethyl acetate extract 2.5 % and methanol extract 1.25% giving larval mortality of 16.67 and 18.33 % respectively.

The insecticidal action exhibited by soxhlet extracts of *S. indica* leaf was found significantly inferior to quinalphos 0.05 % and it showed 35, 43.33 and 51.67 per cent larval mortality at 24, 48 and 72 hours after treatment respectively.

4.2.2.2.3. Comparison of Method of Extraction

4.2.2.2.3.1. Antifeedant Effect

A comparison of the cold and soxhlet methods of extraction showed that the two methods differed significantly in their performance (Table 9). Significantly higher percentage of mean leaf protection (21.12%) was exhibited by cold extracts of *S. indica* leaf compared to its soxhlet extracts (16.32%).

Methanol extract 5% recorded significantly higher percentage mean leaf protection of 31.17. Next best treatment observed was ethyl acetate extract 5%, which recorded 23.81 per cent leaf protection and it was statistically on par with methanol extract 2.5% (23.31%). Statistically significant variation was observed in the mean leaf protection exhibited by ethyl acetate extract 2.5% (14.36%) and methanol extract 1.25% (11.79%). Significantly lower percentage of mean leaf protection (7.89) was exhibited by ethyl acetate extract 1.25% in both extraction methods.

4.2.2.2.3.2. Insecticidal Effect

4.2.2.2.3.2.1. Spraying Method

Among the two methods of extraction evaluated, cold extraction method (50.37 %) was significantly superior to soxhlet extraction method (42.23 %) (Table 9).

The solvent extracts of *S. indica* leaf showed statistically significant variation in larval mortality of *S. litura*. Methanol extract 5 % gave maximum larval mortality (67.80 %) and it showed statistical variation from the other treatments. The next best treatment was ethyl acetate extract 5 % (53.35 %) and it was found statistically on par with methanol extract 2.5 % (51.68 %). Methanol extract 1.25 % showed 34.83 per cent larval mortality and it was statistically on par with ethyl acetate extracts 2.5 and 1.25 % giving 41.63 and 28.95 per cent larval mortality respectively. Minimum

Table 9. Antifeedant and insecticidal effect of cold and soxhlet extracts of *S. indica* leaf in different application methods

Treatments	Antifeedant effect			Insecticidal effect					
				Spraying method			Leaf dip method		
	Extraction methods		Mean percentage leaf protection	Extraction methods		Mean percentage mortality	Extraction methods		Mean percentage mortality
	Cold	Soxhlet		Cold	Soxhlet		Cold	Soxhlet	
Ethyl acetate 1.25%	7.45	8.32	7.89 ^e	33.18 (35.17)	24.89 (29.93)	28.95 ^d (32.55)	15.00	13.33	14.17 ^e
Ethyl acetate 2.5%	17.35	11.36	14.36 ^c	44.98 (42.12)	38.31 (38.24)	41.63 ^c (40.18)	20.00	16.67	18.33 ^{de}
Ethyl acetate 5%	26.87	20.74	23.81 ^b	56.68 (48.84)	50.00 (45.00)	53.35 ^b (46.92)	26.67	21.67	24.17 ^{bc}
Methanol 1.25%	12.78	10.80	11.79 ^d	38.23 (38.19)	31.51 (34.15)	34.83 ^{cd} (36.17)	23.33	18.33	20.83 ^{cd}
Methanol 2.5%	27.63	18.99	23.31 ^b	55.02 (47.88)	48.31 (44.03)	51.68 ^b (45.96)	30.00	23.33	26.67 ^b
Methanol 5%	34.61	27.73	31.17 ^a	73.55 (59.05)	61.77 (51.81)	67.80 ^a (55.43)	41.67	36.67	39.17 ^a
Mean	21.12 ^a	16.32 ^b		50.37 ^a (45.21)	42.23 ^b (40.53)		26.11 ^a	21.67 ^b	

Mean of three replications; Figures in parentheses are arc sin transformed values

percentage larval mortality (28.95) was recorded in ethyl acetate extract 1.25% and it was on par with methanol extract 1.25 % (34.83).

4.2.2.2.3.2.2. Leaf Dip Method

Comparison of cold and soxhlet extraction methods showed that the two methods of extraction differed significantly in their performance (Table 9). Cold extraction (26.11 %) method was significantly superior to soxhlet extraction method (21.67 %).

The treatments showed statistically significant variation in larval mortality against *S. litura*. Methanol extract 5 % recorded significantly higher percentage larval mortality (39.17) among the solvent extracts of *S. indica* leaf. Next effective treatment was methanol extract 2.5 % (26.67 %) and it was statistically on par with ethyl acetate extract 5 % (24.17 %). Methanol extract 1.25 % recorded 20.83 per cent larval mortality and it was on par with ethyl acetate extracts 2.5 and 5 % and the percentage larval mortality in these treatments ranged from 18.33 to 24.17. Ethyl acetate extract 2.5 % (18.33 %) was found statistically similar to methanol extract 1.25 % (20.83 %) and ethyl acetate extract 1.25 % (14.17 %). Minimum percentage larval mortality (14.17) was recorded in ethyl acetate extract 1.25 % and it was statistically on par with ethyl acetate extract 2.5 % (18.33).

The results of antifeedant and insecticidal effects shown by solvent extracts of *S. indica* leaf revealed that the method of cold extraction was significantly superior over soxhlet extraction. Hence cold extraction method was adopted for further studies on the bioefficacy of *S. indica* leaf.

4.2.2.3. Effect on Adult Emergence

The cold extracts of *S. indica* leaf showed statistically significant variation in mortality of larvae and pupae of *S. litura*. However there was no effect on duration of larvae, pupae and adults and the treatments were ineffective to cause larval, pupal and adult malformations (Table 10).

Table 10. Effect of cold extracts of *S. indica* leaf on adult emergence, fecundity and egg hatching of *S. litura*

Treatments	Effect on adult emergence**						Effect on fecundity and egg hatchability***	
	Mean larval duration (Days)	Mean pupal duration (Days)	Mean adult longevity (Days)	Mean pupal weight (g)	Mean percentage larval mortality	Mean percentage pupal mortality	No. of eggs laid per female	No. of eggs hatched
Ethyl acetate 1.25%	13.83±0.58	9.00±0.58	8.33±0.5	0.32±0.03	0*	0*	677.00±67.33	647.40±58.56
Ethyl acetate 2.5%	13.17±0.29	8.83±0.29	8.00±0.58	0.28±0.03	0*	11.67±2.89 ^c	653.00±55.32	610.40±64.86
Ethyl acetate 5%	12.17±0.5	9.17±0.29	7.33±1.00	0.28±0.03	11.67±2.89 ^{ab}	20.81±3.64 ^b	647.20±61.43	598.00±69.85
Ethyl acetate alone	13.00±0.5	9.33±0.58	8.00±0.5	0.30±0.03	0*	0*	666.00±69.31	641.00±70.50
Methanol 1.25%	12.83±0.58	9.00±0.5	7.66±0.58	0.29±0.020	0*	0*	647.60±47.97	611.00±52.76
Methanol 2.5%	12.33±0.58	9.17±0.29	7.00±0.00	0.28±0.20	6.67±2.89 ^b	16.27 ±3.64 ^c	670.80±79.22	625.20±72.71
Methanol 5%	12.33±0.29	8.83±0.29	7.00±0.00	0.25±0.04	16.67±2.89 ^a	31.98±3.01 ^a	650.20±52.23	592.00±53.85
Methanol alone	13.83±0.29	9.17±0.57	8.5±0.58	0.33±0.03	0*	0*	683.80±55.17	651.20±45.01
NSKE 5%	12.17±0.58	8.5±0.5	7.33±0.5	0.25±0.03	13.33±2.89 ^a	38.45±2.97 ^a	677.20±53.46	621.20±3.15
Control	13.83±0.5	9.5±0.5	8.5±0.58	0.34±0.04	0*	0*	667.00±60.75	648.00±69.16
CD (0.05)	NS	NS	NS	NS	5.44	6.52	NS	NS

*Excluded for analysis

**Mean of three replications; Mean ± SD

***Mean of five replications; Mean ± SD

Maximum larval mortality (16.67 %) was recorded in methanol extract 5% and it was statistically similar to NSKE and ethyl acetate extract at 5 % concentration giving 13.33 and 11.67 per cent larval mortality respectively. The least larval mortality (6.67) was recorded in methanol extract 2.5 % and it was statistically on par with ethyl acetate extract 5 % (11.67).

Cold methanol extract of *S. indica* leaf at 5 % concentration recorded 31.98 per cent mortality of pupae and it was statistically on par with NSKE 5 % (38.45 %) The next best treatment was ethyl acetate extract 5 %, which recorded 20.81 per cent larval mortality. Minimum percentage larval mortality (11.67) was recorded in ethyl acetate extract 2.5% and it was statistically on par with methanol extract 2.5 % (16.27).

4.2.2.4. Effect on Fecundity and Egg Hatching

The solvent extracts of *S. indica* leaf had no influence on fecundity and egg hatchability of *S. litura* (Table 10).

4.2.3. Comparison of Plant Parts

The solvent extracts of *Q. indica* flower and *S. indica* leaf were compared for bioefficacy against *S. litura* in terms of antifeedant and insecticidal effect. The results were presented in Table 11.

4.2.3.1. Antifeedant Effect

The two plants differed significantly in their efficacy to impart feeding inhibition to third instar larvae of *S. litura*. *S. indica* leaf extracts performed significantly superior (21.12 per cent leaf protection) compared to *Q. indica* flower extracts (12.72%).

The solvent extracts of plant parts showed statistically significant variation in antifeedant activity against *S. litura*. Methanol extract 5% of both plant parts recorded significantly higher mean leaf protection (27.63 %). Next best treatments in the order of effectiveness were ethyl acetate extract 5%, methanol extract 2.5%, ethyl acetate extract 2.5% and methanol extract 1.25%. The effect of these treatments was

Table 11. Comparison of antifeedant and insecticidal effects of different plant parts on *S. litura*

Solvent extracts	Antifeedant effect			Insecticidal effect					
	Plant parts		Mean percentage leaf protection	Spraying method			Dry film method		
	<i>Q. indica</i> flower	<i>S. indica</i> leaf		<i>Q. indica</i> flower	<i>S. indica</i> leaf	Mean percentage mortality	<i>Q. indica</i> flower	<i>S. indica</i> leaf	Mean percentage mortality
	Ethyl acetate 1.25%	6.37	7.45	6.91 ^f	39.96 (39.21)	33.18 (35.17)	36.54 ^d (37.19)	0*	0*
Ethyl acetate 2.5%	10.92	17.35	14.14 ^d	61.69 (51.76)	44.98 (42.12)	53.38 ^c (46.94)	26.63 (31.07)	6.49 (14.76)	15.15 ^c (22.91)
Ethyl acetate 5%	17.57	26.87	22.22 ^b	81.72 (64.69)	56.68 (48.84)	69.95 ^b (56.76)	34.95 (36.24)	11.56 (19.88)	19.80 ^c (26.42)
Methanol 1.25%	7.35	12.78	10.06 ^e	56.73 (48.87)	38.23 (38.19)	47.44 ^c (43.53)	26.63 (31.07)	9.61 (18.05)	17.28 ^c (24.56)
Methanol 2.5%	13.43	27.63	20.53 ^c	76.98 (61.33)	55.02 (47.88)	66.46 ^b (54.61)	38.31 (38.24)	18.28 (25.31)	27.74 ^b (31.78)
Methanol 5%	20.64	34.61	27.63 ^a	93.51 (75.24)	73.55 (59.05)	84.92 ^a (67.15)	55.02 (47.88)	29.92 (33.16)	42.21 ^a (40.52)
Mean	12.72 ^b	21.12 ^a		70.10 ^a (56.85)	50.37 ^b (45.21)		36.05 ^a (36.90)	13.52 ^b (21.57)	

Mean of three replications; Figures in parentheses are arc sin transformed values

*Excluded for analysis

found to be significantly different and the percentage leaf protection in these treatments range from 10.06 to 22.22. Ethyl acetate extract 1.25% recorded significantly lower percentage of leaf protection (6.91).

4.2.3.2. Insecticidal Effect

4.2.3.2.1. Spraying Method

Q. indica flower extracts was found to be significantly superior (70.10 %) to *S. indica* leaf extracts (50.37 %) to impart larval mortality against *S. litura* (Table 11).

The treatments showed statistically significant variation in larval mortality against *S. litura* by spraying method. Methanol extract 5 % of both plant parts exhibited significantly higher percentage of larval mortality (84.92). It was followed by ethyl acetate extract 5 % (69.95 %) and it was statistically on par with methanol extract 2.5 % (66.46 %). Ethyl acetate extract 2.5 % recorded 53.38 per cent larval mortality and it was statistically on par with methanol extract 1.25 % (47.44 %). Ethyl acetate extract 1.25 % of both plant parts exhibited significantly lower percentage of larval mortality (36.54).

4.2.3.2.2. Dry Film Method

Among the two plant parts compared, cold extracts of *Q. indica* flower performed significantly superior to cold extracts of *S. indica* leaf giving 36.05 and 13.52 per cent larval mortality against *S. litura* (Table 11).

The solvent extracts of plant parts showed statistically significant variation in larval mortality against *S. litura* by dry film method. Methanol extract 5 % of both plants showed statistically significant variation from all other treatments and recorded significantly higher percentage (42.21) of larval mortality against *S. litura*. The next best treatment was methanol extract 2.5 % (27.74 %) and it was significantly different from all other treatments. Least larval mortality (15.15 %) was recorded in ethyl acetate extract 2.5 % and it was found to be statistically on par with methanol extract 1.25 % and ethyl acetate extract 5 % giving 17.28 and 19.80 per cent larval mortality respectively.

Table 12. Effect of treatments on *S. litura* under polyhouse condition

Treatments	Precount	Mean percentage mortality		
		Days after treatment		
		1	2	3
<i>Q. indica</i> flower methanol extract 5%	24.75	44.22±4.64 ^b	57.54±3.01 ^b	61.45±2.16 ^b
<i>S. indica</i> leaf methanol extract 5%	23.5	28.66±1.02 ^c	38.24±3.85 ^c	40.35±1.56 ^c
Quinalphos 25 EC 0.05%	25	53.68±6.29 ^a	72.90±4.24 ^a	84.07±4.14 ^a
<i>B. bassiana</i> (Bb 5) 20g/L	24.5	0*	0*	0*
CD (0.05)	NS	7.28	5.97	4.55

Mean of three replications; Mean ± SD

No larval mortality was recorded in untreated

*Excluded for analysis

Treatments	Mean percentage leaf area damage of <i>S. litura</i> at different intervals after treatment			
	Days after treatment			
	1	3	5	7
<i>Q. indica</i> flower methanol extract 5%	37.56±2.59 ^c	35.71±3.43 ^d	38.82±4.09 ^c (6.31)	48.56±2.86 ^c (7.04)
<i>S. indica</i> leaf methanol extract 5%	48.42±3.34 ^b	46.27±2.94 ^c	47.72±9.79 ^b (6.98)	63.64±2.64 ^b (8.04)
Quinalphos 25 EC 0.05%	19.30±2.37 ^d	12.20±2.44 ^e	14.21±2.70 ^d (3.90)	18.71±3.81 ^d (4.44)
<i>B. bassiana</i> (BB 5) 20g/L	61.96±3.23 ^a	69.69±3.23 ^b	83.46±4.33 ^a (9.19)	92.32±2.69 ^a (9.66)
Untreated	65.29±3.85 ^a	75.73±4.14 ^a	88.11±3.88 ^a (9.44)	95.24±2.40 ^a (9.81)
CD (0.05)	4.70	4.95	(0.62)	(0.37)

Mean of three replications; Mean ± SD; Figures in parentheses are square root transformed values

4.3. EFFECT OF ISOLATED PLANT EXTRACTS ON *S. litura* UNDER POLYHOUSE CONDITION

Methanol extracts of *Q. indica* flower and *S. indica* leaf at 5 % concentration were selected as effective treatments from *in vitro* studies and their efficacy was compared with quinalphos 0.05 % and *B. bassiana* (Bb 5) 20 g/L under polyhouse condition on cowpea. The results are presented in Table 12 and 13.

4.3.1. Mortality of Larvae

All treatments showed statistically significant variation in larval mortality under polyhouse condition (Table 12). Among the two plant extracts, methanol extract of *Q. indica* flower at 5 % concentration exhibited significantly superior larval mortality (44.22 %) compared to the methanol extract 5 % of *S. indica* leaf (28.66 %) on first day after treatment. The effect of the plant extracts was found significantly inferior to quinalphos 0.05 %, which recorded the highest larval mortality of 53.68 per cent against *S. litura*.

Methanol extract of *Q. indica* flower 5 % showed 57.54 per cent larval mortality on second day after treatment. The effect of this treatment was found significantly superior to *S. indica* leaf methanol extract 5 % (38.24 %) and inferior to quinalphos 0.05 % (72.90 %).

The larval mortality (61.45 %) exhibited by methanol extract of *Q. indica* flower 5 % was found significantly superior to *S. indica* leaf methanol extract 5 % (40.35 %) on third day after treatment. The effect of these treatments was found significantly inferior to quinalphos 0.05 % which recorded 84.07 per cent larval mortality.

The biocontrol agent, *B. bassiana* 20 g/L was found ineffective to impart larval mortality of *S. litura*.

4.3.2. Leaf Area Damage

The treatments showed statistically significant variation in percentage leaf area damage of cowpea plants infested by *S. litura* (Table 13).

Among the two plant parts compared, methanol extract of *Q. indica* flower 5% recorded minimum leaf area damage of 37.56 per cent compared to methanol extract of *S. indica* leaf 5 % (48.42 %) on first day after treatment. The effect of these treatments was found significantly inferior to quinalphos 0.05% (19.30 %) and superior to *B. bassiana* 20 g/L (61.96 %). Maximum leaf area damage of 65.29 per cent was recorded in untreated plants.

Methanol extract of *Q. indica* flower 5 % recorded minimum leaf area damage of 35.71 on third day after treatment and it was significantly different from all other treatments. The effect of this treatment was significantly superior to methanol extract of *S. indica* leaf 5 %, which showed 46.27 per cent leaf area damage. The minimum leaf area damage recorded in these plant extracts was significantly superior to *B. bassiana* 20 g/L (69.69 %) and inferior to quinalphos 0.05% (12.20 %). Maximum leaf area damage (75.73 %) was recorded in untreated plants.

Methanol extract of *Q. indica* flower 5 % recorded 38.82 per cent leaf area damage on fifth day after treatment. Minimum leaf area damage of 47.72 per cent was recorded in methanol extract of *S. indica* leaf 5 % and it was found inferior to methanol extract of *Q. indica* flower 5 % and superior to *B. bassiana* 20 g/L (83.46%). Quinalphos 0.05 % exhibited significantly lower percentage of leaf area damage (14.21) and it was found superior to all other treatments. Maximum leaf area damage of 88.11 per cent was recorded in untreated plants.

Among the two plant parts compared, methanol extract of *Q. indica* flower 5% (48.56 %) was found significantly superior to methanol extract of *S. indica* leaf 5% (63.64 %) to impart minimum leaf area damage to cowpea plants infested by *S. litura* on seventh day after treatment. Quinalphos 0.05 % recorded minimum leaf area damage of 18.71 per cent and it was found significantly superior to the plant extracts compared. Maximum leaf area damage of 95.24 per cent was recorded in

untreated plants and it was found statistically on par with *B. bassiana* 20 g/L (92.32%).

Discussion

5. DISCUSSION

Protected cultivation of vegetables which aims at increasing productivity, decreasing production costs and minimizing environmental impact of farming is getting popularized in Kerala as well as in India. *Spodoptera litura* Fabricius is a devastating pest of cowpea and salad cucumber grown under protected condition. Application of chemical pesticides in polyhouses results in serious consequences such as intoxication of people, contamination of water, air and soil, residues in food, resistance in pests and adverse effects on beneficial insects. Plants are important natural sources of bioactive compounds and many such plant compounds have been included in commercial botanical pesticides. Plant derived pesticides are eco-friendly, non-toxic to non-target organisms, non-persistent in nature, besides they do not promote resistance. The rich biodiversity of Kerala opens up a wide opportunity to exploit its diverse flora as better alternative to chemical pesticides.

Considering the above facts, the present study on “Bioefficacy of *Quisqualis indica* L. and *Samadera indica* Gaetrn. against tobacco caterpillar, *Spodoptera litura* Fabricius (Lepidoptera: Noctuidae) in polyhouse condition” was carried out at the College of Agriculture, Vellayani during 2014-2016. In the present study, the aqueous and solvent extracts of *Q. indica* flower and *S. indica* leaf were tested against tobacco caterpillar, *S. litura* for their antifeedant effect, insecticidal effect, effect on adult emergence, fecundity and egg hatchability. The effective plant extracts identified were evaluated in cowpea grown under protected condition.

5.1. BIOEFFICACY OF AQUEOUS PLANT EXTRACTS ON *S. litura*

Studies were conducted on the antifeedant activity of the aqueous extracts of *Q. indica* flower and *S. indica* leaf against third instar larvae of *S. litura* under *in vitro* condition. Results presented in Para 4.1 revealed that aqueous extracts of *Q. indica* flower and *S. indica* leaf were not effective in inhibiting the feeding of *S. litura* larvae with less than 11 per cent mean leaf protection. The larvae exhibited antifeedant activity twenty four hours after treatment and it was absent from second day onwards.

These findings are in agreement with Ladhari *et al.* (2012). They reported antifeedant activity of stem, leaf and siliquae aqueous extracts of *Cleome arabica* L. which showed 8.8 to 21.75 per cent feeding inhibition to third instar larvae of *S. littoralis*. Among the two plant parts, *S. indica* leaf extracts exhibited more feeding inhibition to *S. litura* compared to *Q. indica* flower extracts at 15 % concentration with mean leaf protection of 10.98 and 3.88 per cent respectively (Figure 1). This could be attributed to the presence of phytochemicals present in *S. indica* leaf. NSKE 5 % exhibited significantly superior antifeedant action over the aqueous extracts of *Q. indica* flower and *S. indica* leaf. This is in agreement with Summarwar and Pandey (2013), Ravi (2013) and Razak *et al.* (2014).

No mortality was observed in *Q. indica* flower and *S. indica* leaf aqueous extracts against *S. litura* larvae even at higher concentration of 15 % three days after exposure. (Para 4.1.2). This indicates the aqueous extracts of *Q. indica* flower and *S. indica* leaf do not possess insecticidal effect. The result obtained in this study is in line with Ladhari *et al.* (2012) who reported that the aqueous extracts of *C. arabica* stem and leaves at 20 % concentration did not exhibit mortality against third instar larvae of *S. littoralis* even after three days of exposure.

An incongruity was observed with the effect of aqueous extracts of *Eupatorium triplinerve* M. Vahl reported by Kandagal and Khetagouder (2012). They reported 92 per cent mortality against fourth instar larvae of *S. litura* at 15 per cent concentration of its aqueous leaf extract. This disparity in the effectiveness of the aqueous plant extracts might be due to the variation in the active principle present in the plant.

Observation on aqueous extracts of *Q. indica* flower and *S. indica* leaf on adult emergence, fecundity and egg hatchability revealed that the aqueous extracts did not possess significant growth regulatory activities against *S. litura*. But there are several reports of aqueous extracts of certain plants with significant effect on growth regulatory activities. Kandagal and Khetagouder (2012) reported that the crude

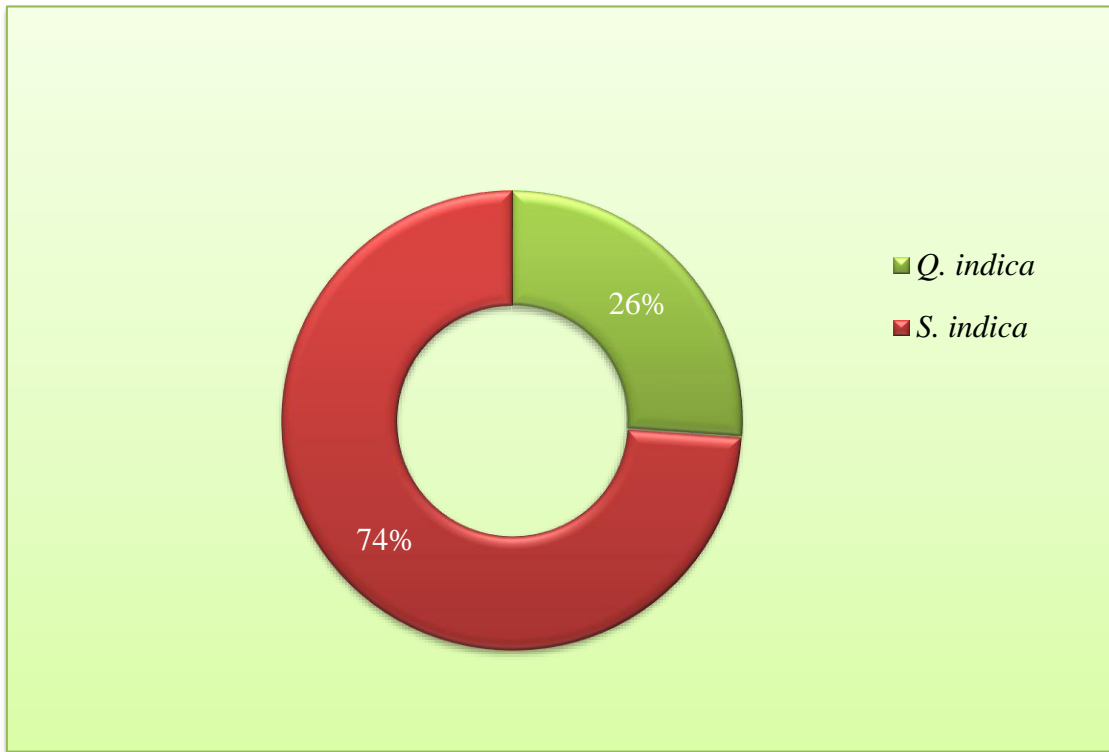


Fig. 1 Antifeedant effect of aqueous extracts of different plants (15%) on *S. litura*

aqueous leaf extracts of *E. triplinerve* at 15 per cent concentration showed 46.66 per cent mortality in prepupae, 20 per cent deformity in pupae and 25.66 per cent deformity in adults. The possible reason for the inefficacy of the aqueous extracts of *Q. indica* flower and *S. indica* leaf may be the chemical nature of the potential compounds present in plant parts, which may require more polar solvents to extract it out. So further studies were conducted using methanol and ethyl acetate solvent extracts.

5.2. BIOEFFICACY OF SOLVENT PLANT EXTRACTS ON *S. litura*

5.2.1. *Q. indica* Flower

Studies conducted on the antifeedant activity of solvent extracts of *Q. indica* flower exhibited significant effect on third instar larvae of *S. litura*. The results presented in para 4.2.1.1. showed that among the different solvent extracts of *Q. indica* flower evaluated, methanol extract at 5 per cent concentration exhibited significantly higher antifeedant action against *S. litura*. The mean leaf protection recorded in cold and soxhlet extracts of *Q. indica* flower ranged from 6.37 to 20.64 and 3.54 to 16.83 per cent respectively. The results showed that the antifeedant effect of *Q. indica* flower extracts was inferior to neem seed kernel extract (NSKE) 5 %. No previous studies have been carried out on the antifeedant effect of *Q. indica* flower extracts on *S. litura* and this clearly highlights the relevance of present investigation.

The two solvents used for the study viz., methanol with high polarity and ethyl acetate with medium polarity were found to be effective to extract the potential compounds present in *Q. indica* flower supposed to be responsible for antifeedant action. Several workers reported the antifeedant effect of methanol extracts of plants viz., *Jatropha nana* Dalz. & Gibs., *Jatropha glandulifera* Roxb. (Bhagath and Kulkarni, 2012) against *S. litura*, *C. arabica* (Ladhari *et al.*, 2012) against *S. littoralis*, *Parthenium hysterophorus* L. and *Ageratina adenophora* (Spreng.) King

& H. Rob (Yogesh *et al.*, 2013) against *S. frugiperda*. The efficacy of ethyl acetate to yield potential plant extracts was supported by Vendan *et al.* (2010), Jeyasankar *et al.* (2010) and Pavunraj *et al.* (2011), who reported the antifeedant effect of ethyl acetate extracts of *Hydnocarpus alpina* Wt., *Syzygium lineare* Wall and *Pergularia daemia* (Forssk) Choiv. against *S. litura*. The maximum antifeedant activity exhibited by the methanol extracts of *Q. indica* flower could be attributed to the polar nature of the plant compounds responsible for the antifeedant action.

Among the different methods of extraction, cold extraction (12.72 %) was found significantly superior to soxhlet extraction (9.63 %) (Figure 2). The effectiveness of cold extraction over soxhlet extraction was earlier given by Bai (1996). She reported that the cold extracts of *Thevetia nerifolia* Juss. exhibited significantly higher mean leaf protection compared to its soxhlet extracts against *Henosepilachana vigintioctopunctata* Fabricius. This may be due to the volatile nature of plant compounds responsible for antifeedant action which may loss in soxhlet extraction since heat is involved in this method.

A decreasing trend in antifeedant activity over exposure time was exhibited by the solvent extracts of *Q. indica* flower against *S. litura*. This might be due to the less persistence of active compounds responsible for antifeedant action present in solvent extracts of *Q. indica* flower. This is in line with Kleeberg and Ruch (2006), who reported the less persistence of azadirachtin due to rapid degradation by sunlight.

Results presented in Para 4.2.1.2. showed the insecticidal activity possessed by the solvent extracts of *Q. indica* flower through spraying and leaf dip method against second instar larvae of *S. litura*. Among the solvent extracts of *Q. indica* flower, maximum insecticidal activity was exhibited by the methanol extract at 5 % concentration by spraying method. The percentage larval mortality recorded in cold and soxhlet extracts of *Q. indica* flower ranged from 39.96 to 93.51 and 34.95 to 97.71 respectively. The insecticidal action exhibited by the cold extracts of *Q. indica*

flower was also assessed by dry film method and the larval mortality ranged from 9.60 to 55.02 per cent.

No larval mortality was recorded in *Q. indica* flower solvent extracts by leaf dip method. This might be due to the specific mode of action exhibited by the active ingredient present in *Q. indica* flower extracts. Wetwitayaklung *et al.* (2007) reported the acetylcholine esterase inhibition activity of methanolic extracts derived from *Q. indica* flower in electric eel. But the reports on its mode of action in insects is lacking.

The toxicity of *Q. indica* flower was supported by Song *et al.* (2014), who reported that the methanolic extracts obtained from the fruits of *Q. indica* possessed significant insecticidal activity against four Coccoidea species (*Eriococcus lagerstroemiae* Kuwana, *Ceroplastes japonicas* Green., *Crisicoccus pini* Kuwana and *Planococcus citri* Risso). The *Q. indica* extracts also possessed significant antibacterial (Kiruthika *et al.*, 2011; Kumar *et al.*, 2014; Kamar *et al.*, 2014; Sangeetha *et al.*, 2015), antifungal (Sanguri *et al.*, 2012; Sarika *et al.*, 2013) and antihelminthic (Sarma *et al.*, 2015) activities.

The results of the present investigation showed that the insecticidal action exhibited by methanol extract of *Q. indica* flower methanol extract at 5 % concentration till 48 hours after treatment was on par with quinalphos 0.05 %. This clearly highlights the potential of methanolic extracts derived from *Q. indica* flower as an effective botanical pesticide. This fact is supported by Song *et al.* (2013). They recorded 95.70 per cent nymphal mortality in the black pine bast scale, *Matsucoccus thunbergiana* Miller and Park and observed that *Q. indica* was equally effective to the insecticide, fenitrothion 50% EC against *M. thunbergiana* in field trials.

The potential of methanol and ethyl acetate to yield toxic plant extracts was reported by several workers. The methanol extracts of plants viz., *Cleome amblyocarpa* Barratte & Murb. (Shadia *et al.*, 2007), *C. arabica* (Ladhari *et al.*, 2012) and *Ailanthus altissima* Swingle (Pavela *et al.*, 2014) were found to be lethal to *S. littoralis* larvae. Jeyasankar *et al.* (2010), Vendan *et al.* (2010) and Baskar *et al.* (2011a) reported the insecticidal effect of ethyl acetate extracts of *S. lineare*, *H. alpina* and *A. tagala* against *S. litura* respectively.

Cold extraction (70.05 %) was found to be significantly superior to soxhlet extraction (64.54 %) to yield plant compounds with significant larval mortality of *S. litura* (Figure 3). This is the first report on the superiority of cold extraction over soxhlet extraction to yield solvent extracts with significant larvicidal action. This difference in efficacy in these extraction methods could be attributed to the heat involved in the extraction procedure. The heating of plant extracts in soxhlet method may cause loss of volatile compounds responsible for insecticidal action.

An increasing trend in insecticidal activity over exposure time was noticed for *Q. indica* flower extracts. This might be due to the cumulative effect of toxic components present in *Q. indica* flower on *S. litura* (Pavela, 2009).

The effect of solvent extracts of *Q. indica* flower on adult emergence, fecundity and egg hatching of *S. litura* was evaluated. The results presented in para 4.2.1.3. and 4.2.1.4. revealed that cold extracts of *Q. indica* flower did not possess significant effect on the duration of different life stages of *S. litura* and also on the number of eggs laid by the adult moth and the number of eggs hatched. This might be attributed to the physiological adaptations of *S. litura* like thick cuticle and presence of detoxifying enzymes. However, cold extracts of *Q. indica* flower exhibited significant larval and pupal mortality ranging from 8.33 to 30 per cent. The result is in line with the findings of Ray *et al.* (2012). They recorded 53.8 per cent larval mortality, 29.6 per cent pupal mortality and 22.3 per cent adult mortality at

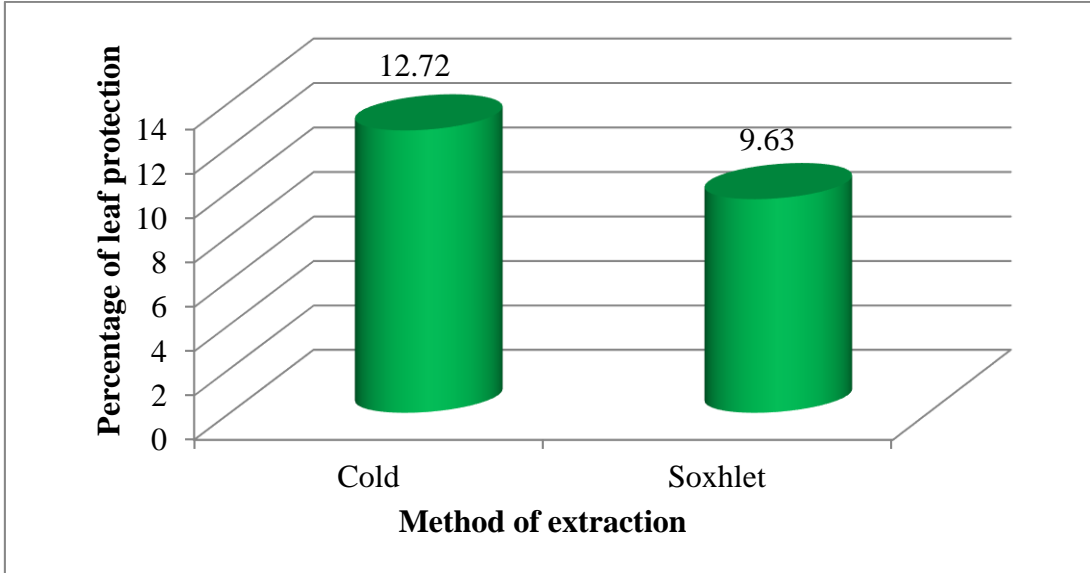


Fig. 2 Antifeedant effect of cold and soxhlet extracts of *Q. indica* flower against *S. litura*

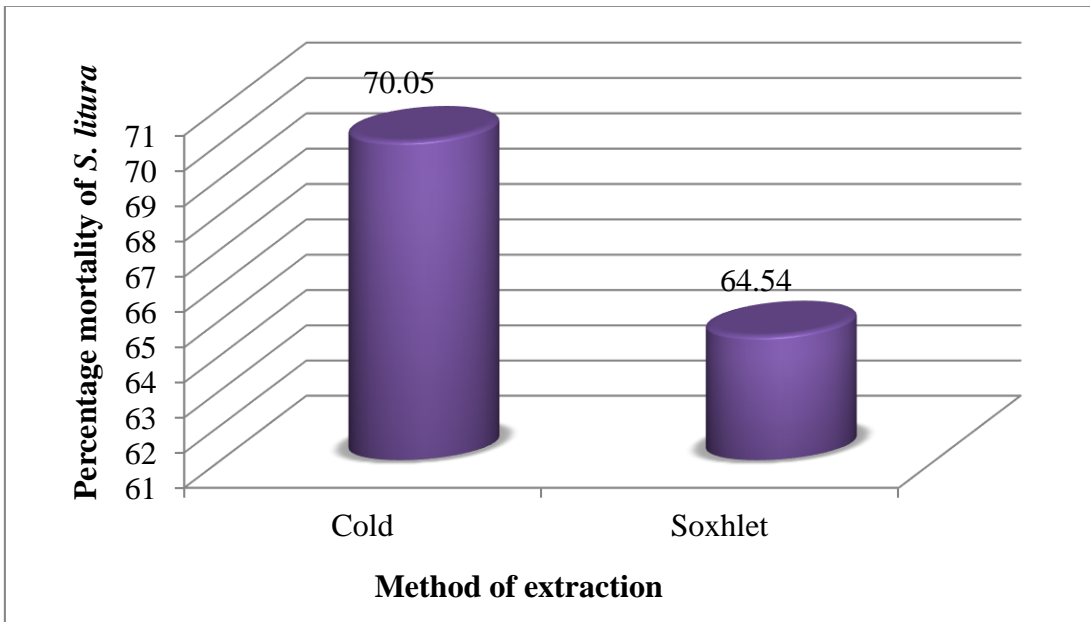


Fig. 3 Insecticidal effect of cold and soxhlet extracts of *Q. indica* flower against *S. litura*

2.5% concentration level for methanol extracts of *T. nerifolia* leaves. This fact was also supported by Pandey and Summarwar (2015) who reported the growth regulatory activities of *Ocimum sanctum* L. on *S. litura*.

The significant bioefficacy exhibited by *Q. indica* flower extracts could be attributed to its phytochemical constitution. The flower extracts of *Q. indica* contained rutin and pelargonidin-3-glucoside, while its seed oil consisted of linoleic, oleic, palmitic, stearic and arachidic acids and a neuroexcitatory amino acid quisqualic acid (Lin *et al.*, 1997). The phytochemical analysis of aqueous extracts of *Q. indica* leaf confirmed the presence of quinone, flavonoids, tannins, phenols, saponins and coumarins (Sangeetha *et al.*, 2015).

5.2.2. *S. indica* Leaf

The results presented in para 4.2.2.1 revealed the antifeedant effect of *S. indica* leaf solvent extracts on third instar larvae of *S. litura*. Among the different solvent extracts, methanol extract at higher concentration of 5 % exhibited significantly superior antifeedant action to *S. litura* larvae. Methanol extract of *Q. indica* flower 5 % recorded 7.45 to 34.61 and 8.32 to 27.73 per cent leaf protection in cold and soxhlet extraction methods respectively. The results of the present investigation showed that NSKE 5 % possessed superior antifeedant action over *S. indica* leaf extracts.

An earlier study by Govindachari *et al.* (2001) supports the finding of the present investigation. They reported that four quassinoids, indaquassin C, samaderins C, B and A isolated from the seeds and bark of *S. indica* exhibited significant antifeedant activity against *S. litura* and indaquassin C was found to be the most effective antifeedant with 62.5 per cent feeding inhibition at 5 µg/cm² concentration.

This shows the significant antifeedant activity observed in *S. indica* leaf might be due to the presence of phytochemicals responsible for feeding inhibition.

The potency of methanol extracts of plant parts to impart feeding inhibition against *S. litura* is clear from the previous reports. The methanol extracts derived from the leaves of *Tinospora crispa* (L.) Hook. F. & Thomson, *Psidium guajava* L. (Elanchezhiyan *et al.*, 2015), *Punica granatum* L., *Cassia fistula* L. and *Erythrina variegata* L. (Thushimanan *et al.*, 2016) exhibited significant antifeedant activity against fourth instar larvae of *S. litura*.

The ethyl acetate extract of *S. indica* leaf exhibited mean leaf protection in the range of 7.45 to 26.87 per cent. This showed the effectiveness of ethyl acetate to extract the active compounds present in plants and this fact is supported by several workers such as Arivoli and Tennyson (2013), Chennaiyan *et al.* (2016a) and Chennaiyan *et al.* (2016b) against *S. litura*.

The solvent extracts of *S. indica* showed a decreasing trend in antifeedant activity over exposure time against *S. litura*. The possible reason could be the instability of active compounds responsible for antifeedant action, which may easily degrade into less toxic compounds (Kleeberg and Ruch, 2006).

Among the two methods of extraction evaluated, cold extraction was found significantly superior over soxhlet extraction with mean leaf protection of 21.12 and 16.32 per cent respectively (Figure 4). This result is in accordance with Bai (1996) and also supported by the findings from the bioefficacy study of *Q. indica* flower.

The results presented in para 4.2.2.2. showed the insecticidal activity exhibited by the solvent extracts of *S. indica* leaf through spraying and leaf dip method against second instar larvae of *S. litura*. The methanol extract of *S. indica* leaf at 5 per cent concentration exhibited significantly higher larvicidal activity against *S. litura*. The percentage larval mortality recorded in cold and soxhlet

extracts of *S. indica* leaf by spraying method ranged from 33.18 to 73.55 and 24.89 to 61.77 respectively, while in leaf dip method it ranged from 15 to 41.67 and 13.33 to 36.67 respectively. The insecticidal action exhibited by the cold extracts of *S. indica* leaf was also assessed by dry film method and the larval mortality recorded ranged from 6.49 to 29.92 per cent.

The significant larval mortality (73.55 %) exhibited by *S. indica* leaf extracts against *S. litura* is reported for the first time in this study. The toxic effect of methanol extracts of *S. indica* leaf against bacteria and fungi was given by Viswanad *et al.* (2011) and Rajasree *et al.* (2012).

The two solvents selected in the present study *viz.*, methanol and ethyl acetate were proved to be effective to yield the toxic compounds from *S. indica* leaf. Similar reports were given by Rathi and Gopalakrishnan (2005), Kamaraj *et al.* (2008), Rathi and Gopalakrishnan (2010) and Chauhan *et al.* (2011) on the lethal effects of methanolic extracts of *Synedrella nodiflora* Gaertn., *Ocimum canum* L., *Lantana wightiana* Wall, and *Clerodendron inerme* (L.) Gaertn against *S. litura* respectively. The toxic effect of ethyl acetate extracts of plants *viz.*, *H. alpina* and *C. fistula* against *S. litura* and *H. armigera* was reported by Vendan *et al.* (2010) and Duraipandiyar *et al.* (2011) respectively.

S. indica leaf extracts showed increasing trend in insecticidal activity over exposure time. This could be attributed to the cumulative effect of toxic compounds and undesirable metabolites present in *S. indica* leaf extracts on *S. litura* (Pavela, 2009).

Regarding the method of application, spraying was more effective than leaf dip method for *S. indica* leaf extracts with mean values of 50.37 and 25.59 per cent larval mortality respectively (Figure 5).

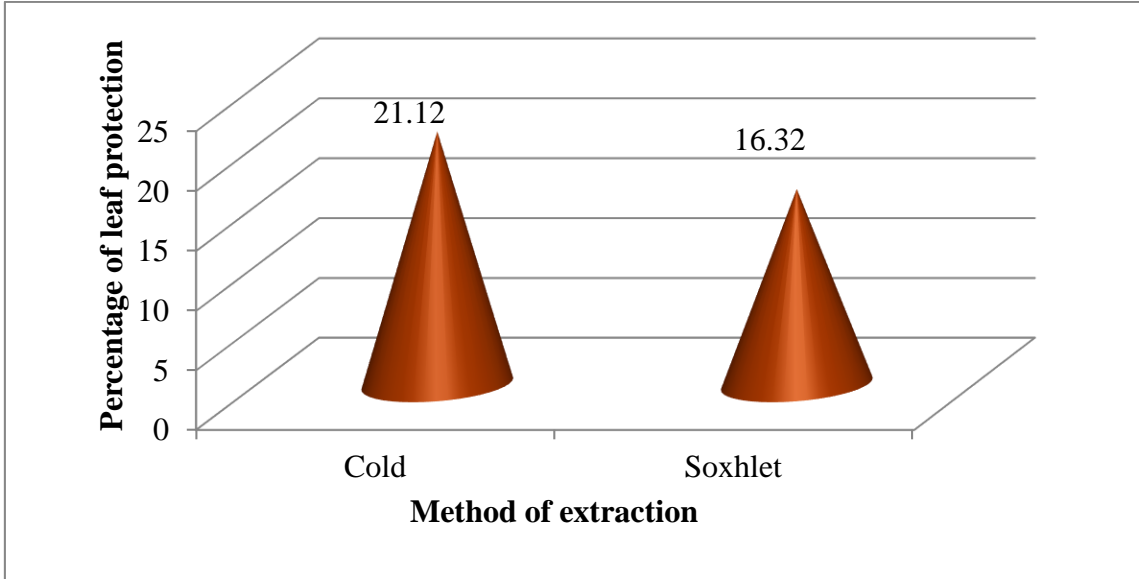


Fig. 4 Antifeedant effect of cold and soxhlet extracts of *S. indica* leaf against *S. litura*

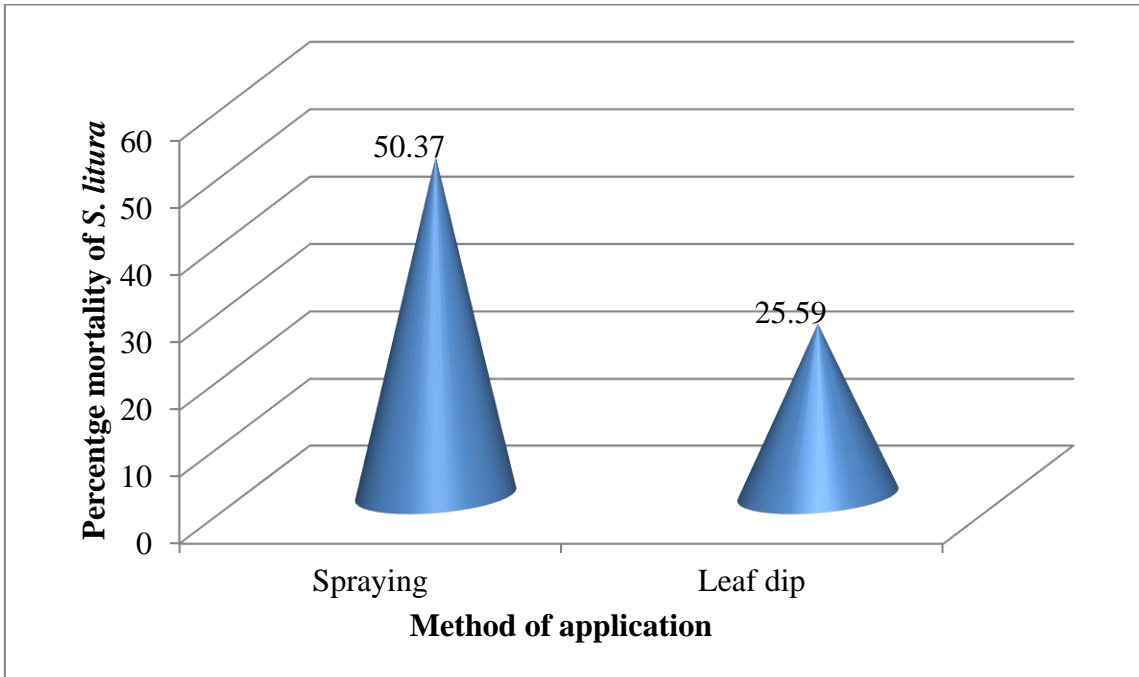


Fig. 5 Effect of different methods of application on insecticidal action of *S. indica* leaf extract against *S. litura*

The *S. indica* leaf extracts obtained through cold extraction method exhibited significantly superior larvicidal action to *S. litura* than soxhlet extraction method in both spraying (50.37 %) and leaf dip methods (26.11 %) (Figure 6 and 7). This is supported by the findings from the bioefficacy studies of *Q. indica* flower extract.

From the results of the bioefficacy study in terms of antifeedant and insecticidal activity, it was concluded that the cold extraction method was significantly superior to soxhlet extraction method. Hence cold extraction method was accepted for further studies.

The data presented on para 4.2.2.3. revealed the effect of solvent extracts of *S. indica* leaf on growth regulatory activities of *S. litura*. The results showed that the cold extracts of *S. indica* leaf did not possess significant effect on the duration of different life stages of *S. litura* and also on the number of eggs laid by the adult moth and the number of eggs hatched. However, the cold extracts of *S. indica* leaf exhibited significant larval and pupal mortality ranging from 6.67 to 16.67 and 11.67 to 31.98 per cent respectively. The growth regulatory action of *S. indica* leaf was also reported by Govindachari *et al.* (2001). They reported that the quassinoid Samaderin C isolated from the seed and bark extracts of *S. indica* increased pupal duration (9.7 days) and pupal mortality (54.1 %) of *S. litura* at 0.5 $\mu\text{g}/\text{cm}^2$ concentration.

The phytochemical constituents might be the reason for the bioefficacy exhibited by *S. indica* leaf. Phytochemical analysis of the leaf and bark extracts of *S. indica* confirmed the presence of phenolic compounds, flavonoids, alkaloids, steroids and terpenoids (Sindhu and Jose, 2015). The ethanol extracts of *S. indica* leaves contain carbohydrates, steroids, alkaloids, terpenoids, flavonoids, tannins and polyphenols, while aqueous extracts contain carbohydrates, alkaloids, flavonoids, tannins and poly phenols (Rajalakshmi and Harindran, 2013).

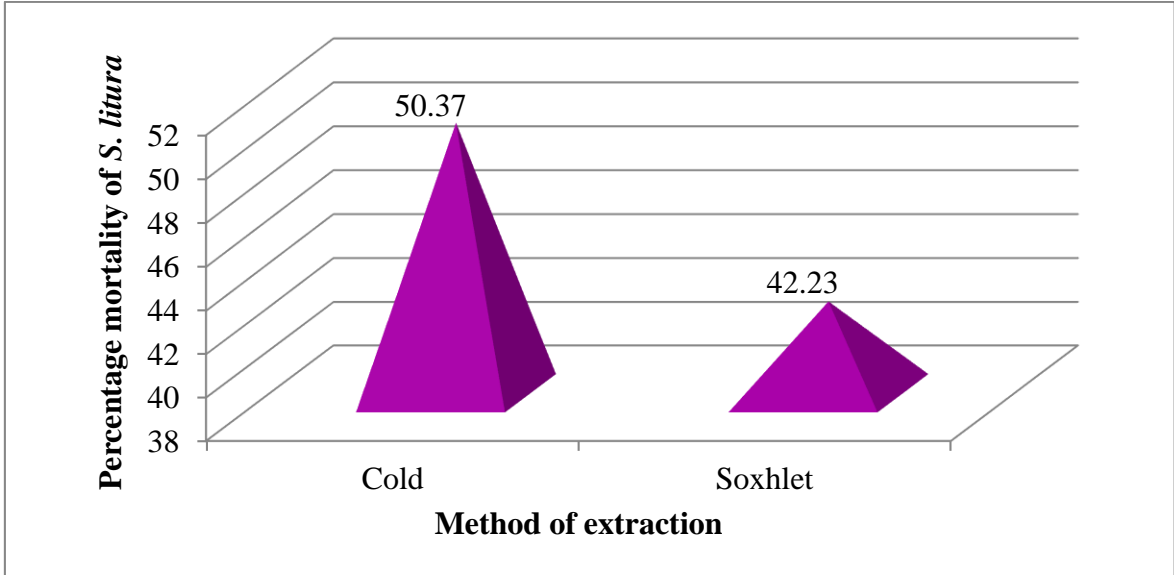


Fig. 6 Insecticidal effect of cold and soxhlet extracts of *S. indica* leaf against *S. litura* (Spraying method)

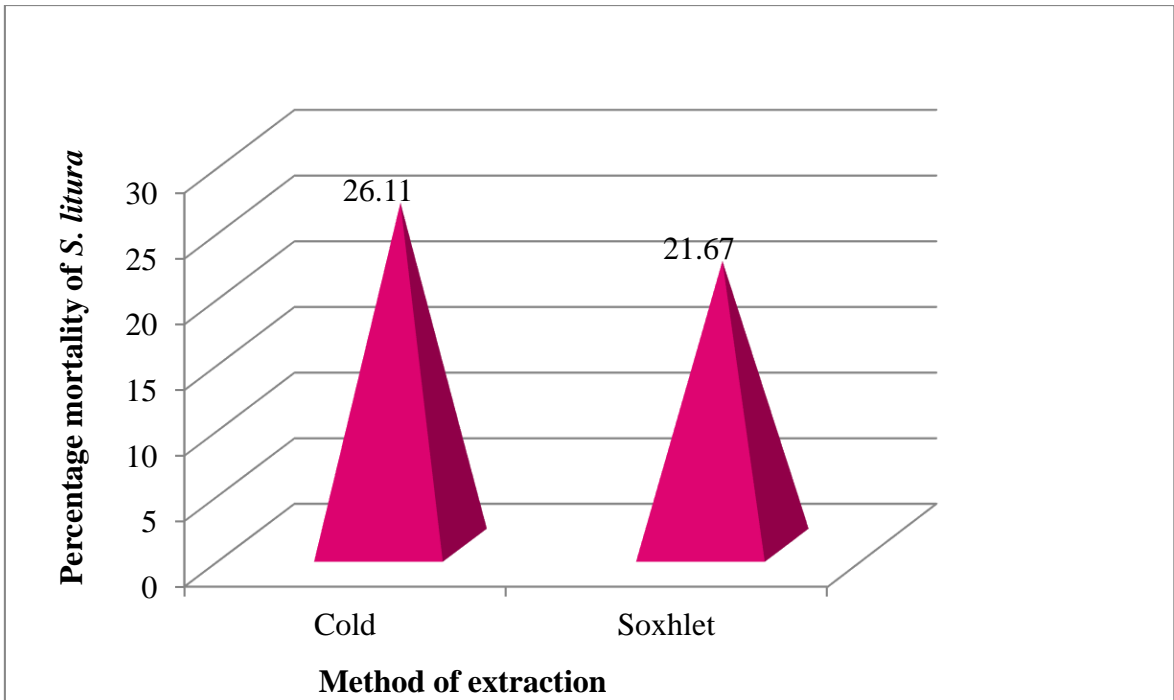


Fig. 7 Insecticidal effect of cold and soxhlet extracts of *S. indica* leaf against *S. litura* (Leaf dip method)

The results presented in para 4.2.3. showed the comparative efficacy of *Q. indica* flower and *S. indica* leaf extracts. Among the two plant parts compared, *S. indica* leaf extracts possessed more antifeedant action to *S. litura* compared to *Q. indica* flower extracts (Figure 8). The insecticidal bioassay revealed the superiority of *Q. indica* flower extracts over *S. indica* leaf extracts in both spraying and dry film method (Figure 9 and 10).

The bioefficacy study conducted under *in vitro* condition clearly highlighted the effectiveness of 5 % cold methanol extract of *Q. indica* flower and *S. indica* leaf against *S. litura*. Hence, these extracts were evaluated under polyhouse condition on cowpea to confirm their efficacy as a pest management strategy.

5.3. EFFECT OF ISOLATED PLANT EXTRACTS ON *S. litura* UNDER POLYHOUSE CONDITION

The results from para 4.3 revealed that cold methanol extract of *Q. indica* flower 5 % succeeded to reduce the *S. litura* population less than half with larval mortality of 61.45 per cent (Figure 11). The methanol extract of *S. indica* leaf 5 % also exhibited pronounced larval mortality, eventhough its insecticidal action was less compared to *Q. indica* flower extract. The larvicidal action of the plant extracts was found to be significantly inferior to quinalphos 0.05 % and superior to the biocontrol agent, *Beauveria bassiana* (Bals.-Criv.) Vuill. (Bb 5). The superiority of insecticide over plant extracts against *S. litura* was supported by Suganthi and Sakthivel (2013). They reported that quinalphos 2ml/L caused 100 per cent mortality of *S. litura*, while Pungam oil 3 % recorded 1.63 larvae/plant one week after treatment. An incongruity was observed with the effect of *B. bassiana* reported by Moorthi *et al.* (2011), Baskar *et al.* (2012b), Gupta and Kumar (2014) and Ummidi and Vadlamani (2014), who recorded significant mortality in *S. litura* larvae caused by *B. bassiana*. This disparity might be due to the difference in isolates of the entomopathogen.

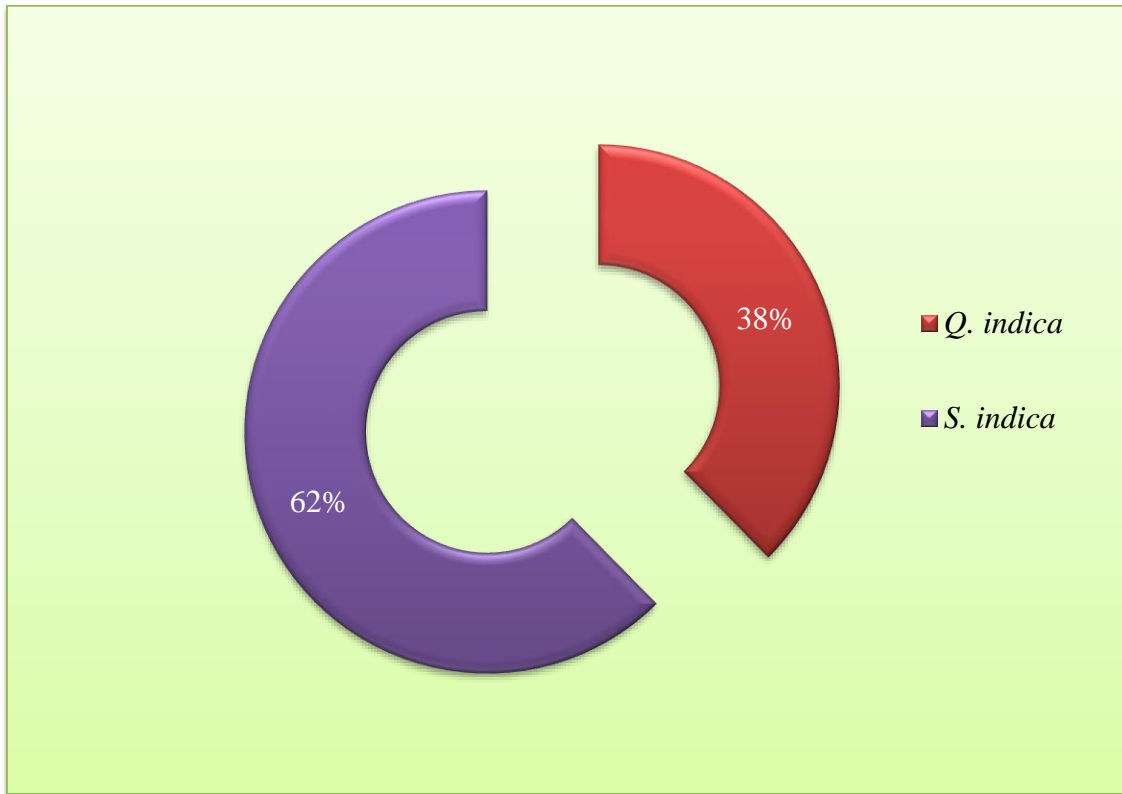


Fig. 8 Antifeedant effect of solvent extracts of different plants on *S. litura*

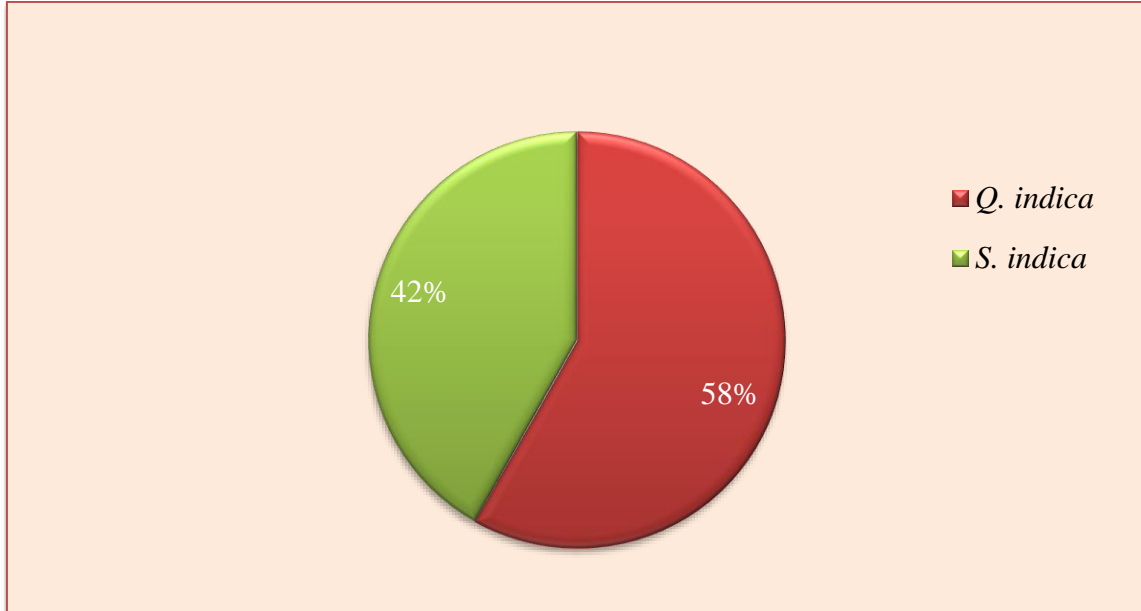


Fig. 9 Insecticidal effect of solvent extracts of different plants on *S. litura* (Spraying method)

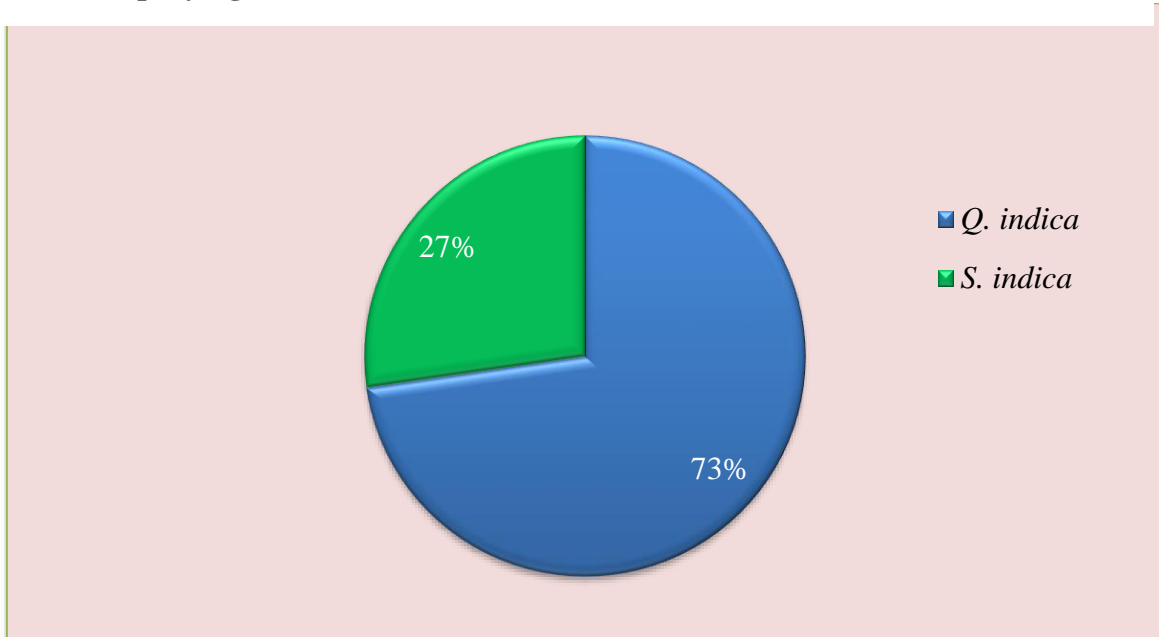


Fig. 10 Insecticidal effect of solvent extracts of different plants on *S. litura* (Dry film method)

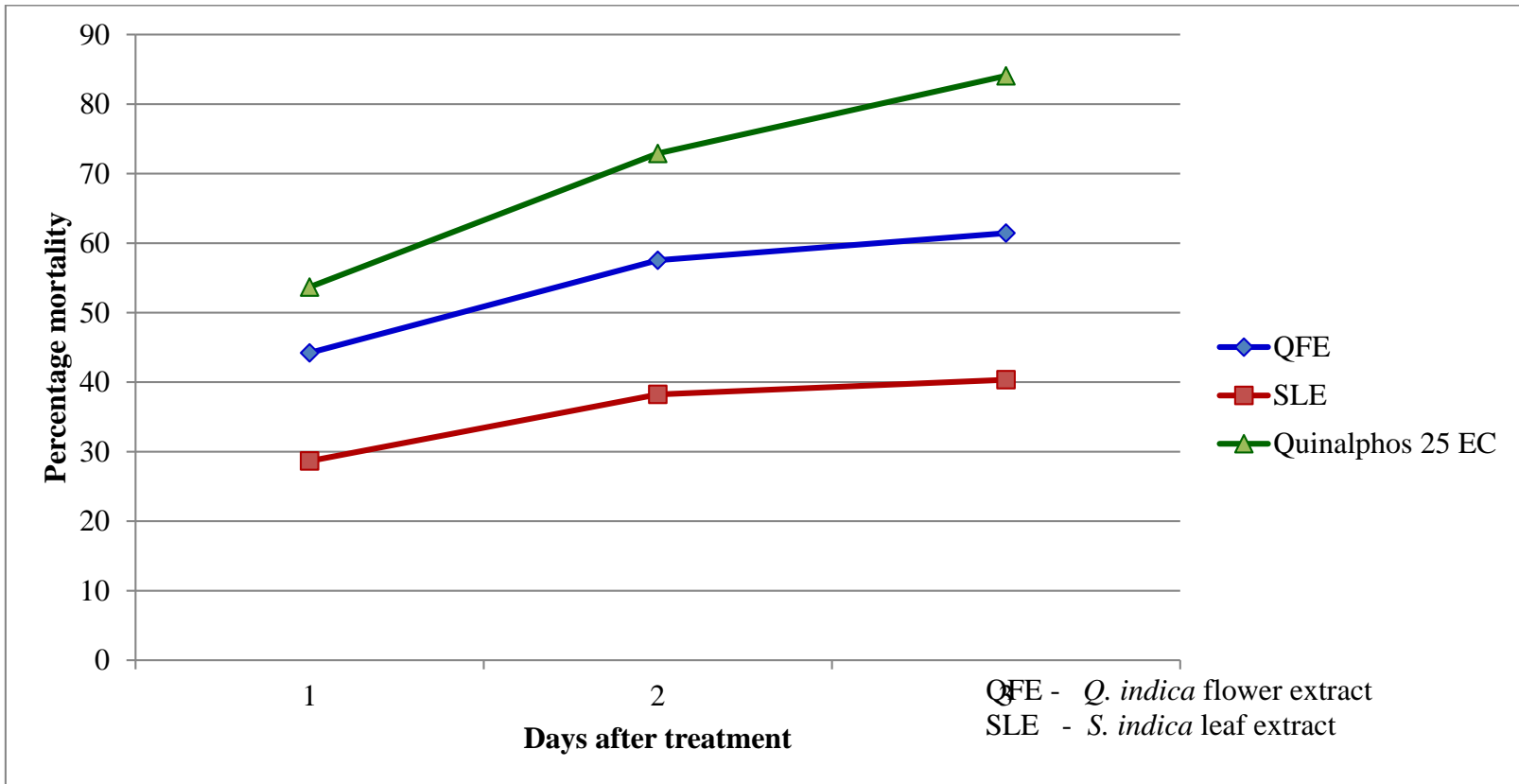


Fig. 11 Effect of plant extracts on larval mortality of *S. litura* (*in vivo*)

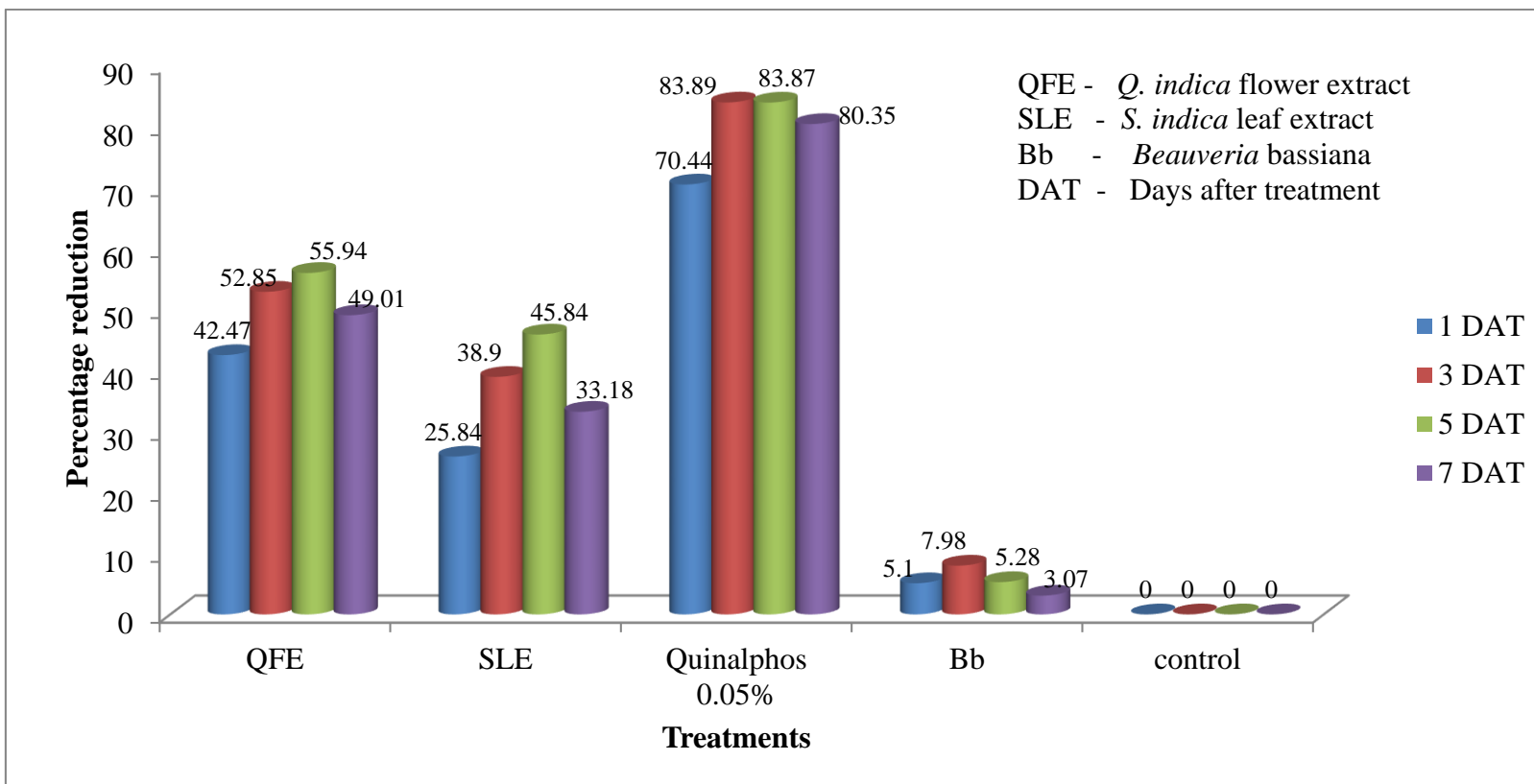


Fig. 12 Effect of plant extracts on leaf damage of cowpea infested by *S. litura* (*in vivo*)

Significant reduction in leaf area damage was observed along with larval mortality. Methanol extract of *Q. indica* flower and *S. indica* leaf at 5 % concentration recorded a reduction in leaf area damage of 42.47 and 25.84 per cent over untreated respectively (Figure 12) at one day after treatment. At one week after treatment, *Q. indica* flower extract and *S. indica* leaf extract treated leaves exhibited a reduction of 49.01 and 33.18 per cent in leaf area damage respectively.

The findings of the present investigation clearly showed that extracts of *Q. indica* flower and *S. indica* leaf were rich in potential molecules responsible for various bioactivities. Methanol extract of *Q. indica* flower was observed superior in insecticidal activity, while maximum antifeedant activity was recorded in methanol extract of *S. indica* leaf. Cold methanol extract (5 %) of both plant parts exhibited significant larvicidal action against earlier instars of *S. litura* under *in vitro* and *in vivo* conditions. Methanol extract of *Q. indica* flower at 5 per cent concentration succeeded to reduce the population of pest into less than half under polyhouse condition. This highlights the potential of *Q. indica* as an ideal substitute for chemical pesticides and it could be a solution for the pesticide residue problems prevalent under polyhouse cultivation. Isolation and identification of potential biocidal molecules from these plants could lead to the development of safer green pesticides.

Summary

6. SUMMARY

Polyhouse cultivation of vegetables for sustainability and higher returns per unit area is fast gaining importance in Kerala. The tobacco caterpillar, *Spodoptera litura* Fabricius is becoming a serious threat under protected cultivation badly affecting the production of cotton, mung bean, soya bean, cabbage, cauliflower, tomato, cucumber, rose, sweet pepper, groundnut, castor and millets. The intensive and indiscriminate use of chemical pesticides in polyhouses to manage *S. litura* resulted in residue problems as well as build up of resistance against many of the conventional insecticides such as organophosphates, carbamates, pyrethroids etc. This warrants a switch over to safer alternatives where botanicals could play a crucial role. The present investigation was undertaken to study the bioactivity of two botanicals, *Quisqualis indica* L. and *Samadera indica* Gaetrn against *S. litura*.

Aqueous extracts of *Q. indica* flower and *S. indica* leaf at 5, 10 and 15 per cent and solvent extracts viz., ethyl acetate and methanol extracts at 1.25, 2.5 and 5 per cent concentrations were screened for bioactivities. Two methods of solvent extraction viz., cold and soxhlet extraction methods were compared for antifeedant and insecticidal activities against *S. litura*.

The antifeedant activity of plant extracts was assessed by non-choice method against third instar larvae of *S. litura* and observations were recorded at 24, 48 and 72 hours after treatment. The aqueous extracts of *Q. indica* flower and *S. indica* leaf at 5, 10 and 15 per cent concentrations showed leaf protection ranging from 0 to 10.98 per cent.

The solvent extracts of *Q. indica* flower and *S. indica* leaf exhibited significant feeding inhibition to *S. litura* larvae with mean leaf protection ranging from 3.54 to 31.87 and 7.45 to 45.62 per cent respectively. Methanol extract exhibited superior antifeedant action than ethyl acetate extract for both plant parts in cold and soxhlet extraction methods. Methanol extract of *Q. indica* flower at 5 per

cent concentration recorded highest leaf protection of 20.64 and 16.83 per cent in cold and soxhlet extraction methods respectively. Leaf protection of 34.61 and 27.73 per cent was recorded in *S. indica* leaf methanol extract 5 per cent by cold and soxhlet extraction methods respectively. Among the methods of solvent extraction, cold extraction was significantly superior to soxhlet extraction giving leaf protection of 12.72 per cent in *Q. indica* flower extract and 21.12 per cent in *S. indica* leaf extract. A comparison between the two plant parts showed that *S. indica* leaf extract was significantly superior in antifeedant activity than *Q. indica* flower extract. Decreasing trend of antifeedant action with increased exposure time was noticed for both cases.

The insecticidal effect of the extracts was assessed through spraying, leaf dip and dry film methods against second instar larvae of *S. litura* and observations were recorded at 24, 48 and 72 hours after treatment. The aqueous extracts of both plant parts did not possess larvicidal action on *S. litura*, while the solvent extracts exhibited significant larval mortality. Spraying method was effective for both the plant extracts, while leaf dip method was effective for *S. indica* leaf extract only. Methanol extract exhibited superior insecticidal action than ethyl acetate extract.

Cold methanol extract of *Q. indica* flower at 5 % concentration was found highly toxic to *S. litura* and showed significant larval mortality of 70, 87 and 93 per cent at 24, 48 and 72 hours respectively after treatment in spraying method. However, 89.07 per cent larval mortality was recorded in soxhlet methanol extract 5 % at 72 hours after treatment. Ethyl acetate extracts of *Q. indica* flower also exhibited significant insecticidal action and recorded 25 to 81.72 and 21.67 to 70.08 per cent larval mortality in cold and soxhlet extraction methods respectively. A comparison between the methods of extraction revealed the superiority of cold extraction (70.05%) over soxhlet extraction (64.54 %) to yield toxic extracts of *Q. indica* flower.

The *S. indica* leaf extract, along with significant antifeedant action, exhibited pronounced insecticidal activity also. The larval mortality recorded in solvent extracts of *S. indica* leaf ranged from 11.67 to 73.55 and 6.67 to 41.67 per cent in spraying and leaf dip method respectively. Among the two methods of extraction, cold extraction method proved as the effective method of extraction for *S. indica* leaf in both spraying (50.37 %) and leaf dip method (26.11 %). The cold methanol extract 5 % of *S. indica* leaf was found to be highly toxic to *S. litura* larvae with 38, 62 and 74 per cent mortality at 24, 48 and 72 hours after treatment in spraying method. Assessment of insecticidal action by leaf dip method recorded 17, 27 and 42 per cent larval mortality at 24, 48 and 72 hours after treatment. The ethyl acetate extract 5 % of *S. indica* leaf also showed significant larvicidal action against *S. litura* with percentage mortality ranging from 28.33 to 56.68 and 8.33 to 26.67 in spraying and leaf dip method respectively. A comparison made between the plant parts showed that *Q. indica* flower extract (70.10 %) exhibited higher insecticidal activity than *S. indica* leaf extract (50.37 %).

The effect of the cold extracts of *Q. indica* flower and *S. indica* leaf was assessed through dry film method. Larval mortality of 36.05 and 13.52 percent was observed in *Q. indica* flower and *S. indica* leaf extracts respectively, suggesting the contact toxicity of plant extracts.

Effect of plant extracts on adult emergence of *S. litura* showed that the aqueous and solvent extracts of both plants did not have any influence on the duration of different life stages of *S. litura*. The cold extracts of *Q. indica* flower and *S. indica* leaf exhibited significant larval and pupal mortality ranging from 8.33 to 30 % and 11.67 to 31.98 % respectively.

Assessment of potential of the selected treatments, methanol extract (5 %) of *Q. indica* flower and *S. indica* leaf was done under polyhouse condition on cowpea. The efficacy was compared with quinalphos 25 EC 0.05 % and biocontrol agent, *Beauveria bassiana* (Bals.-Criv.) Vuill. (Bb 5) 20 g/L. Maximum larval mortality of

84.07% was observed in quinalphos 0.05 % followed by 5 % methanol extract of *Q. indica* flower (61.45 %) and *S. indica* leaf (40.35 %). No larval mortality was observed in *B. bassiana* treated plants. The larvicidal action of the plant extracts was found significantly inferior to quinalphos 0.05 % and superior to *B. bassiana*. The leaf area damage in 5 per cent methanol extract of *Q. indica* flower and *S. indica* leaf was 48.56 and 63.64 per cent respectively. A reduction in leaf area damage of 42.47 and 25.84 per cent over untreated was recorded in methanol extract 5 per cent of *Q. indica* flower and *S. indica* leaf treated leaves respectively.

The study revealed that *Q. indica* flower and *S. indica* leaf extracts possessed significant effect on *S. litura*. The leaf extracts of *S. indica* were superior to *Q. indica* flower extracts to impart feeding inhibition to *S. litura* larvae, while significantly higher percentage larval mortality was recorded in *Q. indica* flower extract compared to *S. indica* leaf extract. Cold methanol extract 5 % of *Q. indica* flower showed 61 per cent larval mortality of *S. litura* under polyhouse condition. Hence, this plant can be effectively included as a component in integrated pest management. Moreover, future studies can be concentrated on the isolation and identification of the potential molecules present in these plant parts, which could possibly lead to the development of safe pesticides.

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7. REFERENCES

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**BIOEFFICACY OF *Quisqualis indica* L. AND *Samadera indica*
GAETRN. AGAINST TOBACCO CATERPILLAR,
Spodoptera litura FABRICIUS (LEPIDOPTERA: NOCTUIDAE)
IN POLYHOUSE CONDITION**

by

ANUSREE S. S.

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Abstract of the thesis

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VELLAYANI, THIRUVANANTHAPURAM-695522

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ABSTRACT

An investigation entitled “Bioefficacy of *Quisqualis indica* L. and *Samadera indica* Gaetrn. against tobacco caterpillar, *Spodoptera litura* Fabricius (Lepidoptera: Noctuidae) in polyhouse condition” was conducted at College of Agriculture, Vellayani during 2014-16. The main objectives were to evaluate the effect of aqueous and solvent extracts of *Q. indica* flower and *S. indica* leaf on behavioural and physiological changes of *S. litura* and to test the potential of the selected plant extracts against *S. litura* infesting cowpea under polyhouse condition.

Aqueous and solvent extracts of *Q. indica* flower and *S. indica* leaf were tested for antifeedant and insecticidal action against larvae of *S. litura* under *in vitro* condition. Antifeedant activity of aqueous extracts of *Q. indica* flower and *S. indica* leaf at 5, 10 and 15 % concentrations showed percentage leaf protection ranging from 0 to 10.98. Solvent extracts *viz.*, ethyl acetate and methanol extracts of *Q. indica* flower and *S. indica* leaf at 1.25, 2.5 and 5 % concentrations showed percentage leaf protection ranging from 13.23 to 45.62. Maximum antifeedant activity (45.62 %) was exhibited by methanol extract 5 % of *S. indica* leaf at 24 hours after treatment.

The extracts obtained through cold and soxhlet extraction methods were compared for the antifeedant activity against *S. litura*. Cold extraction was significantly superior to soxhlet extraction giving leaf protection of 12.72 % for *Q. indica* flower extract and 21.12 % for *S. indica* leaf extract. Decreasing trend of antifeedant action with increased exposure time was noticed for both the plants.

The insecticidal effect of the extracts was assessed through two application methods, spraying and leaf dip method. Spraying method was effective for both the plant extracts, while leaf dip method was effective for *S. indica* leaf extract only. Cold extract of *Q. indica* flower 5% with methanol was found to be highly toxic to *S. litura* larvae with maximum percentage mortality of 93.51 in spraying method. Methanol cold extract 5 % of *S. indica* leaf exhibited 73.55 % mortality in spraying

method and 41.67 % mortality in leaf dip method. Cold extraction method was found to be significantly superior for both *Q. indica* flower (70.05 %) and *S. indica* leaf (50.37 %) than Soxhlet extraction in spraying method. The insecticidal effect assessed through dry film method showed that cold extracts of *Q. indica* flower and *S. indica* leaf exhibited larval mortality of 36.05 % and 13.52 % respectively. An increase in mortality with increased exposure was observed for both plant extracts.

Effect on adult emergence of *S. litura* (deformation and mortality of larvae, pupae and adults, time taken for pupation, pupal duration, pupal weight and adult longevity) showed that the aqueous and solvent extracts of both plants did not have any influence on larvae, pupae and adults. Cold extracts of *Q. indica* flower and *S. indica* leaf exhibited significant larval and pupal mortality ranging from 8.33 to 30 % and 11.67 to 31.98 % respectively. Effect on fecundity and egg hatchability revealed that the plant extracts did not possess significant effect on number of eggs laid and number of eggs hatched.

To assess the potential of the selected treatments, methanol extract (5 %) of *Q. indica* flower and *S. indica* leaf, a pot culture experiment was done under polyhouse condition on cowpea. It was compared with quinalphos 25 EC 0.05 % and biocontrol agent, *Beauveria bassiana* (Bb 5) 20 g/L. The percentage leaf area damage in 5 % methanol extract of *Q. indica* flower and *S. indica* leaf was 48.56 and 63.64 respectively. Maximum larval mortality of 84.07% was observed in quinalphos 0.05 % followed by 5 % methanol extract of *Q. indica* flower (61.45 %) and *S. indica* leaf (40.35 %).

From the above study it is concluded that methanol cold extract (5 %) of flowers of *Q. indica* and leaves of *S. indica* have insecticidal action against earlier instars of *S. litura*. These plants can be exploited for formulating potential green pesticides.