

Original Research Article

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Insecticidal Efficacy of Methanol Extract of Leaves of *Saraca indica* L. against Four Insect Pests Causing Severe Damage to Stored Grains

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ABSTRACT

The methanol extract from leaves of *Saraca indica* was investigated to control insect infestation of stored food commodities viz. maize, rice, cowpea and wheat flour during from *Ostrinia nubilalis*, *Spodoptera littoralis*, *Callosobruchus maculatus* and *Tribolium confusum*. After introducing the test insects, stored food samples were fumigated with methanol extract from leaves of *Saraca indica* at 500 µg/mL (ppm). The methanol extract significantly enhanced feeding deterrence in insects and reduced the grain damage as well as weight loss in fumigated food samples infested with all insects. The methanol extract at different doses significantly reduced oviposition and adult emergence of these insect pests. Regression analysis of data on individuals in treated food samples confirmed that significant reduction of oviposition and adult emergence of *insect pests* decreased with increase in doses. All the four insect pests caused minimum damage to food commodities when fumigated with methanol extract of leaves of *Saraca indica*. The methanol leaf extract of *Saraca indica* exhibited as botanical fumigant in protection of stored maize, rice, cowpea and wheat by enhancing feeding deterrence and reducing grain damage as well as weight loss of *Ostrinus nubilalsis*, *Spodoptera littoralis*, *Callosobruchus maculatus* and *Tribolium confusum*. The ovicidal activity of the methanol extract of leaves of *Saraca indica* against four insect pests reveals that no hatching of larvae were observed till 24 hours. Hatching of larvae were observed from 24 hours to 72 hours. No further hatching was recorded after 72 hour of treatment. Therefore LC50 value of ovicidal activity was recorded at 72 hours of exposure period. At 10 ppm and after 72 hours of incubation *O. nubilalsis*, *S. littoralis*, *C. maculatus* and *T. confusum* the hatching percentage was 80.75%, 78.51%, 76.25% and 78.85% respectively. The hatching of larvae showed a decreasing trend on increasing the concentration of methanol extract of *Saraca indica*. At 1000ppm concentration of leaf extract and after 72 hrs of incubation the hatching percentage of *O. nubilalsis*, *S. littoralis*, *C. maculatus* and *T. confusum* was 26.15%, 21.16%, 22.18% and 24.15% respectively. The larvicidal activity of methanol extract of leaves of *Saraca indica* against four insect pests viz. *Ostrinus nubilalsis*, *Spodoptera littoralis*, *Callosobruchus maculatus* and *Tribolium confusum* was found to be directly related to the exposure time and concentration of the methanol extract.

Keywords

Saraca indica, Leaf extract, *Ostrinia nubilalis*, *Spodoptera littoralis*, *Callosobruchus maculatus*, *Tribolium confusum*, LC50, Larvicide, Ovicide

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Introduction

Saraca indica L. (Ashok) is a small evergreen tree of sub family Caesalpinoideae of family Leguminosae. The leaves are paripinnate, oblong and rigidly sub- coriaceous with 6- 7 leaflets (Ali, 2008). This tree has orange coloured flowers with a beautiful aroma, 7- 8 stamens are found in flower and fruits are smooth, leathery and flat pods including 6- 8 seeds inside (Jain, 1968). Bark of this tree is rich in tannins, flavonoids, steroids, volatile oil, glycosides, and various steroidal glycosides. Leaves contain various carbohydrates, tannins, gallic acid and gallic acid. Flowers are rich in sarcasin, sarcadin, waxy substances, proteins, carbohydrates and steroids. Seeds of this plant contain various fatty acids like oleic, linoleic, palmitic and stearic acid.

Ashok tree has been an integral part of Indian history. It is commonly called a tree which is important to decrease our sorrows. It has got religious significance and is also worshipped by some people in parts of India. It has a number of medicinal properties hence used by physicians since centuries in Unani system of medicine along with Ayurveda (Kokate *et al.*, 2007). It is primarily used for the management of female reproductive problems. Married women in India are known to eat Ashoka flower buds as a ritual to invoke deities for child protection as well as gynecological problems. Women suffering from menorrhagia drink a decoction on an empty stomach in the morning, which is prepared from the bark of Ashoka in water in combination with other herbs such as *Terminalia chebula* and *Coriandrum sativum* (Begum *et al.*, 2014).

In leucorrhoea, the decoction of Ashoka bark in water and milk after evaporation of water is consumed by women. In India, Srilanka, Bangladesh and Pakistan Ashoka bark is used by womenfolk in treating menorrhagia,

menstrual and uterine disorders (Mishra *et al.*, 2013; Mollik *et al.*, 2010).

Saraca indica is a rain- forest tree. It is native of Asia and South America. It is originally distributed in the central areas of Deccan plateau. It is also found in Western Ghats of the Indian subcontinent. It is also widely distributed in the center and the Eastern Himalayas and in the hills of Khasi, Garo and available in West Bengal. It is common to all parts of Indian and other countries. In India it is easily available in West Bengal, Kerala, Maharashtra, Andhra Pradesh and Meghalaya (Kokate *et al.*, 2007; Prajapati *et al.*, 2003).

This plant has cooling properties. It is very useful for the body to bring down excessive heat in the organs due to fatigue or hormonal imbalance. It helps to regulate blood composition and stabilize blood circulation making it optimally available to all the body parts. Its pain relieving action can help relieve painful dysmenorrhea, swelling and pain at any site of the body. In females it is very commonly used to regularize hormones and menstrual cycles. It improves the strength and stamina in young females having menstrual irregularities such as dysmenorrhea and leucorrhoea. Many at times a combination of *Aloe vera* and Ashok is given to females to improve their reproductive health and blood condition. Anemia which is very common health problem in females is also recovered with the right combination of herbs along with Ashok derivatives. It not only works on uterine structures but also helps to cleanse the system so that any kind of microbial infestation that may be causing leucorrhoea and other associated infections in the reproductive organs in females can be checked.

Ashok is also a cardiac tonic that can act as a supportive therapy for people suffering from hypertension, circulatory problems, edema, congestive heart failure etc. Its bark has

natural detoxification properties which make it very useful to improve skin complexion and keep the body free from toxins inside out. Its natural cleansing properties can help the body stay toxin free. When the body has a lot of toxic load free radicals are produced. These free radicals then start damaging the body cells and all signs of ageing, disease and malfunctions are produced. For general pitta aggravated states also, Ashok bark acts as a coolant and helps to relieve thirst, excessive burning sensation, anger, emaciation, sweating etc. These are all common signs of pitta aggravation which can be relieved with the use of Ashok bark in different ways. It also has some digestive properties. Common problems of digestion like bloating, flatulence, burping, colicky pain in abdomen, ascites etc. can be relieved with the use of Ashok. It is not exactly a direct indication of the herb but it does help because all diseases have root from a malfunctioning gut and digestive system overtime.

Cereals and pulses have great biological and nutritional value in developing countries, are lost upto 20-60 per cent by storage insect pests during storage (Arthur and Throne 2003; Babu *et al.*, 2003; Shaaya *et al.*, 1997). Post-harvest deterioration causes economic losses due to obvious decay and adverse changes in the odour, taste, appearance and nutrition value (Phillips and Burkholder 1984; Mondal and Port 1994; Arlian *et al.*, 1996). In addition, the arthropods transfer bacteria and microscopic fungi of pathogen importance on stick on their bodies or disseminate them via faeces (Wilbur and Mills 1978, Hubert *et al.*, 2004). During recent years considerable attention has been paid towards exploitation of plant materials in protection of food commodities from insect infestations. Extracts of some plant species viz. *Lantana camara* (Saxena *et al.*, 1992), *Illicium verum* (Ho *et al.*, 1995), *Tithonia diversifolia* (Adedire and Akinneye, 2004) have been reported to possess strong

insecticidal activity against different storage insects. Plant derived products namely, azadirachtin from *Azadirachta indica*, pyrethrin from *Chrysanthemum cinerariaefolium*, carvone from *Carum carvi* and allyl isothiocyanate from mustard and horseradish oil have been received global attention due to their pesticidal properties and potential to protect several food commodities (Hartmans *et al.*, 1995; Ward, 1998; Varma and Dubey, 1999; Athanassiou *et al.*, 2005). Essential oils produced by different plant genera have been reported to be biologically active and are endowed with insecticidal, antimicrobial and bio regulatory properties (Mishra and Dubey, 1994; Varma and Dubey, 1999; Dubey *et al.*, 2004; Holley and Patel, 2005). The volatility and biodegradability of flavour compounds of angiosperm will be advantageous if they are developed as pesticide insecticide (French, 1985). There may be least chance of residual toxicity by treatment of food commodities with volatile substances of higher plant origin.

In the present investigation the larvicidal activity of methanol extract of leaves of *Saraca indica* (L.) was assayed on the Lepidopteran insects viz. *Ostrinia nubilalis* and *Spodoptera littoralis* and Coleopteran insects viz. *Callosobruchus maculatus* and *Tribolium confusum*. These insects are known to severely damage crops like maize (*Zea mays*), rice (*Oryza sativa*), cotton (*Gossypium herbaceum*), tobacco (*Nicotiana tabaccum*), soybean (*Glycine max*), cowpea (*Vigna unguiculata*), rice (*Oryza sativa*), wheat (*Triticum aestivum*) and wheat flour on storage.

Materials and Methods

Rearing and maintaining insect pests

The lepidopteran insects like *Ostrinia nubilalsis* (European corn borer) and

Spodoptera littoralis (Cotton leaf worm) were reared and maintained on maize and rice grains respectively. Similarly, Coleopteran insects *Callosobruchus maculatus* (Cowpea seed beetle) and *Tribolium confusum* (confused flour beetle) were reared and maintained on cowpea (*Vigna unguiculata*) grains and wheat flour respectively. The insects were maintained and reared at $27 \pm 2^{\circ}\text{C}$ and relative humidity (RH) of $80 \pm 5\%$ following the methods suggested by Babu *et al.*, (2003), Jenkins *et al.*, (2003), Perez-Mendoza *et al.*, (2004) and Arivoli *et al.*, (2011).

Forty adult insects were released separately in 200 g of commodities (maize/rice/cowpea/wheat flour) in plastic containers covered by muslin cloth. After 24 hours, adult insects were removed and the commodities were incubated in a temperature/humidity controlled cabinet ($27 \pm 2^{\circ}\text{C}$ and RH $80 \pm 5\%$) in darkness to obtain same aged insects. Adult insects were 2- 4 days old when used in the bioassays. Larvae were fed on larval food (powdered dog biscuit and yeast in the ratio 3:1) and adult mosquitoes on 10 per cent glucose solution. Pupae were transferred to a disposable cup and it is kept inside the cage.

Preparation of methanol extract

The leaves of *Saraca indica* were collected from the local garden of Ganga Devi College, Patna. Leaves were then sliced and chopped into small pieces, dried under shade and powdered with the help of a hand grinder, weighed and placed in separate conical flasks to add solvents. Methyl alcohol (CH_3OH) (Merck, Germany) was used to prepare methanol extract from leaves. The dried plant sample was soaked with absolute methanol under reflux condition for the methanolic extract preparation. The sample was homogenized with extraction buffer. The supernatant was collected after three rounds of

extraction. The solvent was evaporated under reduced pressure in a rotary evaporator at 40°C . To this thick paste colloidal silicon dioxide was added and dried in vacuum tube dryer. The extract was then stored in deep freezer at -20°C until further test.

Fumigation of maize, rice, cowpea and wheat flour by methanol extract *Saraca indica*

The methanol extract of leaves of *Saraca indica* was used to fumigate the maize, rice, cowpea and wheat grains samples separately by the method adapted by Shaaya *et al.*, (1997) and Kumar *et al.*, (2007). Five hundred gram of samples of each was kept separately in closed plastic containers (35 cm diameter x 16 cm). Care was taken to use uninfested freshly harvested grains and freshly prepared wheat flour. Twenty five individuals of each insect species viz. *Ostrinia nubilalis*, *Spodoptera littoralis*, *Callosobruchus maculatus* and *Tribolium confusum* of mixed sex were introduced in the containers. Requisite amount of the methanol extract of *Saraca indica* was introduced separately in the plastic containers of each of the varieties by soaking in cotton swab so as to procure concentration of 500 ppm. The containers were made airtight.

The grains and flour inoculated with the test insects without methanol extract served as controls. After 24 months of storage at laboratory conditions in a temperature/humidity control cabinet ($27 \pm 2^{\circ}\text{C}$ and RH $80 \pm 5\%$) in darkness the efficacy of *Saraca indica* extract on insect infestation was determined by calculating grain damage (%), weight loss (%) and feeding deterrence (%) of treated and control sets.

The grain damage was determined by counting feeding injuries and emergence holes on the surface of the grains. The weight loss (%) of

samples in the treated and control sets was calculated by fresh weight basis using the formula suggested by Parkin (1956).

$$\text{Weight loss (\%)} = \frac{\text{WI} - \text{W}}{\text{W}} \times 100$$

Where WI and W represents the weight of grains before and after the experiment, respectively

Feeding deterrence was calculated using the feeding deterrent index following Isman (1990):

$$\text{Feeding deterrent index (FDI) [\%]} = \frac{\text{C} - \text{T}}{\text{C} + \text{T}} \times 100$$

Where C and T is the weight loss in the controls and in the fumigated sets, respectively.

Insects Ovicidal Bioassay

The ovicidal bioassay was performed according to the method described by Tennyson *et al.*, (2011) and Puspanathan *et al.*, (2006) with little modifications. For the ovicidal bioassay, 50 eggs of each species were transferred to each of the three replicates of each concentration.

Eggs were exposed to the DMSO and water was treated as control. For determination of LC50 values, a wide numbers of concentrations of the oils were tested against the target species. The number of eggs hatched in control and treatments were recorded and the percentage of ovicidal activity was calculated by the following formula-

$$\text{Percent Ovicidal activity} = \frac{\text{Percent of eggs hatched in control} - \text{Percent of eggs hatched in treated}}{\text{Percent of eggs hatched in control}} \times 100$$

Larvicidal bioassay

Screening of the efficacy of methanol extract leaves of *Saraca indica* was done by performing bioassay studies against different developmental stages of *Ostrinia nubilalis*, *Spodoptera littoralis*, *Callosobruchus maculatus* and *Tribolium confusum*. The larvicidal activity of individual extract was assayed following the technique described earlier by Tong *et al.*, (2013) and WHO guidelines (2005). According to the WHO protocol for larvicides testing for laboratory testing, batches of 20 numbers of healthy 4th instars larvae of each species were transferred to the disposable glasses with the depth between 5-10cm having 100ml of water. A series of concentration from the 1000ppm to 10 ppm were used to examine the larvicidal toxicity of the extract. The LC50 values are recorded after 24, 48 and 72 hour exposure. Each concentration was assayed in triplicate along with one negative control group in water and one positive control group with the DMSO. If the pupation occurred in the exposure time or more than 10% larva was died in the control group, the test was repeated. From the data, LC50 values were determined by probit analysis (SPSS 16).

Statistical analysis

All the experiments were conducted in replicates of three and data was recorded as mean value \pm SE. The statistical analysis was performed by one way analysis of variance and means were compared by least significance difference test ($P < 0.05$) using the SPSS statistical software package (SPSS, ver. 10.0; Chicago. IL, USA). The correlation coefficient was calculated between dose-mortality, dose-oviposition, dose-adult emergence, mortality-oviposition and oviposition-adult emergence using software Origin (Origin 6.0 Northampton, MA, USA). Further, the data was subjected to Student's 't'

test to analyzed the effect of *Saraca indica* extract on grain damage as well as weight loss of grains with control.

The data were corrected for the mortalities with the help of Abbott correction factor and were subjected to probit analysis using SPSS software to estimate LC50 values of effective methanol extract against the mosquito. Again, if mortality in the controls was found above 5%, results with the treated samples were corrected using Abbott's formula (Abbott, 1925). The results obtained have been presented in Table 1–14.

The mortality (%) was corrected using Abbott's formula (1925):

$$\text{Pr} = \frac{\text{Po} - \text{Pc}}{100 - \text{Pc}} \times 100$$

Where, Pr = Corrected mortality (%), Po = Observed mortality (%), Pc = Mortality in the control (%). The results obtained have been presented in Table 1 to 14.

Results and Discussion

The fumigation efficacy of methanol extract of leaves of *Saraca indica* against four insect pests has been presented in Table 1 to 4. From the result it is evident that the methanol extract of leaves of *Saraca indica* significantly protected all the four food commodities viz. Maize, Rice, Cowpea and Wheat on storage from insect pests such as *Ostrinus nubilalsis*, *Spodoptera littoralis*, *Callosobruchus maculatus* and *Tribolium confusum* ($p < 0.05$; LSD) (Table 1-4). The feeding deterrent index (FDI) of *O. nubilalsis*, *S. littoralis*, *C. maculatus* and *T. confusum* on Maize infestation was 87.45%, 88.35%, 89.35% and 88.65% respectively; on Rice infestation was 88.55%, 89.654%, 90.25% and 93.15% respectively; on Cowpea infestation was

90.45%, 91.35%, 90.50% and 91.05% respectively and on Wheat infestation was 93.25%, 87.21%, 93.45% and 87.35% respectively. There was 100% damage of all the four food commodities on storage in control experiment i.e. not fumigated with methanol extract of *Saraca indica*. In control experiment there was maximum loss in weight of food commodities. In case of Maize the loss was 47.25% to 48.45%; of Rice it was 50.25 to 62.55%; of Cowpea 28.45% to 58.90% and of Wheat 47.15% to 58.555% (Table 1- 4). All the four insect pests caused minimum damage to food commodities when fumigated with methanol extract of leaves of *Saraca indica*. The damage to maize grains by *O. nubilalsis*, *Spodoptera littoralis*, *C. Callosobruchus maculatus* and *Tribolium confusum* was 10.25%, 10.35% 9.75% and 10.45% respectively (Table 1). Similarly, the damage to rice grains by *O. nubilalsis*, *Spodoptera littoralis*, *Callosobruchus maculatus* and *Tribolium confusum* was 7.25%, 8.25%, 6.35% and 3.68% respectively (Table 2). A more or less similar pattern of damage was noticed in cowpea and wheat by these insect pests after fumigation with methanol extract of leaves of *Saraca indica* (Table 3 and 4).

In the present study the methanol leaf extract of *Saraca indica* exhibited as botanical fumigant in protection of stored maize, rice, cowpea and wheat by enhancing feeding deterrence and reducing grain damage as well as weight loss of *Ostrinus nubilalsis*, *Spodoptera littoralis*, *Callosobruchus maculatus* and *Tribolium confusum*. The findings are in accordance with Kumar *et al.*, (2007) and Varma and Dubey (2001) who investigated that essential oil of *Cymbopogon martinii*, *Caesulia axillaris* and *Mentha arvensis* protected stored gram and wheat from *C. chinensis*, *S. oryzae* and *T. castaneum* for first 12 months of storage. In the present investigation the shelf life of the *Saraca indica* extract in protection of insect

infestation was 24 months thus more than the extract reported earlier. Plant extracts and essential oils are known to possess repellent, ovicidal and insecticidal activities against various stored grain insects (Hill and Schoonhoven 1981; Desmarchelier 1994). The present findings are also in agreement with the work of Rajesh *et al.*, (2008) who have observed insecticidal activity *Aegle marmelos* essential oil against four stored grain insect pests viz. *Callosobruchus chinensis*, *Rhyzopertha dominica*, *Sitophilus oryza*, and *Tribolium castaneum*. Zaidur Rahmanolium Sabuj *et al.*, (2017) have studied the control potential of *Saraca indica* extract against the adults of stored product pests *Callosobruchus chunensis*, *Sitophilus oryzae* and *Tribolium castaneum* and found a more or less similar result.

In the present investigation the ovicidal activity of the methanol extract of leaves of *Saraca indica* against four insect pests pest, no hatching of larvae were observed till 24 hours. Hatching of larvae were observed from 24 hours to 72 hours. No further hatching was recorded after 72 hour of treatment. Therefore LC50 value of ovicidal activity was recorded at 72 hour of exposure period (Table 5). At 10 ppm and after 72 hours of incubation *O. nibilalsis*, *S. littoralis*, *C. maculatus* and *T. confusum* the hatching percentage was 80.75%, 78.51%, 76.25% and 78.85% respectively. The hatching of larvae showed a decreasing trend on increasing the concentration of methanol extract of *Saraca indica*. At 1000ppm concentration of leaf extract and after 72 hrs of incubation the hatching percentage of *O. nibilalsis*, *S. littoralis*, *C. maculatus* and *T. confusum* was 26.15%, 21.16%, 22.18% and 24.15% respectively (Table 5). The LC50 value along with the regression equation is listed in Table 6-9. The survivability of larvae of these insect pests has been indicated in Table 10. From the result it is evident that the larvae of all the four

insect pests viz. *Ostrinus nubilalsis*, *Spodoptera littoralis*, *Callosobruchus maculatus* and *Tribolium confusum* survived at 10 ppm concentration of methanol extract even after 72 hours of incubation. The survival percentage of the larvae of all the four insect pests decreased with increased concentration of methanol extract and with incubation period. At 1000ppm concentration of methanol extract of *Saraca indica* the larvae of these insect pests could not survive at any incubation time (Table 10).

In the present investigation the larvicidal activity of methanol extract of leaves of *Saraca indica* was studied against four insect pests viz. *Ostrinus nubilalsis*, *Spodoptera littoralis*, *Callosobruchus maculatus* and *Tribolium confusum*.

It was found that the larval mortality was directly related to the exposure time and concentration of the methanol extract (Table 11-14). For *Ostrinus lubilalsis* the LC50 values of the methanol extract at 24h, 48h and 72h was recorded as 181.75ppm, 165.41ppm and 122.43ppm respectively (Table 11). For *S. littoralis* it was 179.65ppm, 161.45ppm and 121.35ppm respectively (Table 12); for *C. maculatus* 176.60ppm, 157.44ppm and 128.25ppm respectively (Table 13) and for *T. confusum* 180.75ppm, 160.35ppm and 124.25ppm respectively (Table 14). The values of sub lethal concentrations are presented in Table 6-9.

In the present study, the methanol extract from the leaves of *Saraca indica* exhibited insecticidal activities against different developmental stages of the four target insect pests viz. *Ostrinus nubilalsis*, *Spodoptera littoralis*, *Callosobruchus maculatus* and *Tribolium confusum*. The findings revealed variation in the potentiality of the methanol extract in different development stages of the four insect pests.

Table.1 Fumigant efficacy of methanol extract of leaves *Saraca indica* on stored maize against four insect pests at 500ppm

Treatment	<i>O. nubilalis</i>			<i>S. littoralis</i>			<i>C. maculatus</i>			<i>T. confusum</i>		
	Grain damage (%)	Weight loss (%)	FDI (%)	Grain damage (%)	Weight loss (%)	FDI (%)	Grain damage (%)	Weight loss (%)	FDI (%)	Grain damage (%)	Weight loss (%)	FDI (%)
Extract	10.25 ^a ±0.31	5.25 ^a ±0.21	87.45 ±0.70	10.35 ^a ±0.31	4.75 ^a ±0.34	88.35 ±0.18	9.75 ^a ±0.21	5.15 ^a ±0.71	89.35	10.45 ^a ±0.31	5.25 ^a ±0.15	88.65 ±0.35
control	100.00 ^b ±0.00	48.45 ^b ±0.31		100.00 ^b ±0.00	47.25 ^b ±0.41		100.00 ^b ±0.00	47.25 ^b ±0.13		100.00 ^a ±0.00	47.75 ^b ±0.32	

±: Standard Error; Means within each column followed by different letter are significantly different ($P < 0.05$, student's *t* test)

Table.2 Fumigant efficacy of methanol extract leaves of *Saraca indica* on stored Rice against four insect pests at 500ppm

Treatment	<i>O. nubilalis</i>			<i>S. littoralis</i>			<i>C. maculatus</i>			<i>T. confusum</i>		
	Grain damage (%)	Weight loss (%)	FDI (%)	Grain damage (%)	Weight loss (%)	FDI (%)	Grain damage (%)	Weight loss (%)	FDI (%)	Grain damage (%)	Weight loss (%)	FDI (%)
Extract	7.25 ^a ±0.31	3.65 ^a ±0.21	88.55 ±0.69	8.25 ^a ±0.26	3.15 ^a ±0.25	89.65 ±0.61	6.35 ^a ±0.21	3.45 ^a ±0.31	90.25	3.68 ^a ±0.32	1.25 ^a ±0.31	93.15 ±0.32
control	100.00 ^b ±0.00	62.55 ^b ±0.27		100.00 ^b ±0.00	61.55 ^b ±0.41		100.00 ^b ±0.00	50.25 ^b ±0.12		100.00 ^a ±0.00	55.35 ^b ±0.30	

±: Standard Error; Means within each column followed by different letter are significantly different ($P < 0.05$, student's *t* test)

Table.3 Fumigant efficacy of methanol extract of leaves *Saraca indica* on stored Cowpea against four insect pests at 500ppm

Treatment	<i>O. nubilalis</i>			<i>S. littoralis</i>			<i>C. maculatus</i>			<i>T. confusum</i>		
	Grain damage (%)	Weight loss (%)	FDI (%)	Grain damage (%)	Weight loss (%)	FDI (%)	Grain damage (%)	Weight loss (%)	FDI (%)	Grain damage (%)	Weight loss (%)	FDI (%)
Extract	4.35 ^a ±0.31	0.53 ^a ±0.23	90.45 ±0.67	3.15 ^a ±0.25	0.55 ^a ±0.25	91.35 ±0.64	6.55 ^a ±0.21	3.25 ^a ±0.33	90.50	2.25 ^a ±0.31	0.56 ^a ±0.30	91.05 ±0.31
control	100.00 ^b ±0.00	58.50 ^b ±0.23		100.00 ^b ±0.00	28.45 ^b ±0.44		100.00 ^b ±0.00	40.25 ^b ±0.16		100.00 ^a ±0.00	37.32 ^b ±0.22	

±: Standard Error; Means within each column followed by different letter are significantly different ($P < 0.05$, student's *t* test)

Table.4 Fumigant efficacy of methanol extract of leaves of *Saraca indica* on stored Wheat against four insect pests at 500ppm

Treatment	<i>O. nubilalis</i>			<i>S. littoralis</i>			<i>C. maculatus</i>			<i>T. confusum</i>		
	Grain damage (%)	Weight loss (%)	FDI (%)	Grain damage (%)	Weight loss (%)	FDI (%)	Grain damage (%)	Weight loss (%)	FDI (%)	Grain damage (%)	Weight loss (%)	FDI (%)
Extract	5.65 ^a ±0.31	0.78 ^a ±0.21	93.25 ±0.65	4.65 ^a ±0.23	0.75 ^a ±0.25	87.21 ±0.63	3.65 ^a ±0.22	0.71 ^a ±0.31	93.45	8.25 ^a ±0.30	4.15 ^a ±0.31	87.35 ^a ±0.31
control	100.00 ^b ±0.00	58.55 ^b ±0.27		100.00 ^b ±0.00	53.05 ^b ±0.42		100.00 ^b ±0.00	52.15 ^b ±0.17		100.00 ^a ±0.00	47.50 ^b ±0.21	

±: Standard Error; Means within each column followed by different letter are significantly different ($P < 0.05$, student's *t* test)

Table.5 Hatching percentage of eggs of four insect pests after treatment of different concentration of *Aegle marmelos* leaf extract

No. of individuals	Conc (ppm)	<i>O. nubilalis</i>			<i>S. littoralis</i>			<i>C. maculatus</i>			<i>T. confusum</i>		
		24 hour	48 hour	72 hour	24 hour	48 hour	72 hour	24 hour	48 hour	72 hour	24 hour	48 hour	72 hour
150	10	60.17 ±1.17	81.45 ±1.15	80.75 ±1.17	51.75 ±1.26	80.41 ±1.27	78.51 ±1.07	51.75 ±1.19	75.45 ±1.27	76.25 ±1.31	49.51 ±1.09	78.75 ±1.25	78.85 ±1.17
	100	45.21 ±1.15	60.35 ±1.16	61.25 ±1.17	35.25 ±1.27	35.15 ±1.37	45.25 ±1.08	41.45 ±1.14	60.15 ±1.16	63.41 ±1.32	35.25 ±1.15	57.15 ±1.32	58.15 ±1.17
	500	25.31 ±1.13	32.25 ±1.14	38.35 ±1.17	27.26 ±1.19	24.16 ±1.15	35.35 ±1.21	32.17 ±1.16	35.36 ±1.10	34.35 ±1.30	22.15 ±1.17	35.21 ±1.31	36.25 ±1.17
	1000	15.27 ±1.12	21.35 ±1.17	26.15 ±1.17	17.41 ±1.18	18.50 ±1.12	21.16 ±1.17	17.15 ±1.17	21.17 ±1.17	22.18 ±1.18	19.35 ±1.18	21.45 ±1.14	24.15 ±1.37

Table.6 Ovicidal activity of *Saraca indica* leaf extract against *Ostrinia nubilalis*

<i>Ostrinia nubilalis</i>					
Time	LC50	Regression Equation	95% confidence level		Chi- square value
72 hours	280.75	Y = 3.86 + 0.54X	Lower Bound .355	Upper Bound .526	72.15

Table.7 Ovicidal activity of *Saraca indica* leaf extract against *Spodoptera littoralis*

<i>Spodoptera littoralis</i>					
Time	LC50	Regression Equation	95% confidence level		Chi- square value
72 hours	273.86	Y = 3.96 + 0.45X	Lower Bound	Upper Bound	71.83
			.345	.554	

Table.8 Ovicidal activity of *Saraca indica* leaf extract against *Callosobruchus maculatus*

<i>Spodoptera littoralis</i>					
Time	LC50	Regression Equation	95% confidence level		Chi- square value
72 hours	282.85	Y = 3.98 + 0.46X	Lower Bound	Upper Bound	72.15
			.345	.521	

Table.9 Ovicidal activity of *Saraca indica* leaf extract against *Tribolium confusum*

<i>Tribolium confusum</i>					
Time	LC50	Regression Equation	95% confidence level		Chi- square value
72 hours	286.75	Y = 3.96 + 0.47X	Lower Bound	Upper Bound	71.37
			.370	.517	

Table.10 Survivability larvae of four insect pests after treatment of different concentration of *Saraca indica* leaf extract

No. of individuals	Conc. (ppm)	<i>O. nubilalis</i>			<i>S. littoralis</i>			<i>C. maculatus</i>			<i>T. confusum</i>		
		24 hour	48 hour	72 hour	24 hour	48 hour	72 hour	24 hour	48 hour	72 hour	24 hour	48 hour	72 hour
60	10	100.00 ±0.0	100.00 ±0.0	100.00 ±0.0	100.00 ±0.0	100.00 ±0.0	100.00 ±0.0	100.00	100.00 ±0.0	100.00 ±0.0	100.00 ±0.0	100.00 ±0.0	100.00 ±0.0
	100	85.21 ±1.15	75.05 ±1.16	65.15 ±1.17	82.25 ±1.27	78.15 ±1.37	67.08 ±1.08	83.45 ±1.14	71.15± 1.16	61.41 ±1.32	79.05 ±1.15	68.07 ±1.32	60.05 ±1.17
	500	30.15 ±1.13	25.50 ±1.14	15.15 ±1.17	27.16 ±1.19	28.16 ±1.15	16.10 ±1.21	28.17 ±1.16	20.41± 1.10	14.25 ±1.30	31.00 ±1.17	26.07 ±1.31	15.15 ±1.17
	1000	0.0	0.0	0.0	0.0	0.0	0.0	10.00 ±1.17	0.0	0.0	10.0 ±1.18	0.0	0.0

Table.11 Larvicidal activity of *Saraca indica* leaf extract against *Ostrinia nubilalis*

<i>Ostrinia nubilalis</i>					
Time	LC50	Regression Equation	95% confidence level		Chi- square value
24 hours	185.75	Y = - 1.49 + 2.78X	Lower Bound	Upper Bound	7.63
			1.895	3.05	
48 hours	165.45	Y = -0.65 + 2.50X	1.73	2.79	8.07
72 hours	121.43	Y = -0.21 + 2.41X	1.63	1.95	6.85

Table.12 Larvicidal activity of *Saraca indica* leaf extract against *Spodoptera littoralis*

<i>Spodoptera littoralis</i>					
Time	LC50	Regression Equation	95% confidence level		Chi- square value
24 hours	179.65	Y = - 1.49 + 2.45X	Lower Bound	Upper Bound	7.81
			1.756	3.015	
48 hours	161.45	Y = -0.21 + 2.41X	1.87	3.04	8.02
72 hours	121.35	Y = -0.17 + 2.12X	1.76	2.82	6.87

Table.13 Larvicidal activity of *Saraca indica* leaf extract against *Callosobruchus maculatus*

<i>Callosobruchus maculatus</i>					
Time	LC50	Regression Equation	95% confidence level		Chi- square value
24 hours	176.60	Y = - 1.46 + 2.41X	Lower Bound	Upper Bound	7.76
			1.758	3.013	
48 hours	157.44	Y = -0.24 + 2.31X	1.85	3.02	8.07
72 hours	118.25	Y = -0.15 + 2.13X	1.71	2.80	6.90

Table.14 Larvicidal activity of *Saraca indica* leaf extract against *Tribolium confusum*

<i>Tribolium confusum</i>					
Time	LC50	Regression Equation	95% confidence level		Chi- square value
24 hours	180.75	Y = - 1.51 + 2.80X	Lower Bound	Upper Bound	7.51
			1.90	3.01	
48 hours	160.35	Y = -0.59 + 2.48X	1.85	3.01	7.04
72 hours	124.25	Y = -0.25 + 2.50X	1.78	2.81	6.83

The findings of the present investigation receive supports from works done by previous researchers. Works on *S. indica* extracts for insects mortality and repellency is scanty, however a lots of work have been done on larvicidal activity. The findings on the test insects mortality through this investigation are supported by Mathew *et al.*, (2009) that revealed the Petroleum ether extract of *S. indica* leaves and the chloroform (CHCl₃) extract of the bark were effective against the larvae of *Culex quinquefasciatus* with respective LC₅₀ values, 228.9 and 291.5ppm, which follows the WHO standard protocols. The results are also supported by Jinu and Jayabaskaran (2015), which yielded that the Pet. ether extract of *S. indica* leaves and CHCl₃ extract of bark exhibited more than 50% larval mortality against *C. quinquefasciatus* larvae at an exposure period of 48h. No such reports have been reported so far on insect repellent activity of *S. indica* extract especially against the adults of the test insects. However, the findings by Singh *et al.*, (2009) showed that *S. indica* leaves were effective against antibacterial activity where ethanol (95%) and water extracts on agar plate *E. coli*, *S. aureus* by inhibitory effects on their growth which is similar to our findings in case of repellency test. However, another finding in the same experiments by Singh *et al.*, (2009) showed that *E. coli* were found active whereas tested against *S. aureus* gave negative results. The findings of the present investigation also gets support from the findings of Sarojini *et al.*, (2011) which revealed that the methanolic extracts were found relatively more potent as an anthelmintic agent due to presence of alkaloids. The mortality results also gets support from the findings of Verma *et al.*, (2010) where *S. indica* methanolic leaves extracts showed that the central nervous system (CNS) of albino mice was depressant. The findings of the inhibitory or mortality results gets support from the findings by

Dabur *et al.*, (2007) that the methanolic extracts of *Saraca indica* exhibited good inhibitory activity against *A. canjani* while it is effective at lower concentrations against other fungi also. The findings of Dubey *et al.*, (2008) revealed that food grain losses due to insect infestation during storage are serious problem, particularly in the developing countries. It is estimated by Ahmed and Grainge, (1986) that more than 20,000 species of field and storage pests destroy approximately one-third of the world's food production, valued annually at more than \$100 billion among which the highest losses (43%) occurring in the developing world. The present investigation was carried out against *Ostrinia nubilalis* (*European corn borer*), *Spodoptera littoralis* (*Cotton leaf worm*), *Callosobruchus maculatus* (*cowpea seed beetle*) and *tribolium confusum* (*Confused flour beetle*) to yield promising fumigating and insecticidal activity as all the four insects are stored product pests and they cause a huge damage in stored products and ultimately cause economic damage. Moreno & Racelis, (2015) finds out that repellency is the system tends to dissuade pests away from a susceptible crop (repellent) what can be called a push approach and our findings in controlling these pests gets support from it. Thus, plants are natural source of these repellent agents, reported in several ethno botanical information. Ali *et al.*, (2017) concluded that plant-derived repellents or insecticides do not pose hazards of toxicity to humans and domestic animals, and are easily biodegraded compared to synthetic compounds, natural products are presumed to be safer for humans. The extracts of *S. indica* leaves can be used in the control of these stored product pests as the results of the investigation showed both repellency and mortality against the test insect pests. This study was attempted to highlight *S. indica* claimed to be used or associated with insect repellent and mortality activity, and it was

found considerable. However, test result on other attributes also support the present finding, such as mortality and repellency for the extracts of *S. indica* against stored product pests.

Stored products cover a major portion of agricultural products but several species of insects infest these in storage condition and causing a huge damage. Using plants with insecticidal properties is therefore an attractive alternative to save them in comparison to the more expensive synthetic pesticides. Various plants by-products have been tried recently with a good degree of success as protectants against a number of stored grain insect pests. The findings of the present study indicate the ovicidal and larvicidal efficacy of extracts of *S. indica* on *Ostrinia nubilalis* (European corn borer), *Spodoptera littoralis* (Cotton leaf worm), *Callosobruchus maculatus* (cowpea seed beetle) and *Tribolium confusum* (Confused flour beetle)

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