



Toxicity and Bioefficacy of Selected Botanicals Protecting Sesame Seeds (*Sesamum Indicum*) from *Tribolium Castaneum*

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Abstract

The study explores the bioefficacies of distinct plant derivatives that have an effect on the improvement of the red flour beetle, *Tribolium castaneum* fed on black sesame seeds. Plant extract powder and oil from *Ocimum sanctum* (Tulsi Oil), *Mentha Piperita* (peppermint oil), and *Eucalyptus globules* (eucalyptus oil) had been evaluated for their oviposition inhibition, residual toxicity, direct toxicity effects, and repellency on *Tribolium castaneum*. The effects confirmed that plant oils had been high-quality in checking insect infestation. The least quantity of F1 adults emerged from sesame seeds treated with collected plant oil. The powdered leaves and extracts of peppermint, tulsi, and eucalyptus, at a 3% mixture, provided good safety for sesame seeds with the aid of decreasing insect oviposition, F1 adult emergence, and sesame infestation rates. The oil treatment did no longer exhibit destructive outcomes on the germination functionality of seeds, even after three months of treatment. The four essential plants oil tulsi showed powerful repellent against *T. castaneum* beetles. In conclusion, the essential oils from the botanical plants may be explored as a potential natural insecticide for stored-product insect pests because of their high repellency and insecticidal activities.

Keywords: Sesame Seeds, Red flour Beetle, Tulsi, Peppermint, Eucalyptus, Toxicity, and Repellency

1. Introduction

The red Tenebrionidae beetle, genus *Tribolium castaneum* Herbst (Coleoptera: Tenebrionidae), is one among the foremost widespread and damaging stored-product pests throughout the globe. Beetles and larvae kill a really large choice of dry vegetable substances, like polished cereal products (Rees, 2004). Management of hold on product insects depends heavily on employment of artificial pesticides and fumigants, that has LED to issue like environmental disturbances, increasing prices of the applying, cuss advance, cuss resistance to pesticides, and deadly effects on non-target organisms, additionally to direct toxicity to users (Isman MB, 2006).

Essential oils possess acute contact, and chemical toxicity to insects (Liu and metallic element, 1999; Sahaf et al., 2008; Abdelgaleil et al., 2009), repellent activity (Wang et al., 2006; Nerio et al., 2009), antifeedant activity (Huang et al., 1999), moreover development and growth repressive activity (Tomova et al., 2005; Waliwitiya et al., 2008). Many volatile oils and volatile compounds are rumored as attractants, repellents, pesticides, oviposition deterrents, and growth inhibitors against several holds on product insects together with *T.*

castaneum, and area unit thought-about as promising cuss management agents (Deshpande et al., 1974; Deshpande and Tipnis, 1977; Regnault-Roger and Hamraoui, 1995; Lee et al., 2004; Mondal and Khalequzzaman, 2006).

Some investigators have tested the toxicity of artificial volatile compounds against hold-on product pests. In an exceedingly chemical toxicity experiment, Lee et al. (2004) found that one; 8-cineole was cyanogenic to *S. oryzae*, *T. castaneum*, and *Rhyzopertha* Dominica adults. Burkholder et al. (1973) have rumored that propanoic acid was cyanogenic to larvae and eggs of *Trogoderma variabile* Ballion and *Attagenus megatoma* (F.) (Coleoptera, Dermestidae) once another to the insects food at a pair of concentration. Propanoic acid may be a present acid. It happens within the mix of volatile compounds emitted by barley grains (Maga, 1978). It's normally utilized by the food trade as a preservative in many food products. Benzaldehyde, the only aromatic organic compound, was rumored to be a safer chemical to regulate hold on grain insect pests (Lee et al., 2004). Toxicity and grain protective the effectiveness of many volatile oils and volatile compounds are checked separately against *T. castaneum* by several investigators (Negahban and Moharrampour, 2007; Mohamed and Abdelgaleil, 2008). However, the additive, or synergistic effects of volatile oils and volatile compounds weren't studied against *T. castaneum*.

Biological management could also be a good strategy for stored-product cuss management. Biology pesticides have the advantage of providing novel modes of action against insects that may cut back the danger of cross-resistance moreover as providing new leads for the look of target-specific molecules (Liu *et al.*, 1999). During this context, several plant products are evaluated for his or her cyanogenic properties against the completely different hold on grain pests, particularly within the type of essential oils (Regnault-Roger, 1997; Rajendran and Sriranjini, 2008).

Plant derived materials area unit additional perishable. Some area units less cyanogenic to mammals could also be additional selective in action and will retard the event of resistance. Their main advantage is that they will be simply and cheaply created by farmers and small-scale industries as crude or part sublimate extracts. Within the last twenty years, tidy efforts are directed at screening plants to develop new biology pesticides as alternatives to the present pesticides. It was rumored that once mixed with stored-grains, leaf, bark, seed power, or oil extracts of plants cut down oviposition rate and suppress adult emergence of bruchids, and conjointly reduced seed harm rate (Talukder and howse 1994; onu and aliya 1995; shaaya et al. 1997; Keita et al., 2001; tapondjou et al., 2002)

Hence, the present research was carried out to determine the oviposition inhibition, residual toxicity, and direct toxicity of some local plant derivatives against the red flour beetle, *Tribolium castaneum* fed on Black sesame seeds (*Sesamum indicum*). Effects of treatments on the viability of Black sesame seeds was also determined.

2. Materials And Methods

2.1. Test Plant Materials

Fresh plant leaves of tulsi, peppermint, eucalyptus (eucalyptus globules labill) were collected from Gandhi park, pattukkottai, Thanjavur (Dt), and neighboring areas during the month of February, washed and air-dried in the shade. Dried leaves were then ground to powder using an electric grinder. The powder extract used in this experiment.

2.2. Collection of plant oil

Essential oils obtained from the local market, India (Kannan & Co, Pattukkotai; commercial producers of plant essential oils and aromatic substances) were used in this study. The scientific name, the common name of oils are *Ocimum sanctum* (Tulsi Oil), *Mentha piperita* (peppermint oil), and *eucalyptus* (eucalyptus globules labill).

2.3. Test insects and maintenance

The Red flour beetle, *Tribolium castaneum* was used for the present experiments. The adult *Tribolium castaneum* was collected from infested seeds purchased from the local market and brought to the laboratory. The culture was established using sesame seeds in a plastic container of 25 X 10 cm and maintained at room temperature 30 ± 2 °C and relative humidity of 70-75%. Sieving the culture separated the adult insects and the adults were used for subsequent experiments. During the study time, the culture in the containers was consistently preserved.

2.4. Sample preparation of test plants

Powder and dust preparation of leaves were made by separately grinding approximately 500g of leaves of (tulsi, peppermint, and eucalyptus) in an electric grinding machine. The resulting powder was passed through a 25-mesh sieve to obtain fine dust. The Talukder and Howse (1993) process has been used to prepare acetone extracts with amendments. Ten grams of ground leaves of tulsi, peppermint, and eucalyptus were separately mixed with 50ml acetone and stirred for 30 minutes using a magnetic stirrer, and then left to stand for 24 hours. The mixture was then filtered through Whatman #1 paper, and the solids were stirred again for 15 minutes with 30ml of acetone and filtered and filtrates were combined. The solvent from the pooled filtered solution was evaporated in a water bath at 65°C. After complete evaporation of solvents, the final crude extracts were weighed and preserved in sealed bottles in a refrigerator at 5°C until used for the insect bioassays.

2.5. Oviposition inhibition effects

Laboratory tests for oviposition inhibition effects were conducted according to the method of Talukder and Howse (1994) with some modifications. Whatman filter paper disks (80mm in diameter) were soaked in a 2 or 3% solution of an extract and air-dried for an hour. Only acetone was provided for the control filter sheets. The treated and control filter paper discs were placed singly at the bottom of Petri dishes (90mm diameter) and 10g of Black sesame seeds were placed on the papers. Five pairs (5 female and 5 male) of *Tribolium castaneum* beetles were released in each petri dish which was covered for the next 7 days allowing them to lay eggs.

The adults were then excluded, and the following data were collected:

The number of seeds in each petri dish as a whole

Number of eggs per 50 seeds in each petri dish from day 27-42

42 days after the set up the following data were recorded:

Percentage of eggs hatching = $(\text{Total egg hatch} / \text{Total eggs in each petri dish}) \times 100$

inhibition rates IR percent = $[(C_n - T_n) / C_n] \times 100$ (where C_n is the number of insects in the control dish and T_n is the number of insects in the handled dish).

2.6. Direct toxicity by dipping method

The leaf extracts of tulsi, peppermint, and eucalyptus were diluted with acetone to make 2,4 and 6% solutions. In the middle of a pad of filter paper were five pairs of adult insects (2-3 days old) twisted in order to enclose them. They were dissolved or regulated for 35 seconds in the diluted extract. The insects were removed, air-dried, and returned to Petri dishes containing 10g of black sesame seeds. Four replications were made for each dose. Mortality was observed 24,48, 72 and 96 hours after treatment. During their regular examination insects were deemed dead any who did not move or react to gentle contact. Insects were checked every day and were declared dead for any who did not move or respond to gentle touches. During their regular examination insects were deemed dead any who did not move or react to gentle contact. Abbott's formula corrected insect mortality data (1925).

2.7. Residual toxicity test

A residual toxicity test was conducted according to the method of Talukder and Howse (1994) with some modifications. Ground leaf powders of tulsi, peppermint, and eucalyptus were mixed with sesame seed at the rate of 2% and 3 % (w/w). The treated foods were then put into separate plastic pots (3.5cm 4cm) so that each pot contained 10g of sesame seeds. Four replications were made of each dose. At the center of the pot containing the seeds were five pairs of adult beetles, which were closed for 7 days with a cover to oviposit.

The control pots contained untreated sesame gram seeds. The adults were then removed from the pots. The following observations were recorded.

A number of F1 adults emerging from each pot (from day 27 to day 42).

At the conclusion of the trial, the seed harm rate was calculated using a random sample of 100 seeds.

Inhibition rates as described above

Percentage seed weight loss= $\frac{U - D}{U} \times 100$ (where, U=Weight of an undamaged sesame seed, D=Weight of a damaged sesame seed, Nd=Number of damaged seeds, and Nu=Number of undamaged seeds)

2.8. Repellent activity of Plant oils against *Tribolium castaneum*

To investigate the repellent behavior of plant oils, 5µl and 10µl of the chosen plant oils were placed in filter paper strips (6X4 cm) and allowed to dry for 5 minutes. The filter paper was then mounted in plastic jars and tied to the olfactometer's arm. Filter paper with no treatment was used as a control. After attaching all of the plastic vials to the arms, fifty newly emerging adults of *T.castaneum* were placed in the olfactometer, and EPI values were determined using the formula of (Sakuma M, Funkami H, 1985) Where Nt represents the number of insects on the tested sample side and Nc represents the number of insects on the control sample side.

2.9. Insecticidal activity of Plant oils

T.castaneum adults were collected from the laboratory colony when they were two weeks old. For each concentration and untreated power, three replications were performed. To test the oil's insecticidal efficacy, 3mm circular filter paper was dipped in 5, 10, and 15 µl of oil and put in small glass jars containing nutritious maize flour. In addition, 20 insects were placed in each jar, and mortality was registered every 24 hours for three days, with the percentage of mortality measured using Abbot's percent corrected formula (Abbott WS.1925)

2.10. Seed germination test

Three months after the application of oil, the viability of treated and regulated seeds is studied. For this test, the tulsi, peppermint, and eucalyptus oil were handled separately at a speed of 5 ml of oil per kg (1 percent v/w) of seeds. The control seeds were solvent but no untreated seed was filled with oil or solvent. The seeds were handled and managed for 2 to 3 hours by air. Then, for three months, 50 seeds from each of the three groups were put separately in glass jars under laboratory conditions but without insects. In Petri dishes, each group of seeds was put on moist filter paper. For the next thirty days, the dishes were monitored for seed germination.

3. Results

3.1. Oviposition inhibition effects of acetone extracts

The total number of eggs laid, egg hatching percentage, and inhibition rates in the treated with tulsi, eucalyptus, and peppermint at a 2 percent extract concentration were compared to the total number of eggs laid, egg hatching percentage, and inhibition rates in the treated with tulsi, eucalyptus, and peppermint. Treatment with tulsi and peppermint leaf extract resulted in the lowest hatching rate. The 3 percent combination showed similar patterns. Tulsi-treated food had the highest oviposition inhibition levels, followed by eucalyptus and peppermint.

Table 1 shows the influence of various plant acetone extracts on oviposition.

Herbal Extract	Complete laying amount of eggs		Number of larvae hatched		Inhibition rate of hatching (%)	
	2%	3%	2%	3%	2%	3%
Tulsi	93b	72b	54b	41c	52a	69a
Eucalyptus	109b	126a	79ab	63b	36b	48b
Peppermint	118ab	139a	72b	78ab	36b	40b
Control	150a	146a	129a	127a	-	-
SEM	10.8	10.8	8.7	8.7	2.3	2.3

At 5 percent (Multiple range measure Duncan) Hatching Inhibition Rate (percent)=[$((Lc-LT)/Lc)$] a 5% degree of chance is dramatically different (Lt =no. of larvae has emerged from control, and Lt =No. of larvae have emerged from handled sesame seeds. The results are significantly different. SEM=Standard error of mean.

3.2. Direct Toxicity effects of acetone extracts

Insect mortality was assessed at three separate rates of 2,4, and 6% after treatment due to direct toxicity of acetone extracts of Tulsi, Eucalyptus, and Peppermint leaves on *Tribolium castaneum* at 24,48, and 72 hours (Table 2). Tulsi>Eucalyptus>Peppermint was the order of toxicity of the three red meal beetle extracts. Mortality percentages were immediately propotional to and after the extract concentration.

Table 2. The direct toxicity effect of various acetone extracts on the Red flour beetle, *Tribolium Castaneum* (via dipping method).

plant extract	Dose(%)	Death rate of insects (%) at		
		24HAT	48HAT	72HAT
Tulsi	2	20abcd (27.33)	45ab (40.22)	76bc (57.73)
	4	28ab (33.72)	56a (48.61)	79abc (63.41)
	6	31a (34.52)	60a (52.78)	89a (69.86)
Eucalyptus	2	16bcd (22.63)	36b (37.24)	67cd (53.24)
	4	20abcd (27.33)	45ab (40.22)	75bc (57.54)
	6	24abc (29.14)	48ab (42.78)	83ab (67.58)
Peppermint	2	10d (20.72)	34.57b (37.59)	57d (49.18)
	4	16cd (23.89)	39b (39.94)	67bcd (54.92)
	6	22abcd (27.67)	43ab (41.23)	76abc (60.57)
Standard Error of mean(Sx)		1.51	1.72	2.23

HAT=Hours after treatment

Sx=Standard error of mean

3.3. Residual Toxicity of Powders

T.Castaneum was assessed against the effectiveness of various powders as seed protectants, by measuring the F1 progeny, the rate of seed injury, the percentage of weight loss, and the inhibition rates. Female beetles were discouraged from ovipositing in black sesame seeds treated with Tulsi, Eucalyptus, and Peppermint leaf powders added at 2% and 3% (w/w). Tulsi and peppermint powder had similar effects on F1 progeny, seed injury, weight loss, and inhibition rates at rates of 2 and 3%, respectively.

Table.3.The Red flour beetle, *Tribolium Castaneum*, was tested for residual toxicity of various powdered leaves.

Herbal Extract	Number of F1 adult appeared		Inhibition rates(%)		Level of Seed damage (%)		weight loss of seed(%)	
	2%	3%	2%	3%	2%	3%	2%	3%
Tulsi	87c	74c	32b	45ab	26c	23c	17c	14bc
Eucalyptus	96c	81c	27bc	39bc	30bc	26bc	21bc	16b
Peppermint	109b	93b	19c	29c	36b	30b	23b	19b
Control	127a	124a	-	-	34a	34a	21a	21a
SEM	5.5	5.1	-	-	4.1	3.2	4.1	3.9

Inside a column, values preceded by separate letters are substantially different at the 5% level of likelihood (Duncan's multiple range test).

Seed Weight Loss(%)=weight loss percentage

SEM stands for standard error of mean

3.4. Repellent activity of Plant oils against *Tribolium castaneum*

T. castaneum has been checked for biological activity at 5 μ l and 10 μ l of various plant oils. The results show that the plant oils tested against the chosen insect pest differed. Table 1 shows the outcome of the repellent behaviour test. The EPI values varied from +1 to -1. These words clearly convey the directional choice's polarity. Positive and negative ideals denoted favourable and unfavourable methods, respectively. In this analysis, the repellent effect of Tulsi oil was greater than other oil studied here. Repellence generally increased as all treatments of the here checked oils increased as repellence increased. At the concentration and this property can be clearly shown at 5 μ l (-0.60 and -0.73 at the 1h and 6h, respectively), and 10 μ l, in tulsi oil, strong repellents were found against *T.castaneum* beetles (-0.56 and -0.81 in 1h and 6h respectively). Eucalyptus oil exhibited increased repellent behavior next to Tulsi oil. At 5 μ l this indicates the value of +0.38 and +0.62 EPI and -0.60 and 0.71 respectively at 1h and 6 h at 10 μ l concentration. The operation of peppermint oil was very low.

Table.4.Plant oils affect the *Tribolium castaneum* of Excess Proportion Index (EPI)

Plant oil	Concentrations tested			
	5µl		10µl	
	1HR	6HR	1HR	6HR
Tulsi oil	-0.60	-0.73	-0.56	-0.81
Eucalyptus oil	+0.38	+0.62	-0.60	-0.71
Peppermint oil	+0.21	-0.08	-0.50	-0.36

Mean of five replications

3.5. Insecticidal activity of Plant oils against *Tribolium castaneum*

Tulsi oil, Eucalyptus oil, and Peppermint oil were tested for insecticidal activity against *T. castaneum* at three concentrations (5, 10, and 15). Table 2 contains information on the insecticidal function of the chosen plant oils. The findings show that there is a wide range of plant oils that can be used to combat the chosen insect pest. In general, higher concentrations and longer durations resulted in higher insect mortality. The insect mortality effectiveness was ranked in the following order: Tulsi oil precedes Eucalyptus oil and is followed by Peppermint oil. Tulsi oil has the highest percentage of mortality (76 and 92 percent at 48 and 72 hours after treatment, respectively). At 72 hours after surgery, eucalyptus oil reported an 86 percent mortality rate. Peppermint oil had a moderate activity on *T.castaneum*.

Table.5.Plant oils have insecticidal efficacy against *Tribolium castaneum*.

Plant oil name	Concentrations tested								
	5µl			10µl			15 µl		
	Exposure Time								
	24hr	48hr	72hr	24hr	48hr	72hr	24hr	48hr	72hr
Tulsi oil	8.0±1.10	8.0±0.84	22.0±0.84	14.0±0.55	44.0±0.89	88.0±0.44	42.0±1.43	76.0±1.14	92.0±0.84
Eucalyptus oil	0±0.00	0±0.00	2.0±0.44	0±55	4.0±0.55	6.0±0.54	2.0±0.64	4.0±0.55	10.0±0.84
Peppermint oil	2.0±0.50	2.0±0.44	2.0±0.64	6.0±0.55	20.0±1.22	22.0±0.84	14.0±0.71	22.0±1.36	32.0±1.30

3.6. Effect of different plant oils on seed germination

The effect of different plant oils (10ml/kg) on the viability of sesame seeds.

Name of oil	Number of Treated seeds	Germination(%)
Tulsi	50	93
Eucalyptus	50	83
Peppermint	50	91
Control	50	84
Untreated	50	89

In Control, seeds were treated with solvent only

In Untreated, seeds were not treated with any solvent

3.7. Statistical analysis

For direct toxicity experiments observed insect mortalities at 24, 48, and 72 hours after exposure, and the data was corrected using Abbott's (1925) formula before being analyzed using ANOVA. Duncan's Multiple Range Test was used to determine the mean values (Duncan 1951). Micrograms per insect (g/insect) were used to calculate the results. Similarly, if the normal deviations of two slopes did not overlap, they were considered significantly different ($P < 0.05$). Data for residual toxicity is converted by means of square root data ($Y = \sqrt{X}$) for the percentages of numbers and arcsins, and evaluated by ANOVA and compared to DMRT by mean values.

4. Discussion

Plant extracts have been used for centuries to detect stored product insects. (Akinyemi et al., 2005) The insecticidal properties of various plant extracts for preserved crop insects have been evaluated. (Adedire Ajayi, 2003) Sandbox potential, Hua crepitans (L.) reported that seed oil for the conservation of *Callosobruchus Maculatus* (F) seeds (Coleoptera: Bruchidae). Plant diseases diary and security. 110(6):602-610 Protection. Mendie, Smith, S.T., A.O., Coker, A.O., Akinyemi, K.O. (2005). Stated that Screening for antisalmonella typhi activity of some medicinal plants in southwestern Nigerian herbal medicine. Herbal pharmacotherapy journal. Rahman A, Talukder FA. 2006. reported the efficacy by pulse bee, *callosobruchus maculatus* of essential Nishida, eucalyptus, and bankalmi plant powder. The admixture of food to pithraj leaf, bark, and seed powder, showed Talukder and Howse (1994), decreased the oviposition rate in pulse beetles. Bhaduri et al., (1985), observed that the bankalmi leaf extract had pulse beetle insecticide properties. Some investigators have shown that various botanical extracts. Das (1986) found that seeds treated with botanical extracts oil did not lose their viability. Onu & Aliyu (1995) reported that though various pepper powders were effective in reducing oviposition and damage of *C. maculatus*, seed quality and viability were not affected. Keita et al. (2001) reported that powders made from essential oils of different basil provided complete protection against *c. maculatus*, and also did not show a significant effect on the seed germination rate. Our results confirmed these studies.

In recent years, plant products with considerable potential as insecticidal compounds have become extremely important. Another essential biological parameter to protect development following harvesting is the control of adults. If plant extracts and oils contain insecticidal compounds that destroy adult insects, egg-laying and progeny growth are reduced. The chemical composition of *C. carvi*'s essential oil from different countries has been extensively researched.

Numerous studies revealed that the essential oil had antimicrobial, antifungal, molluscicidal, nematocidal, antioxidant, and antiaflatoxigenic properties, as well as the ability to prevent cancer (Samojlik *et al.*, 2010). In this analysis, the oils demonstrated differing degrees of insecticide action. There was no mortality in *T. castaneum* with the plant oils at the exposure of 24 hours. Minimum mortality following treatment was estimated at 72 hours. Tulsi oil and Eucalyptus oil in particular exhibited increased insecticide efficacy against the pest. Maximum repellent activity in plant *T. purpurea* powders showed higher EPI values against *T. castaneum* was found in the olfactometer study (Pugazhvendan SR *et al.*, 2009). Both compounds were shown to be effective inhibitors of acetylcholinesterase (AChE) production in larvae of many stored product insects (Badawy MEI *et al.*, 2010). Several stored product insects, including rice weevil (*S. oryzae*), lesser grain borer (*R. Dominica*), red flour beetle (*T. castaneum*), and flat grain beetle (*Cryptolestes pusillus*) (Lopez MD *et al.*, 2010), have been identified.

The essential oils tested were not only effective repellents of *T. castaneum* but also effective insecticides. Oils derived from both *L. sempervirens* and *D. winter*, when applied topically to *T. castaneum* adults, demonstrated medium lethal concentrations equal to those previously recorded for *Elletaria cardamomum* L. oils when applied through touch to *S. zeamais* and *T. castaneum* (Huang *et al.*, 2000) finally The idea that Tulsi Oil's basic oil is a production possibility for stored goods as a new natural fumigant.

5. Conclusion

The current results suggest that botanical derivatives may be useful as commercially available store insect control agents. To some extent, all of the plant extracts examined were useful in decreasing the amount of *T. castaneum*, accompanied by Tulsi, Eucalyptus, peppermint proved to be the most successful of the three test plant materials against the larvae, according to the results of the current analysis. The agricultural sectors of developing countries would benefit from a study to increase the efficiency of botanical derivatives as insecticides because these substances are not only cheap but also less environmentally friendly when it comes to insecticide effects.

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