Journal of Entomology and Zoology Studies
Available online at www.entomoljournal.com

E-ISSN: 2320-7078
P-ISSN: 2349-6800
JEZS 2017; 5(2): 324-335

Received: 14-01-2017
Accepted: 15-02-2017

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Repellent activity and fumigant toxicity of a few plant oils against the adult rice weevil *Sitophilus oryzae* Linnaeus 1763 (Coleoptera: Curculionidae)

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Abstract
Insect pests cause damage to stored grains and processed products by reducing their dry weight and nutritional value. The control of rice weevil infestations has been primarily through the use of fumigants and residual chemical insecticides to augment the more obvious approach of hygiene. Treatment of rice with synthetic insecticides is not recommended because of direct and indirect health hazards to humans. Recognition of such detrimental effects of synthetic insecticides has prompted the development of new alternatives as less obtrusive management strategies that must be ecologically safer with no residual and noxious effects on non-target animals. Plant based formulations are chiefly biodegradable and are recognized as better sustainable and eco-friendly alternatives of synthetic pesticides in food security as their activity may be due to synergistic effects of different active principles leading to different mode of action during their pesticidal action. Therefore, in the present study, the repellent activity and fumigant toxicity of plant oils *viz*., Aniseed, Camphor, Citronella, Eucalyptus, Geranium, Lavender, Lemon, Rosemary, Vetiver and Wintergreen were tested at concentrations of 10 and 50µL against the adults of *Sitophilus oryzae* in the laboratory. Results revealed varying degree of repellency by plant oils on the adults of *Sitophilus oryzae*. Among the plant oils tested, maximum repellency expressed as Excess Proportion Index (EPI) was recorded in camphor. The order of the repellency of plant oils at 10µL on 6 hours of exposure with EPI were Camphor (-0.90) > Wintergreen (-0.88) > Lavender (-0.70) > Citronella (-0.70) > Rosemary (-0.67) > Vetiver (-0.62) > Lemon (-0.57) > Eucalyptus (-0.55) > Geranium (-0.44) > Aniseed (-0.04) and at 50µL it was Camphor (-1.0) > Wintergreen (-0.89) > Citronella (-0.89) > Lemon (-0.89) > Lavender (-0.71) > Vetiver (-0.69) > Geranium (-0.65) > Rosemary (-0.57) > Eucalyptus (-0.52) > Aniseed (-0.50). For fumigant toxicity (expressed in terms of adult mortality), the selected plant oils were tested at 10 and 50µL concentrations for 24, 48 and 72 hours, and Lemon oil exhibited the highest activity and their respective LD_{50} values were 58.86, 44.90 and 40.38µL. Results of this study indicate that plant oils might be useful for managing coleopterous insects in storage especially *Sitophilus oryzae* on rice. Yet, further scrutiny is required to use plant oils as one of the component in IPM programmes which may reduce the application of synthetic chemicals.

Keywords: Plant oils, *Sitophilus oryzae*, repellent activity, fumigant toxicity

1. Introduction

Around the world, agriculture is the main source of subsistence for millions of people [1]. Rice and wheat suffer heavy losses during storage due to infestation of insect pests. According to the Food and Agricultural Organization (FAO) estimate, 10 to 25% of the world's harvested food is destroyed annually by insects and rodent pests [2]. Therefore, crop protection plays a vital and integral role in modern agricultural production; and the everlasting demands on yield as well as intensification of farming practices have increased the problem of pest damage and hence control [3]. Major part of agricultural produce are not only used immediately after harvesting but are also stored in warehouses for a gradual use in other seasons or to export or transfer to other areas. The substantial amount of stored product is lost by various biotic and abiotic factors [4]. Though stored grains can be destroyed by fungi and vertebrate pests, insect pests are often the most important because of the favourable environmental conditions that promote their development [5]. Insect pests are the major problem in storage products throughout the world because they reduce the quantity and quality of stored products [6, 7].

One of the major pests of stored commodities in tropics is *Sitophilus* species, causing considerable economic loss to stored wheat grain [8, 9]. Heavy infestation of these pests due to the lack of proper food hygiene and storage may cause weight loss of as much as 30–40% [10, 11]. Direct feeding on the grain kernels may cause unfavorable effects on food quality, safety and preservation [12]. Rice is the seed of the monocot plant *Oryza sativa* and is the grain with the second highest worldwide production, after maize [13]. Rice, being one of the staple foods mostly consumed by many parts of the world, has been infested by many important insect pests such as *Sitotroga cerealella, Sitophilus oryzae, Sitophilus granarius, Rhyzopertha dominica, Oryzaephilus mercator* as well as *Scirpophaga incertulas* and *Scirpophaga innotata* among others [14, 15].

The "rice weevil", *Sitophilus oryzae* Linnaeus 1763 (Coleoptera: Curculionidae) is a serious and severe insect pest of stored cereals and their products [16, 17] and one of the most widespread and destructive stored product pests of rice throughout the world [18] besides wheat and maize causing heavy losses of stored food grain quantitatively and qualitatively throughout the world [19]. This pest originated from India, has spread worldwide by commerce and now has a cosmopolitan distribution. Cotton [20] has reported that a pair of *Sitophilus oryzae* can reproduce about one million of its species within a period of three months under favourable conditions. The adults are 2mm long with brown/black coloured body and a long snout. Adult rice weevils are able to fly and can survive for up to two years. Being a ubiquitous pest of economic importance, *Sitophilus oryzae* feeds internally by boring into stored grain. Adult weevils feed on rice and lay their eggs inside rice kernels in a small hole covered with a gelatinous excretion near the grain surface just about 90µm deep [21] where the larva develops within the grain, hollowing it out while feeding. The larva feeds within the kernel preferably on the germ of the grain thus removing a large percentage of the proteins and vitamins [18] and consumes the endosperm. It then pupates within the grain kernel and emerges 2–4 days after eclosion [22-24]. The adults feed mainly on the grain endosperm thus reducing the carbohydrate content [18] and leave a large, ragged exit hole in the kernel [25]. The larva of *Sitophilus oryzae* consumes 14mg grain/day and its adult stage consumes 0.4mg grain/day [26].

Control of insects relies heavily on the use of synthetic insecticides including organochlorines (lindane), organophosphates (malathion), carbamates (carbaryl), pyrethroids (deltamethrin) and fumigants such as methyl bromide, phosphine and sulfuryl fluoride. However, the indiscriminate application of synthetic products has led to various problems including toxic residual effects, environmental pollution and development of resistance in insects (Isman, 2006). Therefore, there is an urgent need to develop safe and convenient alternatives. Replacement of conventional synthetic insecticides by bio-rational ones is universally acceptable and practical in approach worldwide [28]. This necessitates continuous research towards substitution of hazardous synthetic insecticides for eco-friendly natural plant products with active safe components, among which are use of powdered plant parts, oils and extracts that result from secondary metabolism in plants [29]. Much effort has, therefore, been focused on plant derived materials for potentially useful products as commercial insect control agents. Considerable research has been carried out to manage stored product insects by using aromatic medicinal plants despite their excellent pharmacological actions [30-32] and several efforts have been focused on the use of plant derived materials including essential oils as bioinsecticides.

Plant essential oils have been used for centuries as fumigants and topical formulations applied to exposed skin and clothing as recorded in writings by ancient Greek, Roman and Indian scholars [33]. Essential oils have a long history in medical and dietary uses [34] and are "generally recognized as safe" [35] even though they demonstrated toxic effects against stored product insects [36] as well as agricultural pests [37, 38]. They may act as fumigants [17, 36, 39-43], contact insecticides [43, 44], antifeedants [38, 45] or repellents [44, 46]. Essential oils are defined as any volatile oil(s) that have strong aromatic components and that give distinctive odour, flavour or scent to a plant. These are the by-products of plant metabolism and are commonly referred to as volatile plant secondary metabolites [47]. Because of the intensity of plant-insect interactions, the plants have well developed defense mechanisms against pests and are excellent sources of new insecticidal substances. Due to their low mammalian toxicity [48] they could be used as alternative sources for controlling a number of insect pests including stored product insects [12]. Plant essential oils and their constituents have been shown to possess potential for development as new fumigants and they may have advantages over conventional fumigants in terms of low mammalian toxicity and low environmental impact. The objective of this study was to screen plant oils as repellents and fumigants against the adults of rice weevil, *Sitophilus oryzae*. Species were chosen because of their world-wide pest status as well as their availability and selection of plant oils for the experiment was based on prior knowledge of their insecticidal activities against related insect species.

2. Materials and methods

2.1 Plant oils

Plant oils used for the present study were obtained from Government recognized aromatic oil supplier, Chennai, Tamil Nadu, India (Table 1).

Table 1: Details of plant oils used for the study

Common Name	Scientific Name	Family
Aniseed	*Pimpinella anisum*	Apiaceae
Camphor	*Cinnamomum zeylanicum*	Lauraceae
Citronella	*Cymbopogon nardus*	Poaceae
Eucalyptus	*Eucalyptus globulus*	Myrtaceae
Geranium	*Pelargonium graveolens*	Geraniaceae
Lavender	*Lavandula officinalis*	Lamiaceae
Lemon	*Citrus aurantium*	Rutaceae
Rosemary	*Rosmarinus officinalis*	Labiatae
Vetiver	*Vetiveria zizanioides*	Poaceae
Wintergreen	*Gaultheria fragrantissima*	Ericaceae

2.2 Test insects

Sitophilus oryzae adults free of exposure to insecticides and pathogens were reared and cultured in uncooked rice. The plastic containers used for rearing of *Sitophilus oryzae* were covered with muslin cloth and the culture was maintained at 27 ±2°C, 65-70% R.H. and light: dark cycle of 12 hours. For the bioassays, the F_1 generation of the adults from the culture was used.

2.3 Repellent bioassay

Repellent bioassay of *Sitophilus oryzae* adults was studied using a multi-arm olfactometer (Figure 1) which consists of a middle glass chamber (5cm diameter) from which five glass tubes (15cm length; 2cm diameter) project outwards with an equal gap between them. The end of each glass arm was fitted with a glass vial (8x6.5cm). Concentrations of 10 and 50µL of the selected plant oils were applied onto filter paper strips (6x4cm) and was allowed to dry for a couple of minutes. The filter paper strips were then placed onto the inner surface of the glass vials which were attached to the arms of the olfactometer. Filter paper strip which did not receive any treatment served as control. After the attachment of the glass vials to the arms of the olfactometer, fifty newly emerged *Sitophilus oryzae* adults were introduced into the olfactometer via the central opening. After 0.5, 1, 2, 4 and 6 hours of treatment, the number of adults found in each glass vial was recorded and the Excess Proportion Index (EPI) was calculated using the formula of Sakuma and Fukami [49].

$$EPI = \frac{Nt - Nc}{Nt + Nc}$$

Where,
Nt is the number of insects present in the treated
Nc is the number of insects present in the control

Figure 1: Multi-arm glass olfactometer

Further, the repellent activity based on the combinations of plant oils (randomly chosen) on the adults of *Sitophilus oryzae* was also determined wherein two different plant oils were mixed in four different ratios (1:4, 2:3, 3:2 and 4:1). Five sets were arrived by combination of two different plant oils (Table 2). From this mixture, concentrations (50µL) of each group were applied onto filter paper strips and experiments were carried out as mentioned above and EPI calculated.

Table 2: Details of repellent study based on different combinations of plant oils

Combination of plant oils	Concentrations of different combinations of plant oils							
	C1		C2		C3		C4	
	10µL	40µL	20µL	30µL	30µL	20µL	40µL	10µL
Wintergreen (W) + Geranium (G)	W	G	W	G	W	G	W	G
Camphor (CA) + Vetiver (V)	CA	V	CA	V	CA	V	CA	V
Lemon (LE) + Lavender (LA)	LE	LA	LE	LA	LE	LA	LE	LA
Rosemary (R) + Aniseed (A)	R	A	R	A	R	A	R	A
Citronella (CI) + Eucalyptus (E)	CI	E	CI	E	CI	E	CI	E

2.4. Fumigant toxicity
Fumigant toxicity of the plant oils were tested against *Sitophilus oryzae*. Ten unsexed adults taken from the laboratory culture were placed in glass vials (volumes of 70mL) with screw caps. Concentrations (10 and 50µL) of plant oils made in acetone were treated on Whatman No. 1 filter paper (2cm diameter) and left for a couple of minutes for acetone to evaporate. The treated filter paper was then pasted on the underside of the screw cap and the cap was tightly closed. Acetone treated filter paper was used as treated control and the filter paper which received neither plant oil nor acetone served as untreated control. A total of three trials were carried with three replicates per trial for each concentration of plant oil and control (treated and untreated). Mortality was determined after 24, 48 and 72 hours from commencement of exposure.

3. Results
Ten different plant oils were tested against the adults of *Sitophilus oryzae* at concentrations of 10 and 50µL for repellent activity and fumigant toxicity tests. In general, repellency increases with increase in concentration in treatment. The result indicated variation among the plant oils tested. The EPI ranged from 0.41 to -0.90. EPI expresses polarity of the directional/ orientation choice. Positive and negative values indicate attractant and repellent activity respectively. At 10µL, camphor oil exhibited the highest repellent activity with an EPI of -0.90 followed by Wintergreen (-0.88), Lavender and Citronella (-0.70), Rosemary (-0.67), Vetiver (-0.62), Lemon (-0.57), Eucalyptus (-0.55), Geranium (-0.44) and Aniseed (-0.04) after six hours of exposure (Figure 2A). At concentration of 50µL, after six hours of exposure the EPI ranged between 0.2 and -1.0. Here,

again, the Camphor oil exhibited the highest repellent activity with -1.0 EPI followed by Wintergreen, Citronella and Lemon (-0.89), Lavender (-0.71), Vetiver (-0.69), Geranium (-0.65), Rosemary (-0.57), Eucalyptus (-0.52) and Aniseed (-0.50) (Figure 2B). Further, the repellent activity by combining two different plant oils, with four different ratios was also studied. Five sets of plant oils were tested with each set containing the four ratios (C1, C2, C3 and C4) mentioned elsewhere. In the first set, a combination of Wintergreen and Geranium was taken up. The results revealed that the C3 ratio of Wintergreen and Geranium exhibited the highest repellent activity with -0.96 EPI (Figure 3A). In the case of Camphor and Vetiver, it was C4 exhibiting the highest activity with -1.0 EPI (Figure 3B). For Lemon and Lavender, C1 showed highest activity of -1.0 EPI (Figure 3C). C2 (-0.90) recorded the highest repellent activity for Rosemary and Aniseed (Figure 3D) and it was C3 and C4 with an EPI -1.0 in the case of Citronella and Eucalyptus (Figure 3E). Amongst, the five set of synergistic plant oils tested, the C4 and C3 of Citronella and Eucalyptus and C1 of Lemon and Lavender showed the highest repellency of -1.0. The fumigant activity (expressed in terms of adult mortality) of the selected ten plant oils tested at 10 and 50µL concentrations for 24, 48 and 72 hours was assessed. Amongst, the plant oils tested, Lemon oil exhibited the highest activity with 38.66, 53.34 and 58.00% respectively at 50µL (Table 2; Figure 4A, 4B & 4C) and its respective LD_{50} values were 58.86, 44.90 and 40.38µL (Table 3; Figure 4D, 4E & 4F).

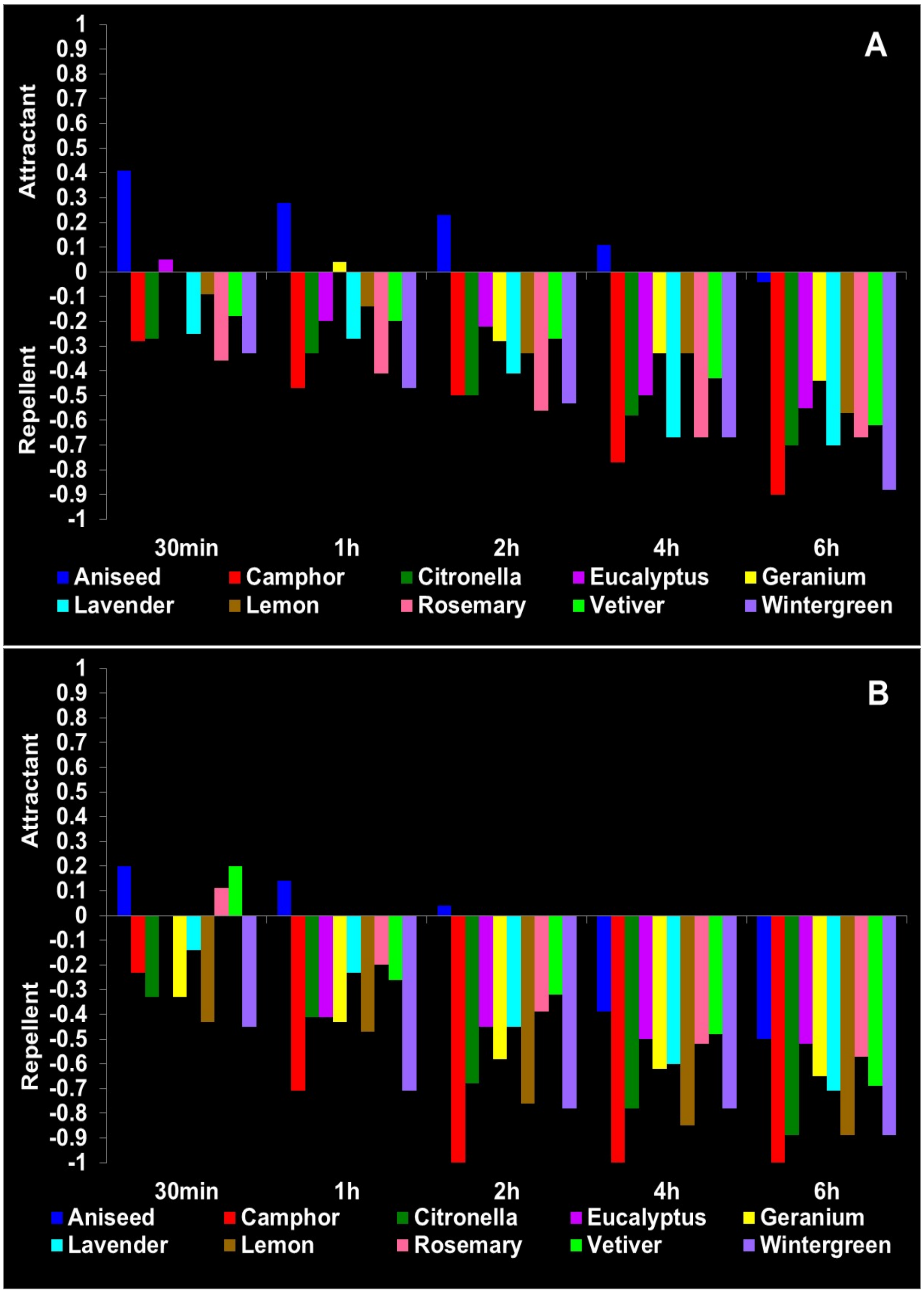

Figure 2: Effect of plant oils on EPI of *Sitophilus oryzae* adults. A: 10µL and B: 50µL

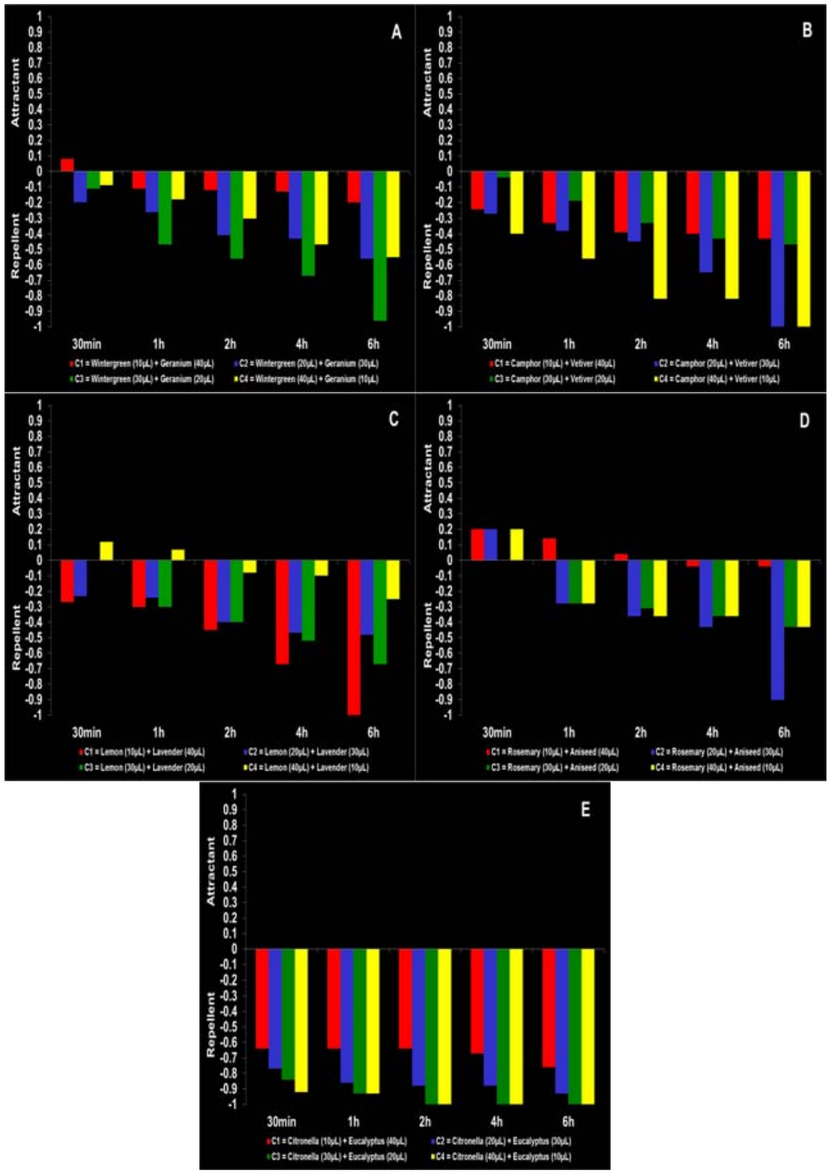

Fig 3: Effect of different combinations (A, B, C, D and E) of plant oils on EPI of *Sitophilus oryzae* adults

Table 3: Fumigant toxicity of plant oils against *Sitophilus oryzae*

Plant oils	24 hours		48 hours		72 hours	
	10µL	50µL	10µL	50µL	10µL	50µL
Aniseed	0.33 ±0.47 (0.66)	1.00 ±0.82 (2.00)	3.66 ±0.47 (7.32)	4.33 ±2.49 (8.66)	8.66 ±4.18 (17.32)	10.66 ±2.49 (21.32)
Camphor	1.66 ±1.25 (3.32)	11.66 ±3.09 (23.32)	5.33 ±3.30 (10.66)	19.00 ±2.16 (38.00)	7.00 ±3.74 (14.00)	24.33 ±0.94 (48.66)
Citronella	1.33 ±0.47 (2.66)	5.00 ±0.82 (10.00)	1.66 ±0.47 (3.32)	9.00 ±1.41 (18.00)	3.66 ±1.70 (7.32)	14.66 ±4.19 (29.32)
Eucalyptus	1.33 ±0.47 (2.66)	16.66 ±7.58 (33.32)	5.00 ±0.82 (10.00)	21.33 ±9.80 (42.66)	11.66 ±3.09 (23.32)	25.33 ±11.11 (50.66)
Geranium	0.66 ±0.94 (1.32)	5.33 ±3.86 (10.66)	1.00 ±0.82 (2.00)	9.33 ±5.73 (18.66)	1.66 ±1.25 (3.32)	13.33 ±8.06 (26.66)
Lavender	0.00 ±0.00 (0.00)	0.00 ±0.00 (0.00)	1.33 ±0.47 (2.66)	1.66 ±0.94 (3.32)	1.66 ±0.94 (3.32)	3.00 ±0.82 (6.00)
Lemon	8.33 ±0.82 (16.66)	19.33 ±2.05 (38.66)	13.33 ±4.18 (26.66)	26.67 ±1.25 (53.34)	16.66 ±3.74 (33.32)	29.00 ±0.47 (58.00)
Rosemary	0.33 ±5.24 (0.66)	1.00 ±0.82 (2.00)	3.33 ±0.47 (6.66)	15.33 ±1.24 (30.66)	9.66 ±2.86 (19.32)	19.66 ±4.49 (39.32)
Vetiver	0.00 ±0.00 (0.00)	0.00 ±0.00 (0.00)	0.33 ±0.47 (0.66)	1.00 ±0.94 (2.00)	0.66 ±0.47 (1.32)	3.66 ±0.94 (7.32)
Wintergreen	3.66 ±0.47 (7.32)	6.33 ±1.24 (12.66)	8.33 ±0.94 (16.66)	9.66 ±0.82 (19.32)	12.33 ±2.05 (24.66)	19.33 ±3.29 (38.66)

Values are mean of three replicates of three trials ±standard deviation; Values in parenthesis denote per cent adult mortality

Table 4: Probit analysis of fumigant toxicity of plant oils against *Sitophilus oryzae*

Plant oils	24 hours		48 hours		72 hours	
	Concentration (µL)					
	LD_{50}	LD_{90}	LD_{50}	LD_{90}	LD_{50}	LD_{90}
Aniseed	176.98	256.78	159.32	266.24	96.53	178.38
Camphor	71.70	110.51	58.73	99.41	49.92	86.78
Citronella	106.27	163.30	81.31	125.82	67.42	110.29
Eucalyptus	60.14	90.81	54.53	91.28	47.42	89.92
Geranium	92.68	136.89	76.20	114.25	67.29	103.47
Lavender	NA	NA	222.05	344.79	154.03	241.53
Lemon	58.86	106.04	44.90	87.51	40.38	83.12
Rosemary	176.98	256.78	65.31	105.78	58.34	107.81
Vetiver	NA	NA	176.98	256.78	109.36	162.11
Wintergreen	116.15	193.03	104.84	192.89	59.57	115.95

LD_{50}: Lethal concentration that kills 50% of the exposed adult; LD_{90}: Lethal concentration that kills 90% of the exposed adult; NA: Not applicable

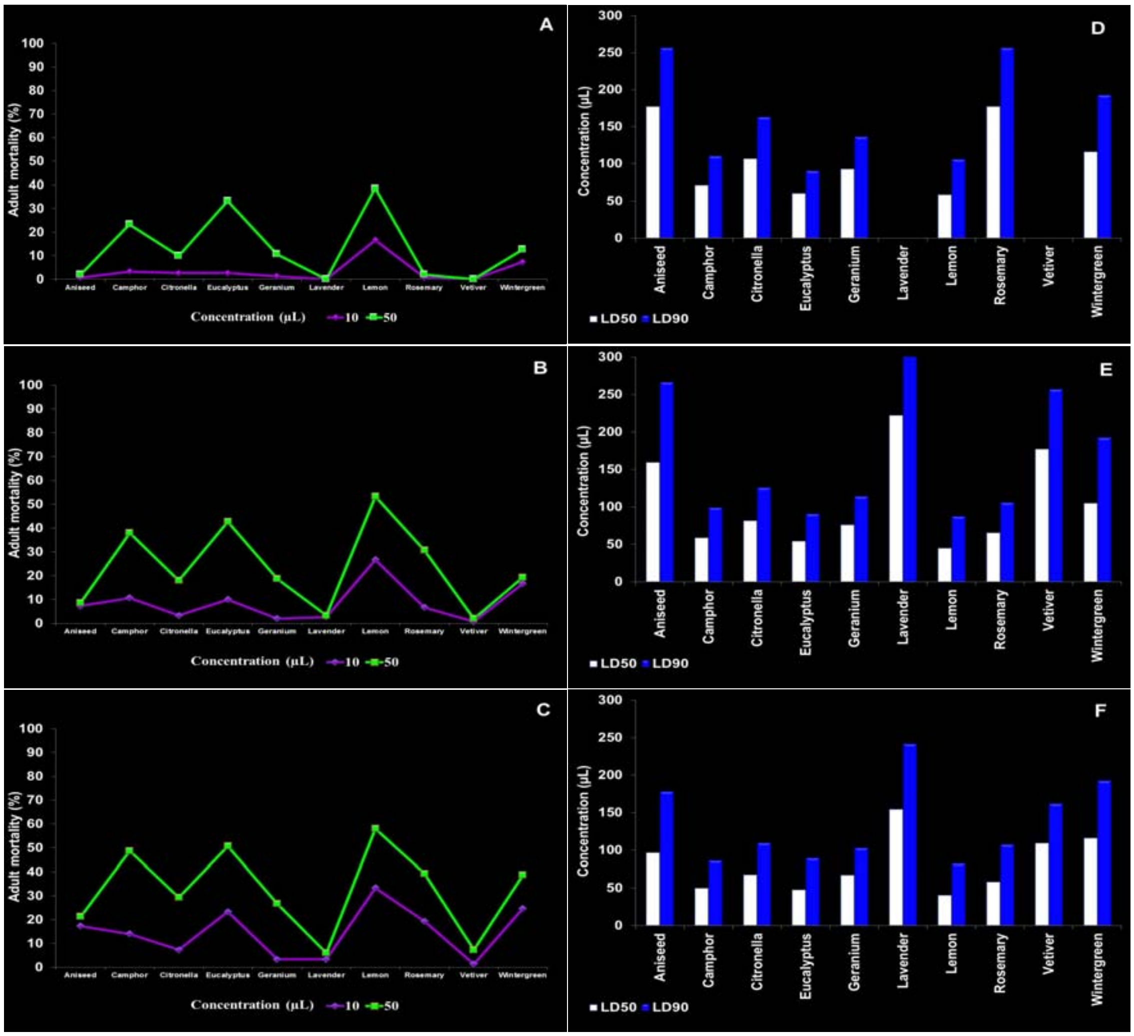

Figure 4: Fumigant toxicity of plant of plant oils and their corresponding probit analysis against *Sitophilus oryzae* adults at A&D: 24; B&E: 48; C&F: 72 hours

4. Discussion

Massive post-harvest losses sustaining in the developing world due to physical, nutritional and quality deterioration of stored grains are caused by insects and the detrimental impact of these losses on food security are well known [50]. Of all the coleopteran pests of stored grains, the most damaging species of storage insects are found in the genus *Sitophilus. Sitophilus oryzae*, an ubiquitous pest of economic importance, is an internal feeding insect pest that bores into stored grain [18]. Usually one larva hollows out small grain during its development. Adults cause further damage by feeding, mainly by attacking previously damaged grain. Rice weevil infestations may also produce a considerable amount of heat and moisture which encourage extensive quality loss, mould growth and growth of other insect populations [51].

Plants and their derivatives have played an integral role in the well-being of human race as they not only serve as food for man but also serve as source of medicine to man and animals. Insects, being major antagonists to man's combat against hunger have posted alarming threats to human security in term of food provision. However, because of the severe damage caused by these insects, man has overwhelmingly relied on the use of synthetic chemical insecticides which have been noted to link with numerous perils which lead to their ban in many developed countries [27, 52-54]. In order to replace these problems associated with chemical insecticides, man has returned to natural products from plants since they are believed to be safer to both human and environmental health. However, many botanicals in the man's habitats are yet to be exploited for their insecticidal potential. Jacobson [55] pointed out that the most promising botanical insect control agents are in the families Annonaceae, Asteraceae, Canellaceae, Lamiaceae, Meliaceae and Rutaceae and potent insecticidal activity against adults of *Sitophilus oryzae* was observed in the plants belonging to the families Apiaceae, Araceae, Lauraceae, Magnoliaceae and Myrtaceae. Since, aromatic plants were most efficient, the activity of their plant oils on *Sitophilus oryzae* was investigated.

Repellency increases the potential value of materials in protecting grains from the attack by stored product pests [56]. EPI value gives an idea about the repellency or attractancy of a chemical substance against an animal tested [57]. The repellent effect of volatile essential oils have an important implication in traditional post-harvest storage system and have potential activity for their local availability making it an attractive candidate in management of stored-grain insect pests [58]. Regnault-Roger [59], Cosimi *et al.* [60] and Nerio *et al.* [61] have recorded the repellent activity of essential oils against stored product pests. In the present study, at minimum exposure time, the plant oils showed attractant/ moderate repellency and high repellency when the exposure time was increased. At maximum exposure of 6 hours, all the plant oils showed repellent activity. The order of the repellency of plant oils at 10µL were Camphor > Wintergreen > Lavender > Citronella > Rosemary > Vetiver > Lemon > Eucalyptus > Geranium > Aniseed and at 50µL, it was Wintergreen > Citronella > Lemon > Lavender > Vetiver > Geranium > Rosemary > Eucalyptus > Aniseed. Whereas for minimum exposure of 30 minutes at 10µL, of the ten plant oils tested, seven plant oils showed repellent activity (Rosemary > Wintergreen > Camphor > Citronella > Lavender > Vetiver > Lemon) whereas two plant oils exhibited attractant activity (Aniseed > Eucalyptus) however Geranium showed neither repellent nor attractant activity and at 50µL, six plant oils showed repellent activity (Wintergreen > Lemon > Geranium > Citronella > Camphor > Lavender) whereas three plant oils exhibited attractant activity (Rosemary > Aniseed > Vetiver) however Eucalyptus showed neither repellent nor attractant activity.

This may be due to the potentiality of phytocompounds present in the plant oils which repels the insect. The toxic and repellent effects of the phytochemicals on stored grain pests depend on several factors among which are the chemical composition of the plant oils and susceptibility of the insect [62]. There are a few techniques for determining if substances repel or attract stored product insects and one such is usage of filter papers in glass test arenas [63]. In the present study, the filter paper technique was used with the aid of a multi-arm glass olfactometer which showed good results wherein Camphor, Citronella, Eucalyptus, Lemon and Wintergreen oil exhibited significant activity. Potential of these above mentioned plant oils have been reported by several researchers against the stored grain pests as well as crop pests [64, 65]. Besides these, Jayakumar and William [66] also reported a similar multi-arm glass olfactometer study, wherein crude solvent (hexane, chloroform and ethyl acetate) extracts of plants *viz*., *Argemone mexicana*, *Artemisia vulgaris*, *Sphaeranthus indicus* and *Tephrosia purpurea* when tested against *Sitophilus oryzae* adults, it was found that the ethyl acetate extract of *Artemisia vulgaris* and *Sphaeranthus indicus* exhibited repellent activity (66.23 and 54.55%) on exposure to one hour at 5% concentration. In addition, in preventing the resistant strains, the importance of mixed formulations containing the insecticides and the non-toxic chemicals was well emphasized. Hence, mixed formulations proved synergistic, additive and antagonistic activity. Therefore, in the present study, combinations of plant oils were used to study its repellent effect against *Sitophilus oryzae*. The combinations exhibiting either the synergistic, additive action would, therefore, help to reduce the actual quantity of the toxicant in the mixed formulation and at the same time enhance or maintain its desired toxicity. On the basis of our results, different combinations of plant oils might be very useful in the protection of stored products and the phytochemical constituents from plant oils can work synergistically, improving their effectiveness as observed in the present study.

The plant oils showed adult mortality when tested for fumigant toxicity. The fumigant toxicity effects of plant essential oils have been widely reported against pests of stored products [11, 40, 67-74]. Rajendran and Sriranjini [36] mentioned that essential oils of plants (mainly belonging to Apiaceae, Lamiaceae, Lauraceae and Myrtaceae) and their components (monoterpenoids and others) were tested for fumigant toxicity, where many of them indicated positive results against stored insect pests including *Sitophilus oryzae*. Notably, Lemon oil showed insecticidal activity against the target insect pest which was lesser at 24 hours period of exposure and thereafter an increase in mortality was recorded at 72 hours after treatment in the present study. Rahman and Schmidt [75] stated that the exposure period was more important than the dosage under the same conditions and the findings of the present study supports and confirms these observations. However, in the present study some of the plant oils did not show any mortality or showed least mortality which might be due to the presence of weak volatile compounds. Ho *et al.* [76] reported that the essential oil of *Evodia rutaecarpa* showed lesser insecticidal activity against *Tribolium castaneum* when the plant oil was extracted with the help of n-hexane. On the other hand the same authors reported that the steam distillated essential oils of the same plant showed higher activity [77] concluding that the potential phytocompounds can be leached out from the plant when steam distillation is applied. The adult mortality might be attributed also to the contact toxicity or to the abrasive effect on the pest cuticle [78], which might also interfere with the respiratory mechanism of insect [12, 79, 80]. Fumigation studies showed that the essential oils had a ‘knock down effect’ on the test insect. Essential oils act by inhibiting insect acetylcholinesterase (AChE) and thus, ultimately blocking the nerve functions. This is in agreement with studies by Obeng-Ofori and Amitaye [81] who observed signs of immobilization with flexed legs and clinging to the grain, outstretched meta thoracic wings from the elytra and paralysis of the dead or dying insects. The enzyme AChE is also the target site of inhibition by organophosphates and carbamate insecticides [82]. The observed rapid action of essential oils could be attributed to their property of acting in the vapour phase, hence gaining entry into the insect’s internal systems with ease through the spiracles; whereas, in topical application procedures, the insect is protected by its exoskeleton against external influences.

Essential oils possess acute contact and fumigant toxicity to insects [42, 71, 83], repellent activity [61, 84], antifeedant activity [85, 86], as well as development and growth inhibitory activity [87, 88]. Essential oils are volatile mixtures of hydrocarbons with a diversity of functional groups and the insecticidal constituents of essential oils are mainly monoterpenoids [89-93]. Most essential oils comprise of monoterpenes compounds that contain 10 carbon atoms often arranged in a ring or in acyclic form, as well as sesquiterpenes which are hydrocarbons comprising of 15 carbon atoms [47]. Repellent activity has been linked to the presence of monoterpenes and sesquiterpenes that cause death of insects by inhibiting AChE activity in the nervous system [94]. Monoterpenes being volatile are more useful as insect fumigants [92, 95-98]. Monoterpenoids have strong toxicity to insects due to high volatility and lipophilic properties can penetrate into insects rapidly and interfere in physiological functions [40, 68]. Due to their high volatility, they have fumigant and gaseous action on stored product insects. The monoterpene carvacrol has broad insecticidal and acaricidal activity against agricultural, stored-product, medical pests and acts as a fumigant being highly toxic to adults of *Sitophilus oryzae* [93]. Besides, menthol, methonene, limonene, β-pipene, α-pipene, pulegone, linalool and linalyl acetate exhibited fumigant toxicity in *Sitophilus oryzae* and inhibited AChE activity [22, 23, 47, 99]. Caryophyllene, a volatile compound, was reported to be a strong fumigant and toxic to *Sitophilus zeamais* [9].

The insecticidal activity varies with plant derived material, insect species and exposure time. The presence of volatile compounds having strong odour would have blocked the tracheal respiration of the insects leading to their death. Similar observation was made by Liu and Ho [83] against *Sitophilus zeamais* and *Tribolium castaneum*. Brown [100] however, pointed out that the amount of fumigant absorbed depends on whether the insect’s initial contact with the fumigant resulted in supplication or stimulation of the tracheal opening. Moreover, the ability of the insect to exclude vapour from its cuticle and prevent dehydration of body fluid plays a vital role in susceptibility or tolerance to fumigants of various life stages of insects particularly beetles and weevils infesting stored products [101]. The toxic effect of essential oils, apart from the variability of phytochemical patterns, involves several other factors. The point of entry of the toxin is one of them where essential oils can be inhaled, ingested or skin absorbed by insects [59].

The presence of volatile compounds is responsible for strong odour that could block the tracheal respiration of the insects leading to their death [102]. The mode of action of oils was partially attributed to interference in normal respiration, resulting in suffocation [79]. Most insects breathe through the trachea which usually leads to the opening of the spiracle. These spiracles might have been blocked thereby leading to suffocation [103, 104]. Essential oils are presumed to interfere with basic metabolic, biochemical, physiological and behavioural functions of insects [105]. Essential oils block the spiracles, resulting in blockage of respiratory siphons (asphyxiation) and death [106, 107]. Further, Rattan [108] reviewed the mechanism of action of essential oils on the body of insects and documented several physiological disruptions, such as inhibition of AChE, disruption of the molecular events of morphogenesis and alteration in the behaviour and memory of cholinergic system. Of these, the most important activity is the inhibition of AChE activity as it is a key enzyme responsible for terminating the nerve impulse transmission through synaptic pathway. Plant oils affect AChE and have an action on the nervous system [109]. Recent research has demonstrated the interference of monoterpenes with AChE activity in insects [110, 111]. Essential oils are lipophilic in nature and can be inhaled or ingested. The rapid action against insect pests is indicative of a neurotoxic mode of action and interference with the neuromodulator octopamine [112] or GABA-gated chloride channels [113]. Several essential oil components act on the octopaminergic system of insects. Octopamine is a neurotransmitter, neurohormone, and circulating neurohormone–neuromodulator, and its disruption results in total breakdown of the nervous system [114]. Thus, the octopaminergic system of insects represents a target for insect control. Low molecular weight terpenoids are too lipophilic to be soluble in the haemolymph after crossing the cuticle, and the proposed route of entry is tracheae [115] and may also bind to target sites on receptors that modulate nervous activity [114] and interrupt normal neurotransmission leading to paralysis and death. Results of this and earlier studies indicate that plant oils might be useful for managing coleopterous insects in storage especially *Sitophilus oryzae* on rice. Yet, further scrutiny is required to use plant oils as one of the component in IPM programmes which may reduce the application of synthetic chemicals.

5. Acknowledgement

The corresponding author is thankful to DST-PURSE Phase II Programme for the financial support.

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