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# *Acute, Lethal and Synergistic Effects of Some Terpenes Against Tribolium castaneum Herbst (Coleoptera: Tenebrionidae)*

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**Abstract.** Terpenes are found in abundance in all plant essential oils. In the present study two pure volatile compounds of terpene group, α-pinene and β-caryophyllene have been evaluated for their repellent, acute toxicity and developmental inhibitory activities alone and in binary combination against red flour beetle *Tribolium castaneum*. In repellency assay, α-pinene and β-caryophyllene repelled *T. castaneum* adults significantly even at 0.025% concentrations. Fumigation of larvae and adults of *T. castaneum* with these two compounds caused lethality in them. Median lethal concentrations ($LC_{50}$) of α-pinene and β-caryophyllene against adults were 0.998 and 1.624 µl/cm$^3$ and against larvae were 1.379 and 1.949 µl/cm$^3$ respectively. In binary combination, the $LC_{50}$ values against adults and larvae were found 1.277 and 1.438 µl/cm$^3$ respectively. Fumigation with two sublethal concentrations viz. 40 and 80% of 24-h $LC_{50}$ of these two compunds alone and in binary combination significantly reduced oviposition potential of adults and inhibited pupation and adult emergence in larvae. All the responses were found concentration-dependent. From the present study, it can be concluded that α-pinene is more active than β-caryophyllene and these two volatile compounds in binary combination shows synergism and thus, can used as efficient insecticidal tool against *T. castaneum* as fumigant either alone or in combination.

**Keywords:** α-Pinene, β-Caryophyllene, *Tribolium castaneum*, Fumigant toxicity, Terpenes.

## Introduction

Stored-grain insect pests have been damaging food grains in granaries and store houses and accounts for 10-40% loss worldwide (MATTHEWS, 1993). In India, this damage approximates for 10% during farm level storage (LAL, 1988). Despite application of improved storage structures and traditional control techniques, 70-90% of food grain is still stored not more than six months to a year at farmer's level. Thus, to protect stored grains from insect infestation exploration of other alternatives becomes quite essential. In this regard, synthetic pesticides came into existence, but uncontrolled use of these chemicals causes great environmental hazards due to their persistent nature, increased risk of neurotoxic, carcinogenic, teratogenic and mutagenic effects in non-target animals (BAKKALI *et al*., 2008; AYAZ *et al*., 2010). Besides, efficacy of these chemicals against stored-grain insect pests varies greatly after treatment and induced resistance against such chemicals (ZETTLER & CUPERUS, 1990; JEMBERE *et al*., 1995; PINTO *et al*., 1997). In another approach, plants and its products have been used in insect pest management programme since time immemorial. Amongst such plant derived products, essential oils have got much attention as fumigants since last two decades (AGRAWAL *et al*., 2001, TRIPATHI *et al*., 2003; CHAUBEY, 2007; SHUKLA *et al*., 2008; ABDEL-SATTAR *et*



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*al.,* 2010; AYAZ *et al.,* 2010; ZAPATA & SMAGGHE, 2010; CHAUBEY, 2011; STEFANAZZI *et al.,* 2011). Essential oils are complex mixture of volatile compounds produced as secondary metabolites. Bioactivity of these essential oils depends on its chemical composition which varies with plant part used for extraction, harvesting time, plant age, nature of the soil and growth conditions (ANGIONI *et al.,* 2006; ISMAN *et al.,* 2007). Efficacy of these essential oils is also affected by proportion of chemical constituents and synergism or antagonism among them (HUMMELBRUNNER & ISMAN, 2001; SAMPSON *et al.,* 2005; ANGIONI *et al.,* 2006).

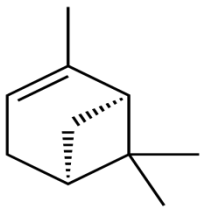
α-Pinene is a monoterpene containing a reactive four-membered ring. It is found as a important constituent in essential oils of *Nepta racemosa* (DABIRI & SEFIDAKON, 2003), *Ferulago* spp. (KHALIGHI-SIGAROODI *et al.,* 2005), *Syzygium aromaticum* (ALMA *et al.,* 2007), *Biden pilosa* (DEBA *et al.,* 2008), *Zingiber officinale* (KOROCH *et al.,* 2007; SASIDHARAN & MENON, 2010), *Eucalyptus* spp. (CHENG *et al.,* 2009; MACIEL *et al.,* 2010), *Citrus* spp. (KAMAL *et al.,* 2011) and *Vicia dadianorum* (KAHRIMAN *et al.,* 2012). β-Caryophyllene, a bicyclic sesquiterpene having cyclobutane ring, has been reported in *Piper cubeba* (LAWLESS, 1995), *Scutellaria pinnati* (GHANNADI & MEHREGAN, 2003), *Ferulago* spp. (KHALIGHI-SIGAROODI *et al.,* 2005), *Syzygium aromaticum* (ALMA *et al.,* 2007), *Biden pilosa* (DEBA *et al.,* 2008), *Eucalyptus* spp. (CHENG *et al.,* 2009; MACIEL *et al.,* 2010), *Citrus* spp. (KAMAL *et al.,* 2011) and *Pistacia lentiscus* essential oils (BURHAM *et al.,* 2011).

In the present study, two pure terpenes, α-pinene and β-caryophyllene have been evaluated for their repellent, acute toxicity and developmental inhibitory activities alone and in binary combination against red flour beetle *Tribolium castaneum*.

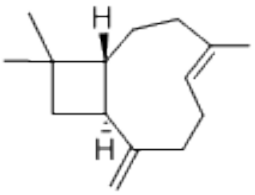
## Materials and methods

*Tested Terpenes*

The two pure terpenes viz. α-pinene and β-caryophyllene were purchased from Sigma Chemicals, USA.



α-pinene




β-caryophyllene

*Insects*

Red Flour beetles *T. castaneum* (Coleoptera: Tenebrionidae) were used to determine insecticide activity of terpenes. The insects were reared on wheat flour in the laboratory at 30±2°C, 75±5% RH and 10:14h (L: D) photoperiod.

*Bioassay Tests*

1. *Repellent activity of terpenes.* Repellency assay was carried out in glass petri dishes (diameter 8.5 cm and height 1.2 cm). Test solutions of serial dilution, 0.2, 0.1, 0.05 and 0.025% of α-pinene and β-caryophyllene were prepared in acetone. Whatmann filter paper was cut into two equal halves and each test solution was applied to filter paper half as uniform as possible using micropipette. The other half of filter paper was treated with acetone only. The volatile terpenes treated and acetone treated halves were dried to evaporate solvent completely. Both treated and untreated halves were then attached with cellophane tape and placed at the bottom in each petri dish. Twenty adults of *T. castaneum* were released at the centre of filter paper disc and then petri dishes were covered and kept in dark. Four replicates were set for each concentration of compounds solution. Number of insects on both treated and untreated halves was recorded after 4 h of start of experiment in mild light.

2. *Fumigant toxicity of terpenes against T. castaneum adults/ larvae.* Fumigant toxicity of α-pinene and β-caryophyllene alone or in binary combination was tested against *T. castaneum* adults and larvae. Ten adults/larvae taken from the laboratory culture were placed with 2 g of wheat flour in petri dishes (diameter 8.5 cm and height 1.2 cm). Filter paper strips (4 $cm^2$) impregnated with test solutions of α-pinene

and β-caryophyllene alone or in binary combination prepared in acetone, was pasted on the under cover of petri dishes. All petri dishes were closed and kept under same environmental conditions described for rearing of insect. Six replicates were set for each test solution as well as control group. After 24 h of fumigation, mortality in adults/larvae was recorded. In binary combination, α-pinene and β-caryophyllene were mixed in equal ratio. In control group, filter paper strips were treated with acetone only.

3. *Oviposition inhibitory activities of terpenes.* Oviposition inhibitory activity of α-pinene and β-caryophylene alone or in binary combination was tested against *T. castaneum* by fumigation method. Adults of mixed sexes were fumigated with filter paper strip (4 $cm^2$) impregnated with 40 and 80% of 24-h $LC_{50}$ of test solutions prepared in acetone as was done in toxicity assay. After 24 h fumigation, adults were transferred to clean petri dishes having wheat flour. After seven days of daily observation, adults were removed and discarded. Number of larvae hatched was counted for treated as well as control groups for ten days continuously. Six replicates were set for each concentration of compounds as well as control group. In binary combination, α-Pinene and β-Caryophylene were mixed in equal ratio. In control group, filter paper strips were treated with acetone only.

4. *Developmental inhibitory activities of terpenes.* Developmental inhibitory activities of α-pinene and β-caryophyllene alone or in binary combination were determined against $4^{th}$ instars larvae of *T. castaneum*. Larvae were fumigated with filter paper strip (4 $cm^2$) impregnated with 40 and 80% of 24-h $LC_{50}$ of test solutions prepared in acetone as was done in toxicity assay. After 24 h of fumigation, treated larvae were transferred to petri dishes having wheat flour. Number of pupae transformed from treated larvae and adults emerged from transformed pupae were recorded. Six replicates were set for each concentration of compounds as well as control groups. In binary combination, α-pinene and β-caryophylene were mixed in equal ratio. In control group, filter paper strips were treated with acetone only.

*Data analysis*

Chi-square test was applied to establish the repellent activity of the tested terpenes (SOKAL & ROHLF, 1973). Median lethal concentration ($LC_{50}$) was calculated by POLO programme (RUSSEL *et al.*, 1977). Correlation and linear regression analysis were conducted to define all concentration-response relationships (SOKAL & ROHLF, 1973). Analysis of variance was performed to test the equality of regression coefficient (SOKAL & ROHLF, 1973).

## Results

*Repellency effect*

In repellency assay, percentage of *T. castaneum* in treated filter paper disc half was 30%, 22.5%, 7.5% and 2.5 at 0.2%, 0.1%, 0.05% and 0.025% concentration of α-pinene (Table 1). Similarly, percentage of insects in treated filter paper disc half was 25%, 17.5%, 10% and 3.75% at 0.2%, 0.1%, 0.05% and 0.025% concentration of β-caryophyllene (Table 1). Chi-square analysis indicated that α-pinene and β-caryophyllene were repellent to *T. castaneum* adults. These two pure allelochemicals showed significant repellent activity even at 0.025%concentration as the hypothesis of ratio 1:1 was rejected (Table 1).

*Larval and adult mortality*

Fumigation of *T. castaneum* larva and adults with α-pinene and β-caryophyllene alone or in binary combination caused toxicity in them by vapour action. Median lethal concentrations ($LC_{50}$) of α-pinene and β-caryophyllene against larvae were 1.379 and 1.949 μl/$cm^3$ and against adults were 0.998 and 1.624 μl/$cm^3$ of air, respectively (Table 2). In binary combination of α-pinene and β-caryophyllene, $LC_{50}$ values against larvae and adult were 1.438 and 1.277 μl/$cm^3$, respectively (Table 2). With regard to larval mortality, regression analysis showed a concentration dependent significant positive correlation of α-pinene

(F = 226.61), β-caryophylene (F = 137.84) and α-pinene and β-caryophyllene mixture (F = 144.12) (P<0.0001, Table 5). Similarly, with regard to adult mortality, regression analysis showed a concentration dependent significant positive correlation of α-pinene (F = 138.35) and β-caryophyllene (F = 72.25), and α-pinene and β-caryophyllene mixture (F = 70.99) (P<0.0001, Table 5).

*Oviposition inhibition*

Fumigation of *T. castaneum* adults with two sublethal concentrations of α-pinene, β-caryophyllene and its binary combination reduced oviposition potential of insect. Oviposition was reduced to 71.91% and 58.54%; and 68.20% and 48.09% of the control when *T. castaneum* adults were fumigated with 40 and 80% of 24-h $LC_{50}$ of α-pinene and β-caryophyllene alone (Table 3). Similarly, oviposition was reduced to 56.4% and 36.52% of control when *T. castaneum* adults were fumigated with 40 and 80% of 24-h $LC_{50}$ of α-pinene and β-caryophyllene binary combination (Table 3). Regression analysis indicates that oviposition potential of the *T. castaneum* showed significant negative correlation with concentration when fumigated with α-pinene (F = 24.13), β-caryophyllene (F = 39.20), and α-pinene and β-caryophyllene binary combination (F = 70.52) (P<0.0001; Table 5).

*Developmental inhibition*

The percentage of larvae transformed into the pupae and percentage of pupae transformed into adult were decreased when fumigated with two sublethal concentrations of α-pinene and β-caryophyllene alone or in binary combination. Pupation in treated larvae was reduced to 70.8% and 55%; 85% and 64.2%; and 64.2% and 34.2% of control when *T. castaneum* larvae were fumigated with 40 and 80% of 24-h $LC_{50}$ of α-pinene and β-caryophyllene alone or in binary combination respectively (Table 4). Adult emergence was reduced to 50.8% and 39.2%; 66.7% and 45.8% and 47.5% and 15.8% of control when *T. castaneum* larvae were fumigated with 40 and 80% of 24-h $LC_{50}$ of α-pinene and β-caryophyllene alone or in binary combination respectively (Table 4). Regression analysis showed a concentration-dependent significant negative correlation of α-pinene fumes with pupation (F = 68.16) and adult emergence (F = 94.44) (P<0.0001; Table 5). β-caryophyllene fumes showed concentration-dependent significant negative correlation with pupation (F = 78.6) and adult emergence (F = 89.55) (P<0.0001; Table 5). Binary combination of α-pinene and β-caryophyllene also showed negative correlation with pupation (F = 180.5) and adult emergence (F = 565.31) (P<0.0001; Table 5).

**Table 1.** Repellenct effect of α-Pinene and β-Caryophyllene against *Tribolium castaneum* adults

| Concentration %(Vol:Vol) | α-Pinene | | | β-Caryophyllene | | |
|---|---|---|---|---|---|---|
| | Treated Mean±SE | Untreated Mean±SE | $\chi^2$-values | Treated Mean±SE | Untreated Mean±SE | $\chi^2$-values |
| 0.2 | 2.5±0.50 | 97.5±0.50 | 36.4[a] | 3.75±0.48 | 96.25±0.48 | 34.5[a] |
| 0.1 | 7.5±0.50 | 92.5±0.50 | 29.2[a] | 10±0.85 | 90±0.85 | 28.1[a] |
| 0.05 | 22.5±0.50 | 77.5±0.50 | 12.4[b] | 17.5±0.29 | 82.5±0.29 | 17.0[a] |
| 0.025 | 30±0.81 | 70±0.81 | 8.2[c] | 25±0.41 | 75±0.41 | 10.2[b] |

[a]Significant at P<0.001, [b]Significant at P<0.01, [c]Significant at P<0.05

**Table 2.** Fumigant toxicity of α-Pinene and β-Caryophyllene alone and in binary combination against *Tribolium castaneum* adults and larvae

| Terpenes | Parameters | $LC_{50}$ (µl/cm³) | LCL (µl/cm³) | UCL (µl/cm³) | g-value | t-ratio | Hetero geneity |
|---|---|---|---|---|---|---|---|
| α-Pinene | Adult mortality | 0.998 | 0.895 | 1.072 | 0.29 | 3.21 | 0.34 |
| | Larval mortality | 1.379 | 1.292 | 1.468 | 0.31 | 3.33 | 0.30 |
| β-Caryophyllene | Adult mortality | 1.624 | 1.204 | 2.025 | 0.32 | 0.41 | 0.41 |
| | Larval mortality | 1.949 | 1.776 | 2.099 | 0.34 | 3.54 | 0.39 |
| α-Pinene + β-Caryophyllene | Adult mortality | 1.277 | 1.13 | 1.424 | 0.35 | 3.17 | 0.30 |
| | Larval mortality | 1.438 | 1.277 | 1.60 | 0.28 | 3.26 | 0.35 |

$LC_{50}$ represents lethal concentration that causes 50% mortality
LCL and UCL represent lower confidence limit and upper confidence limit respectively
g-value, t-ratio and heterogeneity are significant at all probability levels (90, 95 and 99%)

**Table 3.** Oviposition behaviour of *Tribolium castaneum* adults when fumigated with α-Pinene and β-Caryophyllene alone and in binary combination

| Concentration | Number of larvae emerged per adult treated (Mean±SD) | | |
|---|---|---|---|
| | α-Pinene | β-Caryophyllene | α-Pinene + β-Caryophyllene |
| Control | 8.90±0.96 (100) | 8.90±0.96 (100) | 8.90±0.96 (100) |
| 40% of $LC_{50}$ | 6.40±0.84 (71.91) | 6.07±1.03 (68.20) | 5.02±0.42 (56.40) |
| 80% of $LC_{50}$ | 5.21±0.99 (58.54) | 4.28±0.69 (48.09) | 3.25±0.37 (36.52) |

Values in parentheses indicates per cent change control taken as 100%

**Table 4.** Developmental inhibitory activities of α-Pinene and β-Caryophyllene alone and in binary combination against *Tribolium castaneum* by fumigation method

| Conc. | Number of pupa transformed (Mean±SD) | | | Number of adults emerged (Mean±SD) | | |
|---|---|---|---|---|---|---|
| | α-Pinene | β-Caryophyllene | α-Pinene + β-Caryophyllene | α-Pinene | β-Caryophyllene | α-Pinene + β-Caryophyllene |
| Control | 10.0±0.0 (100) | 10.0±0.0 (100) | 10.0±0.0 (100) | 10.0±0.0 (100) | 10.0±0.0 (100) | 10.0±0.0 (100) |
| 40% of $LC_{50}$ | 7.08±0.93 (70.8) | 8.5±0.71 (85) | 6.42±0.32 (64.2) | 5.08±0.97 (50.8) | 6.67±0.76 (66.7) | 4.75±0.31 (47.5) |
| 80% of $LC_{50}$ | 5.50±0.71 (55) | 6.42±0.49 (64.2) | 3.42±0.43 (34.2) | 3.92±0.97 (39.2) | 4.58±0.49 (45.8) | 2.58±0.18 (25.8) |

Values in parentheses indicates per cent change control taken as 100%

**Table 5.** Regression parameters of oviposition and developmental inhibitory effects on *Tribolium castaneum* treated with α-Pinene and β-Caryophyllene alone and in binary combination by fumigation method

| Terpenes | Parameters | Intercept | Slope | Regression Equation | Regression coefficient | F-value |
|---|---|---|---|---|---|---|
| α-Pinene | % Adult mortality | - 10.06 | 0.93 | Y = - 10.06 + 0.93X | 0.965 | 138.35(P<0.0001)* |
| | % Larval mortality | - 11.08 | 0.66 | Y= - 11.08 + 0.66X | 0.947 | 226.61(P<0.0001)* |
| | % Oviposition | 6.83 | 8.41 | Y = 6.84 + 8.41X | - 0.979 | 24.13(P<0.0001)** |
| | % Pupal survival | 6.83 | 2.05 | Y = 6.83 + 2.05X | - 0.985 | 68.16(P<0.0001)** |
| | % Adult emergence | 6.11 | 0.66 | Y = 6.11 + 0.66X | - 0.942 | 94.44(P<0.0001)** |
| β-Caryophyllene | % Adult mortality | - 3.39 | 0.50 | Y = - 3.39+ 0.50X | 0.993 | 72.25(P<0.0001)* |
| | % Larval mortality | - 7.74 | 0.46 | Y = - 7.74 + 0.46X | 0.969 | 137.84(P<0.0001)* |
| | % Oviposition | 6.0 | 1.25 | Y = 6.0 + 1.25X | - 0.992 | 39.2(P<0.0001)** |
| | % Pupal survival | 7.27 | 3.11 | Y = 7.27 + 3.11X | -0.995 | 78.6(P<0.0001)** |
| | % Adult emergence | 6.50 | 1.75 | Y = 6.5 + 1.75X | -0.991 | 89.55(P<0.0001)** |
| α-Pinene + β-Caryophyllene | % Adult mortality | - 7.29 | 0.68 | Y = - 7.29 + 0.68X | 0.965 | 70.99(P<0.0001)* |
| | % Larval mortality | - 4.10 | 0.57 | Y = - 4.1 + 0.57X | 0.993 | 144.12(P<0.0001)* |
| | % Oviposition | 5.71 | 0.04 | Y = 5.71 + 0.04X | - 0.977 | 70.52(P<0.0001)** |
| | % Pupal survival | 5.89 | 1.04 | Y = 5.89 + 1.04X | -0.998 | 180.5(P<0.0001)** |
| | % Adult emergence | 5.34 | 0.22 | Y = 5.34 + 0.22X | -0.972 | 565.31(P<0.0001)** |

*df = 4,25; **df = 2,15

**Discussion**

Use of plant oils and its components as fumigants has received much attention of the scientific communities in pest management programme (AGRAWAL *et al.*, 2001, TRIPATHI *et al.*, 2003; CHAUBEY, 2007; SHUKLA *et al.*, 2008; ABDEL-SATTR *et al.*, 2010; AYAZ *et al.*, 2010; ZAPATA & SMAGGHE, 2010; CHAUBEY, 2011; STEFANAZZI *et al.*, 2011). The volatile components of essential oils can be classified into four main groups viz. terpenes, benzene derivatives, hydrocarbons and other miscellaneous compounds (NGOH *et al.*, 1998). Terpenes and terpenoids are the most representative molecules constituting 90% of the essential oils and allow a great variety of structures with diverse functions (BAKKALI *et al.*, 2008).

Many of the essential oil components of various chemical groups have also been evaluated for their role in insect pest management programme. *Mentha citrata* oil containing linalool and linalyl acetate exhibits fumigant toxicity to rice weevils (SINGH *et al.*, 1989). Linalool has been demonstrated to act on the nervous system affecting ion transport and the release of acetylcholine esterase in insects (RE *et al.*, 2000). DON-PERDO (1996) has studied effect of citrus peel oils and its components against *Callosobruchus maculatus*. Several compounds including the major component of all citrus peel oils, limonene has been found to be insecticidal. A combined study has established that in artificial mixtures, several pure components of citrus peel oil potentiate their individual fumigant activity (DON-PERDO, 1996). Carvone and menthol are effective as fumigant while 1,8-cineole exhibits both contact and fumigant toxicity

against *Tribolium castaneum* and *Callosobruchus maculatus* (TRIPATHI *et al.*, 2001). LEE *et al.* (2001) have reported toxicity of menthol, methonene, limonene, α-pipene, β-pipene and linalool against *Sitophilus oryzae* and proved that these essential oil components exert its toxicity by inhibiting acetylcholine esterase enzyme. *l*-carvone has been reported to cause more fumigant toxicity than its contact toxicity to *Rhizopertha domestica* (TRIPATHI *et al.*, 2003). *Trans*-anethole, thymol, 1,8-cineole, carvacrol, terpineol and linalool have been evaluated as fumigants against *Tribolium castaneum* but only compound to show significant effect against this insect species is trans-anethole (KOUL *et al.*, 2007).

A comparative study has been conducted to assess contact and fumigant toxicities of monoterpenes viz. camphene, camphor, carvone, 1-8-cineole, cuminal-dehyde, fenchone, geraniol, limonene, linalool, menthol and myrcene on *Sitophilus oryzae* and *Tribolium castaneum*. In fumigant toxicity assays, 1-8-cineole has found most effective against *Sitophilus oryzae* and *Tribolium castaneum*. Structure-toxicity investigations reveal that carvone has the highest contact toxicity against the both insects. *In vitro* inhibition studies of acetylcholine esterase from adults of *Sitophilus oryzae* show that cuminaldehyde inhibits enzyme activity most effectively followed by 1-8-cineole, limonene, and fenchone. 1-8-Cineole is the most potent inhibitor of acetylcholine esterase activity from *Tribolium castaneum* larvae followed by carvone and limonene (ABDELGALEIL *et al.*, 2009).

In the present study, α-pinene and β-caryophyllene have been evaluated for their repellent, acute toxicity and developmental inhibitory activities alone and in binary combination against flour insect pest *Tribolium castaneum*. These two compounds alone caused fumigant toxicity in adults and larvae both. Fumigation with two sublethal concentrations viz. 40 and 80% of 24-h $LC_{50}$ of these two compunds significantly reduced oviposition potential of adults and inhibited pupation and adult emergence in larvae. In reducing the dose of active compunds, target multiple site of action and resistance in insects, synergism can play an important role. Essential oil combinations such as thyme, anise and saffron have been demonstrated for synergistic activity (YOUSSEF, 1997). HUMMELBRUNNER & ISMAN (2001) reported that different combinations of monoterpenes produced synergistic insecticidal effects. Present study indicates that α-Pinene and β-Caryophylene in binary combination shows synergism and reduces the egg laying capacity and inhibits pupation and adult emergence in *T. castaneum*. These earlier reported findings clearly support the result of the present study. The mode of action of these essential oils is yet to be confirmed but it appears that death of the adults, larvae, oviposition inhibition and development inhibition may be due to the suffocation and inhibition of different biosynthetic processes of the insect metabolism (DON-PERDO, 1989). Rapid action of essential oils or its constituents against insect pests is an indicative of neurotoxic actions (KOSTYUKOVSKY *et al.*, 2002; PRIESLEY *et al.*, 2003; ISMAN *et al.*, 2007). Recent researches have demonstrated the interference of monoterpenes with acetylcholinesterase enzyme activity in insects (ZAPATA & SNAGGHE, 2010). Thus, it can be suggested that fumigants from volatile oils of plant origin could have greater potential in future on the basis of their efficacy, economic value and use in large-scale storage.

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