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Fumigant, contact, and repellent activities of essential oils against the darkling beetle, *Alphitobius diaperinus*

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Abstract

The fumigant, contact, and repellent activities of four essential oils extracted from *Citrus limonum* (Sapindales: Rutaceae), *Litsea cubeba* (Laurales: Lauraceae), *Cinnamomum cassia*, and *Allium sativum* L. (Asparagales: Alliaceae) against 6th instars and adults of the darkling beetle, *Alphitobius diaperinus* (Panzer) (Coleoptera: Tenebrionidae), one of the main pests of materials and products of *Juncus effuses* L. (Poales: Juncaceae) during the storage period, were assayed, and chemical ingredients were analyzed with gas chromatography-mass spectrometry in this study. While the major ingredients found in *C. limonum* and *C. cassia* were limonene and (E)-cinnamaldehyde, the main constituents of *L. cubea* were D-limonene, (E)-3,7- dimethyl-,2,6- octadienal, (Z)-3,7- dimethyl- ,2 ,6- octadienal, and diallyl disulphide (18.20%), while the main constituents of and *A. sativum* were di-2-propenyl trisulfide and di-2- propenyl tetrasulfide. The fumigation activities of *A. sativum* and *C. limonum* on *A. diaperinus* adults were better than those of the other two essential oilss. The toxicities of *A. sativum* and *C. limonum* were almost equitoxic at 96 hr after treatment. Essential oils from *Allium sativum* and *L. cubeba* also showed good contact activities from 24 hr to 48 hr, and toxicities were almost equitoxic 48 hr post-treatment. The repellent activities of *A. sativum* and *L. cubeba* oils on 6th instars were also observed, showing repellence indexes of 90.4% and 88.9% at 12 hr after treatment, respectively. The effects of *A. sativum* on AChE activity of 6th instars of *A. diaperinus* were strongest compared to the other essential oils, followed by *C. limonum*, *L. cubeba,* and *C. cassia*. These results suggest that the essential oils of *C. limonum* and *A. sativum* could serve as effective control agents of *A. diaperinus*.

Keywords: AChE activity, fumigant toxicity, repellent toxicity
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Introduction

At present, *Juncus effuses* L. (Poales: Juncaceae) is being widely cultivated in southwest China and is used as an important material for summer sleeping mats; it is exported to Japan, Korea, and some other southeast Asian countries, generating about $50 million per year in revenue (Li et al. 2009). *Alphitobius diaperinus* (Panzer) (Coleoptera: Tenebrionidae) is one of the most destructive pests of *J. effuses* in storage, and control treatments heavily depend on the use of synthetic insecticides and fumigants. However, excessive chemical applications have serious health hazards for human applicators, customers' skins, the environment, etc. (Wang et al. 2011). These problems required the developmentment of selective, environmentally-acceptable insect control alternatives (Copping and Menn 2000). Plant-derived natural chemicals have been known as secondary metabolites and possible alternatives to synthetic chemical insecticides and displayed strong biological activities (Cosimi et al. 2009; Nesci et al. 2011). In recent years, the search for new plant oils with insecticidal activity as possible alternatives to synthetic chemical insecticides has become a hotspot in pesticide research (Stefanazzi et al. 2011). Essential oils (EO) are volatile, small molecule, complex secondary metabolites that have been widely researched for their fumigant, contact, and repellant activities and have been developed as stored-product pest repellents or antifeedants (Jin et al. 2010).

The aim of this study was to determine the main components and examine the insecticidal activities of EOs extracted from four aromatic plants, *Citrus limonum* (Sapindales: Rutaceae), *Litsea cubeba* (Laurales: Lauraceae), *Cinnamomum cassia*, and *Allium sativum* L. (Asparagales: Alliaceae), against *A. diaperinus* and the effects on AChE activity of *A. diaperinus*. The results will give an insight into the potential of the tested EOs as effective alternatives to synthetic insecticides against *A. diaperinus* occurring storage.

Materials and methods

Insect cultures

Larvae and adults of *A. diaperinus* were reared in laboratory and maintained in darkness in incubators at 27 ± 2°C and 70 ± 5% RH. They were fed with a mix of wheat bran, maize powder, and peanut cake at an 8:1:1 weight proportion and 15% water. Sixth instars and adults two days post-eclosion were used in all experiments.

Essential oil extraction and gas chromatography-mass spectrometry

Leaves of *C. limonum, L. cubeba*, *C. cassia*, and *A. sativum* were collected during the autumn from Wenjiang District, Chengdu, China, located at 30°41′48.23″ N, 103°49′57.46″ E. The EOs were extracted with hydrodistillation using a modified Clevenger apparatus about 3–4 hr, dried over anhydrous sodium sulfate, and refrigerated at 4°C (Mahnaz et al. 2012).

The oils were analyzed in authors' laboratory by gas chromatography-mass spectrometry (GC-MS) with an Agilent 6890 chromatograph (www.agilent.com) connected to a 5973N mass spectrometer equipped with a capillary column (HP-5, 30 m × 0.25 mm, 0.25 µm). The GC oven temperature was held at 50°C for 2 min, programmed at 5°C min^{-1} ramp to 240°C, and then held at the temperature for about 15 min. The injector and detector temperature was 250°C, the carrier gas was He (1 ml min^{-1}, split ratio 1:50), and the samples and n-alkanes (consecutive C8-C40, purchased from AccuStandard,

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www.accustandard.com) were diluted in acetone (injection of 2 μL). Mass spectra were recorded at 70 eV, and the mass range was m/z 30–600 amu (Dalila et al. 2012). The compounds were identified by comparing their retention indices (Kovats indices) (Santos et al. 2011) with those of known compounds and their mass spectra with those stored in the MS database (NIST98 MS DATA). The relative percentage amounts were obtained directly from the GC peak areas.

Fumigant activity

The fumigant bioactivities of EOs were detected with the sealing jar method (Deng et al. 2004). Whatman No.1 filter papers were cut in filter paper strips (1.5 cm × 5 cm) and suspended in a 250 mL jar after adding 3.5 μL EO or acetone (control) on each strip (concentration of 14 μL/L). Thirty adults were added, and the vial quickly was covered with sealing plastic film. Each treatment was set for triplications and placed at 27 ± 2°C and 70 ± 5% RH. Mortalities and corrected mortalities after 48 hr and 96 hr were corrected for control mortality by using Abbott's (1925) formula. EOs with stronger activities were screened out and their toxicities were assayed on 6th instars and adults at the concentrations of 2, 4, 8, 16, 32, and 0 (control) μL/L. The LC_{50} and LC_{95} (lethal concentration) values and their 95% confidence intervals (CI) values 48 hr and 96 hr after treatment were calculated with POLO 2.0 (LeOra Software, www.leorasoftware.com) respectively.

Contact activity and toxicity

The contact activities of EOs on 6th instar *A. diaperinus* were assayed with an impregnated paper assay (Stefanazzi et al. 2011). EOs were diluted with acetone and applied to Whatman No. 1 filter paper (6 cm diameter) at a 40 μg/cm^2 concentration, using 1 mL acetone as the control. When the solution had absolutely volatilized about 15 min, 30 6th instar *A. diaperinus* were placed into a glass Petri dish with teflon coated on the inner wall to prevent escaping. Mortality and corrected mortality were calculated 24 hr and 48 hr post-treatment. Contact toxicities of EOs were assayed at the concentrations of 5 μg/cm^2, 10 μg/cm^2, 20 μg/cm^2, 40 μg/cm^2, 80 μg/cm^2, 160 μg/cm^2 and 0 (control), and all insect were cultivated at 27 ± 2°C and 70 ± 5% RH. All treatments were set for triplications. The LC_{50} and LC_{95} values and their 95% CI after 24 hr and 48 hr were calculated as the description of fumigant activity.

Repellent activity

The repellent activities of EOs on 6th instar *A. diaperinus* were determined as described by Wang et al (2006) and Yao et al (2008) with some modifications. Whatman No. 1 filter paper (diameter 12.5 cm) was cut in half, EOs were applied to half of a filter-paper disc as uniformly as possible, and other half of the filter paper was treated with acetone alone as the control. Three concentrations, 200, 400, 800 μg/cm^2, were held by dissolving different volumes of EO in L ml acetone. The treated and control half discs were air-dried a tabout 20°C for 15 min to evaporate the solvent completely. Treated and untreated halves were attached to their opposites using adhesive tape and fixed in Petri dishes (diameter 12.5 cm) with Teflon coated on the inner wall to prevent escaping. Thirty 6th instar larvae were released at the center of each filter paper disc. The dishes were then covered and held in an incubator at room temperature, and three replications were set for each concentration. After 12 , 24, and 48 hr, the number of larvae present at each amount of treated or control halves were calculated, and the distribution coefficient was calculated as follows:

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$$DC=\frac{C-T}{C+T}\times 100\%$$

where C is the number of insects present on the control areas of the discs and T is the number of insects present on the treated side.

AChE Activity Assay

The effects of EOs on AChE activities of 6th instar *A. diaperinus* were assayed with the colorimetric method as described by Abdelgaleil et al (2009) and Yeom et al (2012) with some modifications. Fifteen 6th instars larvae treated with *C. limonum*, *L. cubeba*, and *A. sativum* EOs or nothing (control) were quick-frozen with liquid nitrogen at 6, 12, 24, 36, 48, 60, 72, and 96 hr after treatment and separately homogenized in 10 mL of 0.1 M ice-cold phosphate buffer (pH 7.0) using a mortar. Homogenates were centrifuged (7,000 rpm for 20 min at 0ºC), and supernatants were used as the enzyme source for determination of AChE activity, using acetylcholine bromide as substrate. Enzyme aliquots (50 µL) and DTNB (100 µL of 0.01 M) were added to 0.1 M phosphate buffer (pH 8.0, 2.8 mL). Mixtures were incubated at 37ºC for 15 min. Reactions were started by adding acetylcholine bromide (30 µL) followed by incubation at 37ºC for 10 min. Absorbance was measured at 412 nm using a UV 2000-Spectrophotometer (NanoDrop, www.nanodrop.com). Time-courses of AChE activity were examined, and each treatment was corrected by blanks for nonenzymic hydrolysis. All the experiments were performed in triplicate. Inhibition percentage of AChE activity was calculated as follows:

$$\text{AChE inhibition (\%)}=\frac{OD_B-OD_T}{OD_B}\times 100$$

where OD_B is the optical density of blank enzyme and OD_T is the optical density of treatment.

Statistical analyses

In the experiment, the fumigant, contact, and repellent activities and inhibition on AChE activity of EOs were compared using analysis of variance (ANOVA) followed by Duncan's test for multiple -comparison ($P < 0.05$). The recorded mortality data in fumigant and contact toxicity tests were adjusted for mortality in the control using Abbott's formula, analyzed by one-way ANOVA, and means were compared using Duncan's test at $P < 0.05$ through an SPSS version 17.0 software package (IBM, www.ibm.com) in Microsoft Windows 7 operating system (www.microsoft.com). The figure of inhibition on AChE activity was drawn by SigmaPlot version 10.0 software (www.sigmaplot.com).

Results

The main ingredients of test EOs

According to the GC-MS data (Table 1), the main ingredient of *C. limonum* was limonene, and *L. cubea* was mainly constituted of D-limonene, (Z)-3, 7-dimethyl-, 2, 6- octadienal, and (E)-3, 7-dimethyl-, 2, 6-octadienal. *C. cassia* mainly contained methyl salicylate and (E)-cinnamaldehyde, and *A. sativum* mostly was composed of diallyl disulphide, di-2-propenyl trisulfide, and di-2-propenyl tetrasulfide.

Fumigant activity

Fumigant activities of EOs on the adults of *A. diaperinus* showed that *A. sativum* had the strongest fumigant activities 48 to 96 hr post-treatment, followed by *C. limonum*, *C. cassia*, and *L. cubeba* (Table 2). No insect mortality was observed in the control.

The toxicities of *A. sativum* and *C. limonum* seemed to be stronger on 6th instars larvae (Table 3) than on adults (Table 4). Neverthe-

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less, the toxicities of the EOs were almost equitoxic (overlapping confidence intervals) at 96 hr after treatment. No insect mortality was observed in the control.

Contact activity

The contact activities of EOs on 6th instar *A. diaperinus* are shown in Table 5. The activity of *A. sativum* was the highest, followed by *L. cubeba*.. However, the contact activities of the other two EOs were not ideal, causing < 50% adjusted mortality during the experiment period.

The contact toxicity of *A. sativum* on 6th instars of *A. diaperinus* was stronger than that of *L. cubeba*. The lower LC_{50} value of *A. sativum* was recorded 24 hr after treatment and was not equitoxic with that of *L. cubeba* (not overlapping confidence intervals). Nevertheless, the toxicity of *L. cubeba* was enhanced quickly and seemed to be equitoxic with that of *A. sativum* (48 hr after treatment (overlapping confidence intervals) (Table 6). No insect mortality was observed in the control.

Repellent activity

The repellent activity of *A. sativum* was the strongest among the tested oils, followed by *L. cubeba* (both significant). The repellent activities of the other two EOs were not significant 12, 24, and 48 hr after treatment (Table 7). Nevertheless, the repellent activities of all EOs decreased with the elongation of experiment.

Inhibiton of EOs on AChE activity

A. sativum showed the strongest inhibition on AChE activity in 6th instar *A. diaperinus*, followed by *C. limonum*, *L. cubeba*, *C. cassia.* All the essential oilss inhibition activities were significant ($P < 0.01$). Meanwhile, the inhibitions of EOs on AChE activity also became stronger and stronger with time and displayed time-course effects. Inhibitions of all oils except for *A. sativum* were < 50% at 36 hr, but quickly enhanced from 48 hr to 96 hr (Figure 1).

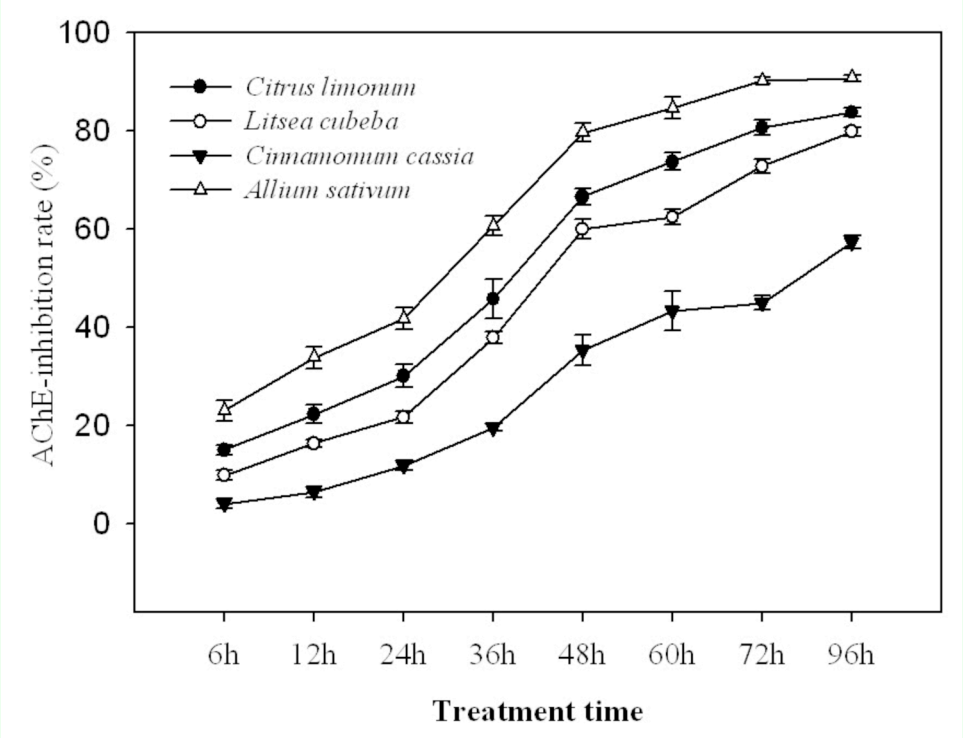

Figure 1. The time-course of EOs on AChE activity of 6th instar *A. diaperinus*. Note: The AChE activity of tested EOs on 6th instar *A. diaperinus* was the average values of triplication. High quality figures are available online.

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Discussion

The results on the compositions of the *A. sativum* (Park and Shin 2005; Almeida et al. 2009; Yang et al. 2010; Martinez-Velazquez et al. 2011), *C. limonum* (Almansa et al. 2002; Ponce et al. 2004; Moreira et al. 2005; Pouvova et al. 2008; Rekha and Vijayalakshmi 2010), and *L. cubeba* (Jiang et al. 2009; Seo et al. 2009) essential oils are in agreement with literature data on other insects. The fumigant, contact, and repellent activities of the tested EOs on adults and/or nymphal stages of *A. diaperinus* in present study showed that essential oils obtained from Sichuan Province acted against *A. diaperinus*. Furthermore, the efficacies of fumigant and contact activities were relatively enhanced with increasing amount of doses and exposure times. Similar findings were also shown in other investigations showing the toxic efficacy of EOs from some other aromatic plant against insects and mites (Benzi et al. 2009).

Some ingredients (such as sulphide, limonene, pinene, etc.) from EOs have been shown to act as the major role of insecticidal activities, including fumigant, contact, and repellent. Yang et al. (2010) reported that *A. sativum* used against adult rice weevils, *Sitophilus oryzae* (L.), and red flour beetles, *Tribolium castaneum* (Herbst), could reduce the survival of eggs or larvae to adult stage. Gusmão et al. (2013) reported the main ingredients of EOs extracted from *Eucalyptus staigeriana* were limonene (28.75%), geranial (15.20%), and neral (12.16%), and from *Foeniculum vulgare* were limonene (41.82%), (E)-anethole (17.91%), and α-pinene (11.13%), and contact and fumigation toxicities of *F. vulgare* and *E. staigeriana* on the adults of *Callosobruchus maculates* had LC_{50} values of 178.13, 345.57 μg/mL and 2.58, 7.85 μL/L of air, respectively. Yeom et al. (2013) also researched 11 kinds of Myrtaceae plant essential oils, including *Eucalyptus polybractea*, *Eucalyptus smithii*, and so on, and they displayed 100% fumigant toxicity against adult male German cockroaches (7.5 mg/L air). The essential oil constituents, including terpinolene, α-terpinene, and terpinen-4-ol, demonstrated strong fumigant toxicity against adult males and females, and eugenol, isoeugenol, methyl eugenol, and terpinen-4-ol showed strong contact toxicity against adult males of *B. germanica.* According to our results, the compounds of sulphide (diallyl-disulphide, methyl-2-propenyl-risulfide, di-2-propenyl-tetrasulfide, etc.), limonene (limonene, D-limonene), and pinene (*α*-Pinene, *β*-Pinene) were the main ingredients of EOs extracted from *A. sativum*, *C. limonum*, and *L. cubeba* and could lead to the insecticidal activities on tested pests.

AChE is an important target for insecticides, and inhibition by phytochemicals from plant essential oils has been reported in previous research (Abdelgaleil et al. 2009; Kang et al. 2013). Yeom et al. (2013) found isoeugenol from Myrtaceae plant essential oils exhibited inhibition activity against male acetylcholinesterase, with IC_{50} values of 0.22 mg/mL. Kim et al. (2013) also revealed some compounds from Apiaceae essential oils on acetylcholinesterase inhibitory activity of *Sitophilus oryzae* and found that *α*-Pinene showed the highest inhibition rate (97.36%), followed by *β*-pinene (54.96%) and limonene (51.23%) at a concentration of 1 mg/mL. The IC_{50} of α-pinene reached 0.019 mg/mL. Based on our data, the EOs of *A. sativum*, *C. limonum*, and *L. cubeba* all displayed good inhibition on AChE activity of 6th instar *A. diaperinus*. We did not detect the inhibition on AChE activity and IC_{50} of singular ingredients on the tested pest, which could lead to

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insecticidal activity (including sulphide, limonene and pinene, eugenol), but we can assume how these compounds function based on the previously mentioned reports.

In general, our results indicate that EOs of *A. sativum*, *C. limonum*, and *L.cubeba* and their components could be developed for managements of *A. diaperinus*. For the practical use of these oils and their constituents as new control agents, the safety of the oils and their components to humans and non-target organisms and their mode of action should be further investigated.

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References

Abbott WS. 1925. A method of computing the effectiveness of an insecticide. *Journal of Economic Entomology* 18: 265–267.

Abdelgaleil SAM, Mohamed MIE, Badawy MEI, El-arami SAA. 2009. Fumigant and contact toxicities of monoterpenes to *Sitophilus oryzae* (L.) and *Tribolium castaneum* (Herbst) and their inhibitory effects on acetylcholinesterase activity. *Journal of Chemical Ecology* 35: 518–525.

Almansa MS, Hernandez JA, Jimenez A, Botella MA, Sevilla F. 2002. Effect of salt stress on the superoxide dismutase activity in leaves of *Citrus limonum* in different rootstock-scion combinations. *Biologia Plantarum* 45: 545–549.

Almeida TF, Camargo M, Panizzi RDC. 2009. Effect of plant extract on control of *Colletotrichum acutatum* the causal agent of the black spot of strawberry. *Summa Phytopathologica* 35: 196–201.

Benzi VS, Murray AP, Ferrero AA. 2009. Insecticidal and insect-repellent activities of essential oils from Verbenaceae and Anacardiaceae against *Rhizopertha dominica*. *Natural Product Communications* 4: 1287–1290.

Copping LG, Menn JJ. 2000. Biopesticides: a review of their action, applications and efficacy. *Pest Management Science* 56: 651–676.

Cosimi S, Rossi E, Cioni PL, Canale A. 2009. Bioactivity and qualitative analysis of some essential oils from Mediterranean plants against stored-product pests: Evaluation of repellency against *Sitophilus zeamais* Motschulsky, *Cryptolestes ferrugineus* (Stephens) and *Tenebrio molitor* (L.). *Journal of Stored Products Research* 45: 125–132.

Dalila H, Pier LC, Monia BHK, Guido F, Mohamed HBH. 2012. Chemical composition and bioactivities of three *Chrysanthemum* essential oils against *Tribolium confusum* (duVal) (Coleoptera: Tenebrionidae). *Journal of Pesticide Science* 3: 367–379.

Deng Y, Wang J, Ju Y, Zhang H. 2004. Comparison of fumigation activities of 9 kinds of essential oils against the adults of maize weevil, *Sitophilus zeamaise* Motschulsky (Coleoptera:Curculionidae). *Chinese Journal of Pesticide Science* 6: 85–88.

Gusmão NMS, Oliveira JV, Navarro DMAF, Dutra KA, Silva WA, Wanderley MJA. 2013. Contact and fumigant toxicity and repellency of *Eucalyptus citriodora* Hook., *Eucalyptus*

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staigeriana F., *Cymbopogon winterianus* Jowitt and *Foeniculum vulgare* Mill. essential oils in the management of *Callosobruchus maculatus* (FABR.) (Coleoptera: Chrysomelidae). *Journal of Stored Products Research* 54: 41–47.

Jiang ZL, Akhtar Y, Bradbury R, Zhang X, Isman MB. 2009. Comparative toxicity of essential oils of *Litsea pungens* and *Litsea cubeba* and blends of their major constituents against the cabbage looper, *Trichoplusia ni*. *Journal of Agricultural and Food Chemistry* 57: 4833–4837.

Jin M, Zhang Y, Ye J, Huang C, Zhao M, Liu W. 2010. Dual enantioselective effect of the insecticide bifenthrin on locomotor behavior and development in embryonic-larval zebrafish. *Environmental Toxicology Chemistry* 29: 1561–1567.

Li XY, Qian Y, Wang X, Chen J, Song Y. 2009. Preliminary study on the community structure of storage insects of *Juncus effuses* L. products in Wujiang area. *Journal of Anhui Agricultural Science* 37: 17545–17546, 17548.

Kang JS, Kim E, Lee SH, Park IK. 2013. Inhibition of acetylcholinesterases of the pinewood nematode, *Bursaphelenchus xylophilus*, by phytochemicals from plant essential oils. *Pesticide Biochemistry Physiology* 105: 50–56.

Kim SW, Kang J, Park IK. 2013. Fumigant toxicity of Apiaceae essential oils and their constituents against *Sitophilus oryzae* and their acetylcholinesterase inhibitory activity. *Journal of Asia-Pacific Entomology* 16: 443–448.

Mahnaz K, Alireza F, Hassan V, Mahdi S, Mohammad Reza A, Abbas H. 2012. Larvicidal activity of essential oil and methanol extract of Nepeta menthoides against malaria vector *Anopheles stephensi. Asian Pacific Journal of Tropical Medicine* 5: 962–965.

Martinez-Velazquez M, Rosario-Cruz R, Castillo-Herrera G, Flores-Fernandez JM, Alvarez AH, Lugo-Cervantes E. 2011. Acaricidal effect of essential oils from *Lippia graveolens* (Lamiales: Verbenaceae), *Rosmarinus officinalis* (Lamiales: Lamiaceae), and *Allium sativum* (Liliales: Liliaceae) against *Rhipicephalus microplus* (Acari: Ixodidae). *Journal of Medical Entomology* 48: 822–827.

Moreira MR, Ponce AG, Del Valle CE, Roura SI. 2005. Inhibitory parameters of essential oils to reduce a foodborne pathogen. *Lwt-Food Science and Technology* 38: 565–570.

Nesci A, Barra P, Etcheverry M. 2011. Insecticidal activity of synthetic antioxidants, natural phytochemicals, and essential oils against an Aspergillus section Flavi vector (*Oryzaephilus surinamensis* L.) in microcosm. *Journal of Pest Science* 84: 107–115.

Park IK, Shin SC. 2005. Fumigant activity of plant essential oils and components from garlic (*Allium sativum*) and clove bud (*Eugenia caryophyllata*) oils against the Japanese termite (*Reticulitermes speratus* Kolbe). *Journal of Agricultural and Food Chemistry* 53: 4388–4392.

Ponce AG, Del Valle CE, Roura SI. 2004. Natural essential oils as reducing agents of peroxidase activity in leafy vegetables. *Lebensmittel-Wissenschaft Und-Technologie-Food Science and Technology* 37: 199–204.

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Pouvova D, Kokoskova B, Pavela R, Rysanek P. 2008. Effectivity of plant essential oils against *Clavibacter michiganensis*, in vitro. *Zemdirbyste-Agriculture* 95: 440–446.

Rekha CR, Vijayalakshmi G. 2010. Influence of natural coagulants on isoflavones and antioxidant activity of tofu. J*ournal of Food Science and Technology-Mysore* 47: 387–393.

Santos FFC, Silva Amaral LD, Rodrigues-Filho E. 2011. Composition of essential oils from *Cupressus lusitanica* and a *Xylariaceous fungus* found on its leaves. *Biochemical Systematics and Ecology* 39: 485–490.

Seo SM, Kim J, Lee SG, Shin CH, Shin SC, Park IK. 2009. Fumigant antitermitic activity of plant essential oils and components from Ajowan (*Trachyspermum ammi*), Allspice (*Pimenta dioica*), Caraway (*Carum carvi*), Dill (*Anethum graveolens*), Geranium (*Pelargonium graveolens*), and Litsea (*Litsea cubeba*) oils against Japanese termite (*Reticulitermes speratus* Kolbe). *Journal of Agricultural and Food Chemistry* 57: 6596–6602.

Stefanazzi N, Stadler T, Ferrero A. 2011. Composition and toxic, repellent and feeding deterrent activity of essential oils against the stored-grain pests *Tribolium castaneum* (Coleoptera: Tenebrionidae) and *Sitophilus oryzae* (Coleoptera: Curculionidae). *Pest Management Science* 67: 639–646.

Wang CF, Yang K, Zhang HM, Cao J, Fang R, Liu ZL, Du SS, Wang YY, Deng ZW, Zhou L. 2011. Components and insecticidal activity against the maize weevils of *Zanthoxylum schinifolium* fruits and leaves. *Molecules* 16: 3077–3088.

Wang J, Zhu F, Zhou XM, Niu CY, Lei CL. 2006. Repellent and fumigant activity of essential oil from *Artemisia vulgaris* to *Tribolium castaneum* (Herbst) (Coleoptera : Tenebrionidae). *Journal of Stored Products Research* 42: 339–347.

Yao YJ, Cai WL, Yang CJ, Xue D, Huang YZ. 2008. Isolation and characterization of inseticidal activity of (Z)-asarone from *Acorus calamus* L. *Insect Science* 15: 229–236.

Yang FL, Liang GW, Xu YJ, Lu YY, Zeng L. 2010. Diatomaceous earth enhances the toxicity of garlic, *Allium sativum*, essential oil against stored-product pests. *Journal of Stored Products Research* 46: 118–123.

Yeom HJ, Kang JS, Kim GH, Park IK. 2012. Insecticidal and acetylcholine esterase inhibition activity of apiaceae plant essential oils and their constituents against adults of *German cockroach* (Blattella germanica). *Journal of Agricultural and Food Chemistry* 60: 7194–7203.

Yeom HJ, Kang J, Kim SW, Park IK. 2013. Fumigant and contact toxicity of Myrtaceae plant essential oils and blends of their constituents against adults of German cockroach (*Blattella germanica*) and their acetylcholinesterase inhibitory activity. *Pesticide Biochemistry and Physiology* 107: 200–206.

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Table 1. Chemical constituent of essential oils extracted from *C. limonum, L. cubeba, C. cassia*, and *A. sativum*. RT = retention time.

Essential oil	Compound	Composition %	RT (min)	Retention index
C. limonum	Limonene	64.53	6.603	1032.8
	1-methyl-4-(1-methylethyl)-1,4-Cyclohexadiene	17.36	7.153	1041.4
	(Z)-3,7-dimethyl-2,6-Octadienal	2.3	10.444	1092.8
	(E)-3,7-dimethyl-2,6-Octadienal	3.44	10.968	1200.9
	(Z)- 3,7-dimethyl-2,6-Octadien-1-ol, acetate	1.26	12.522	1223.8
	2,6-dimethyl-6-(4-methyl-3-pentenyl)-Bicyclo[3.1.1] hept-2-ene	1.54	13.741	1241.8
	(S)-1-methyl-4-(5-methyl-1-methylene-4-hexenyl)-Cyclohexene	1.9	14.874	1258.5
	Other compounds	7.64	-	-
L. cubeba	α-Pinene	1.61	6.571	1032.3
	β-Pinene	1.13	7.993	1054.5
	D-limonene	12.34	10.171	1088.5
	3,7-dimethyl-1,6-octadien-3-ol	1.77	13.016	1231.1
	(Z)-3,7-dimethyl-2,6-octadienal	25.44	19.835	1435.6
	(E)-3,7-dimethyl-2,6-octadienal	33.93	21.459	1462
	Caryophyllene	1.41	26.641	1652.7
	Other compounds	22.38	-	-
C. cassia	Benzyl-alcohol	0.36	7.704	1050
	Methyl salicylate	10.77	12.168	1218.6
	(E)- cinnamaldehyde	74.43	14.729	1256.4
	eugenol	5.5	17.22	1293.1
	Tetracyclo[5.3.0.0<2,6>.0<3,10>]deca-4,8-diene	0.19	18.198	1408.4
	3-phenyl-2-Propenoic acid	4.78	19.858	1435.7
	7-Methyl-1-naphthol	1.04	21.648	1465.1
	Other compounds	2.93	-	-
A. sativum	Diallyl-disulphide	18.2	13.917	1244.4
	methyl-2-propenyl- trisulfide	6.92	16.901	1288.4
	1,3,5-Trithiane	1.19	17.393	1295.7
	di-2-propenyl-trisulfide	27.94	27.478	1668.1
	7-Chloro-s-triazolo[1,5-c]pyrimidin-5-(1H)-one	1.96	30.087	1817.6
	di-2-propenyl-tetrasulfide	3.58	31.595	1846.6
	di-2-propenyl-tetrasulfide	14.57	41.557	2169.7
	Other compounds	25.65	-	-

Table 2. The fumigant activity of EOs on *A. diaperinus* adults.

Essential oil	Number of insects tested	Adjusted mortality of *A. diaperinus* (%) (± SE)	
		48 hr	96 hr
C. limonum	90	34.4 ± 4.0 b[a]	53.3 ± 3.8 b[a]
L. cubeba	90	12.2 ± 2.9 c	23.3 ± 3.8 c
C. cassia	90	18.9 ± 2.9 c	31.1 ± 4.8 c
A. sativum	90	71.1 ± 4.0 a	85.6 ± 2.9 a
Control	90	0 d	0 d
$F_{4,14}$	-	76.3	85.1
P	-	< 0.001	< 0.001

[a]Means within a column followed by the same letters are significantly different ($P < 0.05$) as determined by Duncan's test.

Table 3. LC_{50} values from fumigant activity of *A. sativum* and *C. limonum* to 6th instar *A. diaperinus*.

Essential oil	Treatment time (hr)	Number of insects tested	LC_{50} (μL/L) (95% CI[a])	LC_{95} (μL/L) (95% CI[a])	Slope ± Se	Chi–square (d.f.)	*P*-value*
A. sativum	48	540	6.8 (5.5–8.4)	123.5 (74.6–252.2)	1.30 ± 0.12	1.79 (4)	< 0.01
	96	540	4.2 (3.4–5.2)	70.8 (45.6–130.6)	1.29 ± 0.12	1.90 (4)	< 0.01
C.limonum	48	540	8.8 (7.0–11.5)	256.0 (131.3–692.4)	1.13 ± 0.12	1.65 (4)	< 0.01
	96	540	5.8 (4.6–7.2)	141.2 (80.4–320.3)	1.18 ± 0.12	1.16 (4)	< 0.01
Control	–	90	–	–	–	–	–

[a]LC_{50} or LC_{95} values are considered significantly different when the 95% confidence intervals (CI) do not overlap. *Goodness-of-fit test is significant at $P < 0.05$.

Table 4. LC_{50} values from fumigant activity of *A. sativum* and *C. limonum* to *A. diaperinus* adults.

Essential oil	Treatment time (hr)	Number of insects tested	LC_{50} (μL/L) (95% CI[a])	LC_{95} (μL/L) (95% CI[a])	Slope ± Se	Chi–square (d.f.)	*P*-value*
A. sativum	48	540	8.4 (6.9–10.4)	131.3 (80.5–260.8)	1.38 ± 0.13	3.50 (4)	< 0.05
	96	540	5.7 (4.3–7.7)	85.1 (45.5–239.8)	1.40 ± 0.12	4.64 (4)	< 0.05
C.limonum	48	540	11.8 (9.5– 15.1)	214.9 (121.0–491.4)	1.30 ± 0.13	1.75 (4)	< 0.01
	96	540	7.2 (5.9–8.9)	125.1 (76.0–252.8)	1.33 ± 0.12	1.76 (4)	< 0.01
Control	–	90	–	–	–	–	–

[a]LC_{50} or LC_{95} values are considered significantly different when the 95% confidence intervals (CI) do not overlap. *Goodness-of-fit test is significant at $P < 0.05$.

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Table 5. The contact activity of EOs on 6th instar *A. diaperinus*.

Essential oil	Number of insects tested	Adjusted mortality of *A. diaperinus* (%) (± SE)	
		24 hr	48 hr
C. limonum	90	30.0 ± 2.0 c[a]	46.7 ± 1.9 c[a]
L. cubeba	90	43.3 ± 3.3 b	64.4 ± 2.9 b
C. cassia	90	30.0 ± 3.3 c	27.8 ± 2.9 c
A. sativum	90	64.4 ± 2.9 a	76.7 ± 3.9 a
Control	90	0 d	0 d
$F_{4,14}$	-	79.6	120.9
P	-	0.001	0.001

[a]Means within a column followed by the same letters are not significantly different ($P > 0.05$) as determined by Duncan's test.

Table 6. The contact toxicity of EOs on 6th instar *A. diaperinus*.

Essential oil	Treatment time (hr)	Number of insects tested	LC_{50} (µL/L) (95% CI[a])	LC_{95} (µL/L) (95% CI[a])	Slope ± Se	Chi–square (d.f.)	*P*-value*
A. sativum	24	540	33.0 (27.7–39.5)	378.8 (256.5–641.5)	1.55 ± 0.13	0.25 (4)	< 0.001
	48	540	19.0 (14.0–25.0)	191.9 (116.3–430.3)	1.64 ± 0.13	5.24 (4)	< 0.05
C.limonum	24	540	49.0 (40.4– 60.8)	700.3 (431.5–1376.9)	1.42 ± 0.12	1.88 (4)	< 0.05
	48	540	24.0 (19.7– 28.9)	338.7 (224.5–596.4)	1.43 ± 0.13	1.57 (4)	< 0.01
Control	–	90	–	–	–	–	–

[a]LC_{50} or LC_{95} values are considered significantly different when the 95% confidence intervals (CI) do not overlap. *Goodness-of-fit test is significant at $p < 0.05$.

Table 7. Repellent activity of EOs on 6th instar *A. diaperinus*.

Essential oil	Repellence index after 12 hr treatment (%) (± SE)			Repellence index after 24 hr treatment (%) (± SE)			Repellence index after 48 hr treatment (%) (± SE)		
	200[a]	400	800	200	400	800	200	400	800
C. limonum	44.6 ± 1.2 b[b]	54.7 ± 3.2 b	66.5 ± 3.7 b	33.9 ± 2.0 c	44.7 ± 3.2 b	57.6 ± 2.7 c	27.1 ± 2.0 b	34.7 ± 4.2 b	51.1 ± 2.9 c
L. cubeba	68.7 ± 6.1 a	80.0 ± 1.9 a	88.3 ± 0.9 a	60.9 ± 2.8 a	72.9 ± 1.6 a	77.2 ± 2.0 b	50.9 ± 2.8 a	62.9 ± 1.6 a	69.3 ± 3.0 b
C. cassia	54.8 ± 3.7 b	54.0 ± 2.0 b	71.5 ± 4.3 b	45.5 ± 3.0 b	44.0 ± 2.0 b	62.6 ± 2.6 c	34.7 ± 4.3 b	38.0 ± 1.0 b	48.5 ± 1.0 c
A. sativum	75.1 ± 1.7 a	82.4 ± 1.2 a	90.4 ± 1.6 a	64.9 ± 3.4 a	75.3 ± 1.0 a	83.9 ± 1.4 a	57.5 ± 2.9 a	67.4 ± 2.1 a	76.4 ± 2.0 a
Control	0 c	0 c	0 c	0 d	0 c	0 d	0 c	0 c	0 d
$F_{4,14}$	80.2	277.9	191.8	104.5	255.6	269.1	68.1	142.3	196.4
P	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

[a]µg/cm^2 filter paper. Means in the same column followed by the same letters differ significantly ($P < 0.05$) according to ANOVA. The number of insects tested for each treatment was 90.

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