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Repellent Activity of Eight Essential Oils of Chinese Medicinal Herbs to *Blattella germanica* L.

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Abstract: Eight essential oils of Chinese medicinal herbs (*Angelica sinensis*, *Curuma aeruginosa*, *Cyperus rotundus*, *Eucalyptus robusta*, *Illicium verum*, *Lindera aggregate*, *Ocimum basilicum*, *and Zanthoxylum bungeanum*) were obtained by hydrodistillation and the essential oil of *Eucalyptus robusta* leaves was analyzed by gas chromatography-mass spectrometry (GC-MS). A total of 22 components of the essential oil of *E. robusta* were identified. The principal compounds in *E. robusta* essential oil were α-pinene (28.74%) and 1,8-cineole (27.18%), spathulenol (6.63%), globulol (6.53%) and *ρ*-menth-1-en-8-ol (5.20%). The 8 essential oils and two main components, α-pinene and 1, 8-cineole of the essential oil of *E. robusta* were evaluated repellency against nymphs of the German cockroaches. Strong repellency (Class V) was obtained for *Cyperus rotundus* and *Eucalyptus robusta* essential oils and α-pinene and 1, 8-cineole. However, *Illicium verum* essential oil possessed weak (Class I) repellency. At a concentration of 5 ppm, all the 8 essential oils and the two compounds showed repellent activity after one hour exposure. At 1 ppm concentration, essential oil of *Cyperus rotundus* showed strong repellency and Class IV repellency was obtained for essential oil of *E. robusta* and the two compounds after one hour exposure. However, essential oils of *I. verum* and *Lindera aggregata* showed strong attractiveness to the German cockroaches at a concentration of 1 ppm.

Keywords: *Blattella germanica*; *Eucalyptus robusta*; repellency; essential oil composition; α-pinene; 1, 8-cineole.

1. Introduction

The German cockroach, *Blattella germanica* (L.), is an important pest of homes, restaurants, and commercial food processing facilities worldwide. They are a major public health concern in

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hospitals, kitchens, and food manufacturing plants because they are able to carry a variety of bacteria and other pathogenic organisms. Body parts, cast skins, and feces of cockroaches are human allergens, 2nd in importance only to the house dust mites [1]. Cockroaches are urban pests that can be difficult to control, especially in environments such as schools, hospitals, and elder care facilities. Concern over health implications from the use of residual and broad insecticidal spray treatments has been impetus for research on alternative methods. Repellents may play a very important role in some situations or in some special space where the insecticides are not able to use [2]. Moreover, highly repellent insecticides, such as pyrethrum, can be useful when used to detect infestations in areas where visible inspection is limited [3] and one of the methods to assess relative abundance of cockroaches (flush and count) [4]. Essential oils derived from plants have been traditionally used to protect stored grain and to repel flying insects in the home [5]. Many essential oils have been screened for repellent activity against cockroaches and some of them possess potential to be developed as natural repellents [6-10]. In the present study, an attempt has been made to evaluate the repellent activity of essential oils extracted from 8 Chinese medicinal herbs (*Angelica sinensis*, *Curuma aeruginosa*, *Cyperus rotundus*, *Eucalyptus robusta*, *Illicium verum*, *Lindera aggregate*, *Ocimum basilicum*, *and Zanthoxylum bungeanum*) against German cockroaches. Essential oil of *Eucalyptus robusta* was analysed by GC and GC-MS and its two main components (α-pinene and 1, 8-cineole) of the oil were also evaluated for repellent activity against German cockroaches.

2. Materials and Methods

2.1. Plant Material

Eight Chinese medicinal herbs (5 kg each) were purchased from Anguo Herb Market (Anguo, Hebei Province, China) (Table 1). The herbs were identified, and the voucher specimens were deposited at the Department of Entomology, China Agricultural University. The herbs were firstly ground to powder using a grinding mill (Retsch Muhle, Germany) and were subjected to hydrodistillation using a modified Clevenger-type apparatus for 6 h and extracted with *n*-hexane. Anhydrous sodium sulphate was used to remove water after extraction. Essential oils were stored in airtight containers in a refrigerator at 4°C for subsequent experiments. (+)-α-Pinene, eucalyptus (1,8-cineole, 99%) and permethrin were purchased from Sigma-Aldrich Chemical Co. (P.O.Box 14460, St. Louis, MO 63178, USA). Permethrin was used as a positive control, because it has been widely used in the survey of cockroach population density in China.

2.2. Insects

German cockroaches were obtained from laboratory cultures maintained at 26-28°C, 70-80% RH, and a photoperiod of 12:12 (L:D) h. Dry rat food (Laboratory Animal Centre, Chinese Academy of Medicinal Sciences, Beijing) and water were supplied ad libitum. The unsexd nymphs used in the experiments was about one week old after hacthing.

2.3. Repellent assays

Circular white filter paper No. 40 (9 cm diameter, Whatman international Ltd. Maidstone, England), divided in two halves, were used [11]. One of the halves was treated with 0.5 ml of acetone; the other half was treated with 0.5 ml acetone solutions of essential oils/compounds. Each essential oil was assayed at two concentrations of 5 and 1 ppm (w/v) after preliminary experiments. After solvent evaporation (2 min), each treated half-disc was then attached lengthwise, edge-to-edge, to a control half-disc with adhesive tape to form a full disc. Precautions were taken so that the attachment did not prevent the free movement of the insects from one half to another, but a small distance between the filter-paper halves was left to prevent seepage of the test samples from one half to the other. Each filter paper was then placed in a petri dish (Diameter 9 cm) covered with fluon to prevent insects from escaping. The petri dish had a seam orientated in one of four randomly selected directions to avoid any incidental stimuli affecting the distribution of insects. The orientation of the seam was changed in replicates. Ten nymphs of cockroaches were released in the middle of each filter-paper circle and a plastic cover with some small holes was placed on the petri dish. Five replicates were used. Counts of the insects present on each filter paper disc half were made after 1 hr and subsequently at hourly intervals up to the fourth hour. No significant difference was detected between the repellency of acetone impregnated and plain filter papers in tests designed to check any possible influence of acetone on the insects. The average of the counts was converted to percentage repellency (PR) as PR = 2 (C-50) Where C is the percentage of insects on the untreated half. Positive values (+) express repellency and negative values (-) attractancy. The averages were then categorised according to the following scale (Table 1) [12,13]

Table 1. The scale used to categorise repellency of the essential oils

Class	Percent repulsion (%)
0	0.01-01
I	0.1 - 20
II	20.1-40
III	40.1-60
IV	60.1-80
V	80.1-100

Percent repellency (PR) was analysed using analysis of variance (ANOVA) and Tukey's tests after transforming them into arcsine percentage values.

2.4. Gas Chromatography-Mass Spectrometry

The essential oil of *Eucalyptus robusta* leaves was subjected to GC-MS analysis on an Agilent system consisting of a model 6890N gas chromatograph, a model 5973N mass selective detector (EIMS, electron energy, 70 eV), and an Agilent ChemStation data system. The GC column was an HP-5ms fused silica capillary with a 5% phenyl-methylpolysiloxane stationary phase, film thickness of 0.25 µm, a length of 30 m, and an internal diameter of 0.25 mm. The GC settings were as follows: the initial oven temperature was held at 60°C for 1 min and ramped at 10°C min^{-1} to 180°C for 1 min, and then ramped at 20°C min^{-1} to 280°C for 15 min. The injector temperature was maintained at 270°C. The samples (1 µL) were injected neat, with a split ratio of 1: 10. The carrier gas was helium at flow rate of 1.0 mL min^{-1}. Spectra were scanned from 20 to 550 m/z at 2 scans s^{-1}. Most constituents were identified by gas chromatography by comparison of their retention indices with those of the literature [2,3] or with those of authentic compounds available in our laboratories. The retention indices were determined in relation to a homologous series of *n*-alkanes (C_8–C_{24}) under the same operating conditions. Further identification was made by comparison of their mass spectra with those stored in NIST 05 and Wiley 275 libraries or with mass spectra from literature [14]. Component relative percentages were calculated based on GC peak areas without using correction factors.

3. Results and Discussion

The information of essential oils of 8 Chinese medicinal herbs was summarized in Table 2. A total of 22 components were identified in the essential oil of *E. robusta* leaves, accounting for 97.61% of the total oil (Table 3). The main components of the oil were α-pinene (28.74%) and 1,8-cineole (27.18%) followed by spathulenol (6.63%), globulol (6.53%) and *ρ*-menth-1-en-8-ol (5.20%). In the previous studies, major components of the essential oil of *E. robusta* from Yunnan province China were 1, 8-cineole (66.42%) and α-pinene (27.18%) [22]. Moreover, the major constituent in *E. robusta* oil from Brazil was α-pinene (73.0%) [23]. α-Pinene and 1, 8-cineole were two main components of *E. robusta* essential oil although there were geographic variations in chemical composition of the oil.

At a concentration of 5 ppm, all the essential oils and the two compounds showed repellent activity against the German cockroaches after one hour exposure. Strong repellency (Class V) was obtained from *C. rotundus* and *E. robusta* essential oils and α-pinene and 1,8-cineole after one hour exposure (Table 4). Essential oils of *C. aeruginosa* and *O. basilicum* showed the same level of repellency (Class IV) as that of positive control, permethrin. However, essential oils of *A. sinensis*, *L. aggregata*, and *Z. bungeanum* possessed Class III repellency against German cockroaches while *I. verum* essential oil showed weak repellency (Class I) after one hour exposure (Table 4). Repellency of 6 essential oils decreased to Class II and I and that of *C. aeruginosa* and *I. verum* essential oils decreased to zero after four hours exposure (Table 4).

Table 2. Essential oils of Chinese medicinal herbs used for the repellent experiment.

Essential oil	Family	Source	Main components	Ref.
Angelica sinensis (Oliv.) Diels	Umbelliferae	Roots	Ligustilide, *n*-butyldenephthalide	15
Curuma aeruginosa Roxb	Zingiberaceae	Roots	Curcumenol, curzerenone	16
Cyperus rotundus L.	Cyperaceae	Tubers	α-Cyperone, cyperene	17
Eucalyptus robusta Smith	Myrtaceae	Leaves	1,8-Cineole, α-pinene	-
Illicium verum Hook. f.	Magnoliaceae	Fruits	*trans*-Anethole, limonene	18
Lindera aggregata (Sims) Kosterm	Lauraceae	Root	α-Phellandrene, lindene, linderene	19
Ocimum basilicum L	Labiatae	Leaves	Linalool, (*Z*)-cinnamic acid Me ester	20
Zanthoxylum bungeanum Maxim.	Rutaceae	Fruits	Terpinen-4-ol, 1,8-cineole	21

Table 3 Chemical composition of essential oil of *Eucalyptus robusta* leaves.

RI	Compound	Percent Composition
931	α-Pinene	28.74
1033	1,8-Cineole	27.18
1078	Linalool oxide	0.40
1094	Linalool	1.17
1109	β-Fenchol	1.10
1138	*trans*-Pinocarveol	3.27
1167	Borneol	3.00
1179	Terpinen-4-ol	1.18
1196	Myrtenol	0.19
1208	ρ-Menth-1-en-8-ol	5.24
1226	*cis*-Carveol	0.23
1432	β-Gurjunene	0.95
1426	β-Caryophyllene	2.99
1437	Aromadendrene	1.58
1454	α-Humulene	0.56
1458	*allo*-Aromadendrene	1.12
1489	Eremophilene	0.59
1575	Spathulenol	6.63
1581	Globulol	6.53
1585	*epi*-Globulol	0.77
1592	Viridiflorol	3.00
1640	α-Cadinol	0.70
	Total identified	97.61
	Monoterpenoids	71.70
	Sesquiterpenoids	25.91

Table 4. Repellency of 8 essential oils from Chinese medicinal herbs and two active components against German cockroaches at a concentration of 5 ppm

Essential oil	1 hr		2 hr		3 hr		4 hr	
Angelica sinensis	57.7±2.5d	III	43.3±2.9e	III	12.5±1.5d	I	4.3±0.9e	I
Curuma aeruginosa	68.9±2.6c	IV	52.1±2.7d	III	24.6±1.7c	II	0	-
Cyperus rotundus	89.4±2.5a	V	85.4±2.7ab	V	48.7±2.5b	III	36.5±1.6b	II
Eucalyptus robusta	81.2±1.7b	V	60.4±3.3c	IV	47.9±2.3b	III	27.4±0.8c	II
Illicium verum	14.3±2.6e	I	8.6±0.7g	I	4.3±0.4e	I	0	-
Lindera aggregata	58.5±4.1d	III	33.9±2.8f	II	28.3±2.1c	II	14.9±1.2d	I
Ocimum basilicum	72.2±1.7c	IV	64.5±2.5c	IV	20.5±1.9e	II	16.0±1.5d	I
Zanthoxylum bungeanum	58.0±4.2d	III	53.3±3.0d	III	48.8±3.9b	III	29.2±2.3c	II
Permethrin	77.9±3.2b	IV	65.4±2.6c	IV	66.5±5.3a	IV	19.3±1.6d	I
α-Pinene	84.8±2.4a	V	82.4±2.6b	V	72.1±3.3a	IV	52.9±3.5a	III
1,8-Cineole	89.4±1.9a	V	91.7±2.8a	V	69.3±2.9a	IV	56.6±2.8a	III

Values are the mean repellency (%) ± SEM of five replicates of 10 nymphs each. Means within a column followed by the same lower case letter are not significantly different ($P<0.05$, ANOVA and Tukey's tests).

Table 5. Repellency of 8 essential oils from Chinese medicinal herbs and two active components against German cockroaches at a concentration of 1 ppm

Essential oil	1 hr		2 hr		3 hr		4 hr	
Angelica sinensis	32.1±2.4d	II	8.4±0.5f	I	4.5±0.3e	I	0	-
Curuma aeruginosa	52.7±1.8c	II	42.7±1.4d	II	14.3±1.1d	I	0	-
Cyperus rotundus	84.5±2.2a	V	82.5±2.7a	V	47.9±2.2a	III	12.9±1.5d	I
Eucalyptus robusta	71.4±3.0b	IV	39.0±2.4d	II	33.3±1.5b	II	28.5±1.0c	II
Illicium verum	-73.4±1.1g	-	-58.0±2.1g	-	-24.1±1.7f	-	-8.3±0.5e	-
Lindera aggregata	-65.3±3.7lf	-	-56.2±3.4g	-	-43.2±2.3g	-	-38.5±2.4f	-
Ocimum basilicum	56.7±4.5c	III	52.4±2.3c	III	49.9±2.1a	III	28.4±1.8c	II
Zanthoxylum bungeanum	37.2±3.2d	II	28.2±2.2e	II	23.7±1.5c	II	12.4±0.7d	I
Permethrin	12.1±1.0e	I	8.2±0.6f	I	4.1±0.2e	I	0	-
α-Pinene	79.8±3.4ab	IV	69.2±1.9b	IV	48.9±1.8a	III	34.6±1.5b	II
1,8-Cineole	74.8±2.2b	IV	66.3±1.5b	IV	52.9±1.7a	III	44.7±1.8a	III

Values are the mean repellency (%) ± SEM of five replicates of 10 nymphs each. Means within a column followed by the same lower case letter are not significantly different ($P<0.05$, ANOVA and Tukey's tests).

At a conentation of 1 ppm, the positive control (permethrin) showed weak repellency (Class I) against *B. germanica* after one hour exposure. Strong repellency (Class V) was obtained from *C. rotundus* essential oil and the two compounds and *E. robusta* essential oil possessed Class IV repellency after one hour exposure. Class III repellent activity was obtained from *O. basilicum* essential oil. Essential oils of *A. sinensis*, *C. aeruginosa* and *Z. bungeanum* showed Class II repellency (Table 5). Interestingly, the essential oils of *I. verum* and *L. aggregata* showed strong attractiveness to *B. germanica* (Table 5). Compared with the positive control, the two compounds showed stronger repellent activity against the German cockroaches (Table 5).

α-Pinene has been shown to possess repellent activity against many insects, such as American cockroaches, *Periplaneta Americana* [22], mosquitoes [40], stored product insects [24] and common tick [25]. However, α-pinene also attracted several scolytids and associated beetles (e.g. *Tomicus piniperda*, *Hylurgops palliates*, *Glischrochilus quadripunctatus*, and *Thanasimus formicarius*) [28] and the weevil *Scyphophorus acupunctatus* [27]. 1, 8-Cineole has showed strong repellent activity against several insects, e.g. stored product insects (*S. granarius*, *S. zeamais*, *T. castaneum* and *Prostephanus truncates*) [26, 30], workers of red imported fire ants, *Solenopsis invicta* [29]. 1, 8-

cineole was effective against mosquitoes *Culex pipiens molestus* bites offering complete protection for 2 h [25]. No attract activity of 1, 8-cineole to insects was found so far. The above findings suggested that α-pinene and 1, 8-cineole may have potential to be developed as new natural repellents in the control of cockroaches.

Some essential oils derived from Chinese medicinal herbs possess contact and fumigant toxicity against insects (especially stored product insects and health important insect pests) and also have feeding deterrent activity against insects [31-33]. However, there is very little information on repellent activities of Chinese medicinal herbs against insects. Several authors have reported that *Ocimum* oils were strong repellents against adult mosquitoes, stored product insects, and ticks [34-37]. Moreover, an active compound, eugenol was isolated from *O. basilicum* and possess repellent activity against the tick (*Ixodes ricinus*) [38]. Piperitone isolated from CH_2Cl_2 extract of *Z. bungeanum* showed strong repellent activity against insects [39]. However, no report is available on repellent activity of essential oils of the other Chinese medicinal herbs against insects.

All the Chinese medicinal herbs screened in this study are already used in flavouring, pharmaceuticals, and confectionary and are considered non-toxic to humans. This study demonstrates that the essential oils of Chinese medicinal herbs, *C. rotundus*, *E. robusta*, and *O. basilicum* had strong repellent activity against the German cockroaches. The isolation and identification of the bioactive compounds in those essential oils are of utmost importance so that their potential application in controlling cockroaches can be fully exploited.

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References

[1] R.J. Brenner (1995). Understanding and Controlling the German Cockroach. Oxford University Press, New York. pp. 77-92.

[2] G. Nalyanya, C.B. Moore and C. Schal (2000). Integration of repellents, attractants, and insecticides in a "Push-pull" strategy for managing German cockroach (Dictyoptera: Blattellidae) populations, *J. Med. Entomol.* **37**, 427-434.

[3] C.L. Wang and G.W. Bennett (2006). Study of integrated pest management and baiting for German cockroach management in public housing, *J. Econ. Entomol.* **99**, 879-883.

[4] F.F. Dong, Y.D. Dong, R.W. Dong, C.Y. Li, F.G. Pan and X.Y. Wang (1998). Studies on new formulation of repellent, *Chin. J. Vector Biol. Contr.* **9**, 198-201(in Chinese with English abstract).

[5] M.B. Isman (2000). Plant essential oils for pest and disease management, *Crop Prot.* **19**, 603-608.

[6] K.S. Chang and Y.J. Ahn (2002). Fumigant activity of (*E*)-anethole identified in *Illicium verum* fruit against *Blattella germanica*, *Pest Manag. Sci.* **58,** 161-166.

[7] A.G. Appel, M.J. Gehret and M.J. Tanley (2001). Repellency and toxicity of mint oil to American and German cockroaches (Dictyoptera: Blattidae and Blattellidae), *J. Agric. Urban Entomol.* **18**, 149-156.

[8] C.J. Peterson, J. Zhu and J.R. Coats (2002). Identification of components of osage orange fruit (*Maclura pomifera*) and their repellency to German cockroaches, *J. Essent. Oil Res.* **14**, 233-236.

[9] C.J. Peterson, L.T. Nemetz, L.M. Jones and J.R. Coats (2002). Behavioral activity of catnip (Lamiaceae) essential oil components to the German cockroach (Blattodea: Blattellidae), *J. Econ. Entomol.* **95**, 377-380.

[10] G. Schultz, E. Simbro, J. Belden, J. Zhu and J. Coats (2004). Catnip, *Nepeta cataria* (Lamiales: Lamiaceae) - a closer look: seasonal occurrence of nepetalactone isomers and comparative repellency of three terpenoids to insects, *Environ. Entomol.* **33**, 1562-1569.

[11] Z.L. Liu and S.H. Ho (1999). Bioactivity of the essential oil extracted from *Evodia rutaecarpa* Hook f. et Thomas against the grain storage insects *Sitophilus zeamais* Motsch. and *Tribolium castaneum* (Herbst), *J. Stored Prod. Res.* **35**, 317-328.

[12] A.A. Ferrero, C.S. Chopa, J.O.W. Gonzalez and R.A. Alzogaray (2007). Repellence and toxicity of *Schinus molle* extracts on *Blattella germanica*, *Fitoterapia* **78**, 311-314.
[13] F.A. Talukder and P.E. Howse (1995). Evaluation of *Aphanamixis polystachya* as a source of repellents, antifeedants, toxicants and protectants in storage against *Tribolium castaneum* (Herbst), *J. Stored Prod. Res.* **31**, 55-61.
[14] R.P. Adams (2001). Identification of Essential Oil Components by Gas Chromatography/Quadrupole Mass Spectroscopy. Allured: Carol Stream, IL.
[15] D.E Wedge, J.A. Klun, N. Tabanca, B. Demirci, T, Ozek, K.H.C. Baser, Z. Liu, S. Zhang, C.L. Cantrell and J. Zhang (2009). Bioactivity-guided fractionation and GC/MS fingerprinting of *Angelica sinensis* and *Angelica archangelica* root components for antifungal and mosquito deterrent activity, *J. Agric. Food Chem.* **57**, 464-470.
[16] M.Y. Tang, L.F.Sun and H.W.Wang (2000). Study on the chemical constituents of essential oil from Chinese traditional drug EShu (Tubers of Curcuma), *Chem. Ind. Forest Prod.* **20**, 65-69 (in Chinese with English abstract).
[17] S. Kilani, J. Ledauphin, I. Bouhlel, M. Ben Sghaier, J. Boubaker, I. Skandrani, R. Mosrati, K. Ghedira, D. Barillier and L. Chekir-Ghedira (2008). Comparative study of *Cyperus rotundus* essential oil by a modified GC/MS analysis method. Evaluation of its antioxidant, cytotoxic, and apoptotic effects**,** *Chem. Biodiver.* **5**, 729-742.
[18] M.B. Gholivand, M. Rahimi-Nasrabadi and H. Chalabi (2009). Determination of essential oil components of star anise (*Illicium verum*) using simultaneous hydrodistillation-static headspace liquid-phase microextraction-gas chromatography mass spectrometry, *Anal. Lett.* **42**, 1382-1397.
[19] Z.Q. Du, H.L. Xia, H.X. Jiang, B.F. Zhang and F. Meng (2003). GC-MS analysis of volatile oil of *Lindera aggregate*, *Chin. Trad. Herbal Drugs* **34,** 308-310.
[20] J.W. Zhang, S.K. Li and W.J. Wu (2009). The main chemical composition and *in vitro* antifungal activity of the essential oils of *Ocimum basilicum* Linn. var. *pilosum* (Willd.) Benth, *Molecules* **14**, 273-278.
[21] Y.W. Gong, Y.F. Huang, L.G. Zhou, X.Y. Shi, Z.J. Guo, M.A. Wang and W.B. Jiang (2009). Chemical composition and antifungal activity of the fruit oil of *Zanthoxylum bungeanum* Maxim (Rutaceae) from China, *J. Essent. Oil Res.* **21**, 174-178.
[22] H.D. Sun, J.K. Ding, L.S. Ding and Y.F. Yi (1985). The chemical constituents of *Eucalyptus* oil, *Acta Botanica Yunnanica* **7**, 351-354 (in Chinese with English abstract).
[23] P. Sartorelli, A.D. Marquioreto, A. Amaral-Baroli, M.E.L. Lima and P.R.H. Moreno (2007). Chemical composition and antimicrobial activity of the essential oils from two species of *Eucalyptus*, *Phytotherapy Res*. **21**, 231-233.
[24] S.P. Ngoh, L.E.W. Choo, F. Pang, Y. Huang, M.R. Kini and S.H. Ho (1998). Insecticidal and repellent properties of nine volatile constituents of essential oils against the American cockroach, *Periplaneta americana* (L.), *Pestic. Sci.* **54**, 261-268.
[25] A.F. Traboulsi, S. El-Haj, M. Tueni, K. Taoubi, N.A. Nader and A. Mrad (2005). Repellency and toxicity of aromatic plant extracts against the mosquito *Culex pipiens molestus* (Diptera: Culicidae), *Pest Manag. Sci.* **61**, 597–604.
[26] C. Yoon, S.H. Kang, S.A. Jang, Y.J. Kim and G.H. Kim (2007). Repellent efficacy of caraway and grapefruit oils for *Sitophilus oryzae* (Coleoptera: Curculionidae), *J. Asia-Pacific Entomol.* **10**, 263-267.
[27] K. Paalsson, T.G.T. Jaenson, P. Baeckstrom and A.K. Borg-Karlson (2008). Tick repellent substances in the essential oil of *Tanacetum vulgare*, *J. Med. Entomol.* **45**, 88-93.
[28] L.M. Schroeder and A. Lindeloew (1989). Attraction of scolytids and associated beetles by different absolute amounts and proportions of α-pinene and ethanol., *J. Chem. Ecol.* **15**, 807-817.
[29] A. Altuzar, E.A. Malo, H. Gonzalez-Hernandez and J.C. Rojas (2007). Electrophysiological and behavioural responses of *Scyphophorus acupunctatus* (Col., Curculionidae) to *Agave tequilana* volatiles, *J. Appl. Entomol.* **131**, 121-127.
[30] D. Obeng-Ofori, C. Reichmuth, J. Bekele and A. Hassanali (1997). Biological activity of 1,8-cineole, a major component of essential oil of *Ocimum kenyense* (Ayobangira) against stored product beetles, *J. Appl. Entomol.* **121**, 237-243.
[31] J. Chen (2009). Repellency of an over-the-counter essential oil product in China against workers of red imported fire ants, *J. Agric. Food Chem.* **57**, 618-622.
[32] S.F. Chiu (1989). Recent advances in research on botanical insecticides in China, *ACS Symposium* **387**, 69-77.
[33] Z.L. Liu, S.H. Goh and S.H. Ho (2007). Screening of Chinese medicinal herbs for bioactivities against *Sitophilus zeamais* Motschulsky and *Tribolium castaneum* (Herbst), *J. Stored Prod. Res.* **43**, 290-296.

[34] D.P. Papachristos and D.C. Stamopoulos (2002). Repellent, toxic and reproduction inhibitory effects of essential oil vapours on *Acanthoscelides obtectus* (Say) (Coleoptera: Bruchidae), *J. Stored Prod. Res.* **38**, 117-128.

[35] M.J. Pascual-Villalobos and M.C. Ballesta-Acosta (2003). Chemical variation in an *Ocimum basilicum* germplasm collection and activity of the essential oils on *Callosobruchus maculatus*. *Biochem. Syst. Ecol.* **31**, 673-679.

[36] F. Erler, I. Ulug and B. Yalcinkaya (2006). Repellent activity of five essential oils against *Culex pipiens*, *Fitoterapia* **77**, 491-494.

[37] K. Murugan, P. Murugan and A. Noortheen (2007). Larvicidal and repellent potential of *Albizzia amara* Boivin and *Ocimum basilicum* Linn against dengue vector, *Aedes aegypti* (Insecta:Diptera:Culicidae), *Bioresour. Technol.* **98**, 198-201.

[38] S. D. Fabbro and F. Nazzi (2008). Repellent effect of sweet basil compounds on *Ixodes ricinus* ticks, *Exp. Appl. Acarol.* **45**, 219-228.

[39] W.S. Bowers, F. Ortego, X. You and P.H. Evans (1993). Insect repellents from the Chinese prickly ash *Zanthoxylum bungeanum*, *J. Nat. Prod.* **56**, 935-938.

[40] T.G.T. Jaenson, K. Paalsson and A.K. Borg-Karlson (2006). Evaluation of extracts and oils of mosquito (Diptera: Culicidae) repellent plants from Sweden and Guinea-Bissau, *J. Med. Entomol.* **43**, 113-119.