FULL PAPER

Analysis of the Essential Oil of *Elsholtzia ciliate* Aerial Parts and Its Insecticidal Activities against *Liposcelis bostrychophila*

by **Mei Ping Zhao**[a]), **Xin Chao Liu**[a]), **Daowan Lai**[b]), **Ligang Zhou**[b]), and **Zhi Long Liu***[a])

[a]) Department of Entomology, China Agricultural University, Haidian District, Beijing 100193, P. R. China (phone/fax: +86-10-62732800; e-mail: zhilongliu@cau.edu.cn)

[b]) Department of Plant Pathology, China Agricultural University, Haidian District, Beijing 100193, P. R. China

Water-distilled essential oil from *Elsholtzia ciliate* (Labiatae) aerial parts at flowering stage was analyzed by gas chromatography/mass spectrometry (GC/MS). Thirty-six compounds, accounting for 98.3% of the total oil content, were identified, and the main components of the essential oil were dehydroelsholtzia ketone (26.5%), (*R*)-carvone (16.6%), elsholtzia ketone (14.6%), and D-limonene (4.1%). The essential oil contained higher amounts of monoterpenoids (83.4%) than of sesquiternoids (8.3%). Bioactivity-directed chromatographic separation of the essential oil on repeated silica gel columns led to the isolation of three monoterpenoids. The essential oil possessed fumigant toxicity against the booklice (*Liposcelis bostrychophila*) with an LC_{50} value of 475.2 μg/l, while the isolated constituents, (*R*)-carvone, dehydroelsholtzia ketone, and elsholtzia ketone had LC_{50} values of 417.4, 658.2, and 547.3 μg/l, respectively. The essential oil also exhibited contact toxicity against *L. bostrychophila* with an LC_{50} value of 145.5 μg/cm^2. (*R*)-Carvone, dehydroelsholtzia ketone, and elsholtzia ketone exhibited acute toxicity against the booklice with LC_{50} values of 57.0, 151.5, and 194.1 μg/cm^2, respectively. The results indicated that the essential oil and the isolated constituents have potential for the development into natural insecticides/fumigants for the control of insects in stored grains.

Introduction. – Botanical pesticides have the advantage of providing novel modes of action against insects that can reduce the risk of cross-resistance as well as offering new leads for the design of target-specific molecules. During a screening program for new agrochemicals from Chinese medicinal herbs and local wild plants, the essential oil of *Elsholtzia ciliate* (Labiatae) aerial parts at flowering stage was found to possess insecticidal activity against booklice (*Liposcelis bostrychophila* Badonnel). Booklice have posed an alarming threat to stored grains, especially in the storage facilities, where controlled atmosphere and insecticide combined treatments are commonly practiced [1]. At present, infestations of stored product insects are typically controlled by fumigation or insecticidal treatment of commodities and surfaces, which has led to problems such as pest resurgence and increasing costs of application originating from the development of resistance to insecticides. In addition, these pesticides can have lethal effects on non-target organisms and can cause direct toxicity to applicators [2]. Strong resistance to a currently used fumigant, phosphine, has been detected in several psocid populations from China, Australia, and Indonesia [1]. It is urgent to look for new fumigants as alternatives for the currently used fumigant. In line with this, essential oils and their constituents may provide an alternative to currently used fumigants/pesticides to control stored-food insects [3]. Many essential oils and constituent compounds derived from the essential oils of Chinese medicinal herbs can exert fumigant and contact toxicity as well as repellency against grain storage insects [4 – 7].

E. ciliate is an herbaceous plant up to 30 – 50 cm high and grows wild in all the provinces of China except Qinghai province and Xinjiang Uygur Autonomous Region [8]. It is also distributed in Cambodia, India, Japan, Laos, Malaysia, Mongolia, Myanmar, Russia, Thailand, and Vietnam [8]. The aerial parts of this herb are used in Traditional Chinese Medicine for the treatment of blood clotting, gastralgia, dysphonia, jaundice, diarrheic, throat infections, and as an astringent, antipyretic, and antiviral medicine for the treatment of cough and fever [9]. There are several publications on the chemical composition of the essential oil of *E. ciliata* from natural habitat [10 – 15] as well as of samples cultivated in Europe and Vietnam [16]. The essential oil of *E. ciliate* exhibited strong antimicrobial activity against six bacterial strains and the yeast strain correlating with the diseases of infectious intestines and stomach [9]. However, a literature survey has shown that there is no report on chemical composition and insecticidal activity of the essential oil of *E. ciliate* aerial parts against the booklice. The present investigation was therefore undertaken to investigate the chemical constituents and insecticidal activities of the essential oil and its active constituents against the booklice.

Results and Discussion. – *Chemical Composition of the Essential Oil.* The yield of essential oil of *E. ciliate* aerial parts was 0.03% (yellow, *v*/*w* based on dry weight) while its density was determined to be 0.91 g/ml. A total of 36 components from the essential oil of *E. ciliate* aerial parts were identified, accounting for 98.3% of the total oil

DOI: 10.1002/hlca.201500232

(*Table 1*). The principal constituents of *E. ciliate* essential oil were dehydroelsholtzia ketone (26.5%), (*R*)-carvone (16.6%), elsholtzia ketone (14.9%), and D-limonene (4.1%). The essential oil contained higher amounts of monoterpenoids (83.4%) than of sesquiternoids (8.3%; *Table 1*). It had some differences to the essential oil of *E. ciliate* aerial parts measured in the previous reports [14 – 16]. For example, thymol (8.9%), carvacrol (5.6%), dehydroelsholtzia ketone (4.1%), and elsholtzia ketone (3.9%) were the main constituents in the essential oil of *E. ciliate* aerial parts harvested from Qingyang City (106.50° N and 35.50° E, Gansu Province, P. R. China) [15] while the essential oil of *E. ciliate* aerial parts collected from Jingshan (112.50° N and 30.50° E, Hubei Province, P. R. China) mainly contained carvacrol (23.8%), thymol (18.3%), *p*-cymene (6.5%), and *o*-cymene (6.0%) [14]. Moreover, *Tian* [9] determined that the essential oil of *E. ciliate* leaves collected from of the Qinba Mountain of Shaanxi Province in China (107.01° N and 33.04° E) mainly contained linalool (12.1%), caryophyllene (11.0%), eugenol (9.7%), caryophyllene oxide (9.6%), 1-octen-3-ol (8.9%), verbenone (6.9%), and spathulenol (7.2%), and the oil derived from the flowers had linalool (11.5%), *β*-ionone (11.1%), eugenol (10.6%), caryophyllene (9.6%), and isocaryophyllene (4.2%). Moreover, the essential oil of *E. ciliate* collected from La Chung Valley in the Himalayan region of India mainly contained rosefuran (84.8%) and 1,8-cineole (4.6%) [10], and dehydroelsholtzia ketone (65.2%), elsholtzia ketone (7.6%), *p*-cymene (4.3%), and camphor (3.6%) were the major constituents in the second essential oil sample from Chung Thang region of India [10]. The major constituents of the essential oils of *E. cilliata* leaves

Table 1. *Chemical Constituents of the Essential Oil Derived from* Elsholtzia ciliate

Peak Number	Compound name and class	*RI*[a])	Percent Composition [%]
	Monoterpenoids		83.4
1	*α*-Pinene	931	1.4
2	*β*-Pinene	981	0.5
3	*β*-Myrcene	991	3.6
4	4-Carene	1002	1.5
5	*p*-Cymene	1025	0.3
6	D-Limonene	1029	4.1
7	1,8-Cineole	1032	3.5
8	(*Z*)-*β*-Ocimene	1038	1.0
9	*γ*-Terpinene	1059	1.0
10	Linalool	1100	1.3
11	*α*-Fenchol	1104	0.6
12	Citronellal	1152	0.5
13	Lavandulol	1162	0.7
14	Elsholtzia ketone	1199	14.6
15	*p*-Cymen-8-ol	1182	0.9
16	*α*-Terpineol	1191	0.6
17	1,6-Dihydrocarveol	1195	2.3
18	*cis*-Carveol	1226	0.8
19	Geranial	1240	1.1
20	(*R*)-Carvone	1242	16.6
21	Dehydroelsholtzia ketone	1277	26.5
	Sesquiterpenoids		8.3
22	*β*-Elemene	1391	0.2
23	Caryophyllene	1420	0.5
24	Aromadendrene	1431	0.7
25	*α*-Humulene	1454	1.3
26	Germacrene D	1480	2.1
27	*α*-Farnesene	1503	0.5
28	*β*-Bisabolene	1510	0.1
29	*δ*-Cadinene	1520	0.5
30	Nerolidol	1567	0.6
31	Spathulenol	1578	0.6
32	Caryophyllene oxide	1583	1.2
	Others		6.6
33	Octen-3-ol	980	2.3
34	3-Octanone	982	1.5
35	Acetophenone	1066	2.4
36	*cis*-Jasmone	1396	0.4
	Total identified		98.3

[a]) Retention index (*RI*) as determined on a *HP-5MS* column using the homologous series of *n*-hydrocarbons.

from three environmentally similar sites at Mr. *Muhak* in Korea were dihydrotagetone (62.7%), β-caryophellene (5.0%), and germacrene D (4.0%) [12]. The amount of dihydrotagetone fluctuated significantly during the season, ranging from 40.6 to 81.6% [12]. The above findings suggested that it is necessary to standardize the essential oil of *E. cilliata* before the essential oil being used for commercial production, because great variations were observed in the chemical composition of the essential oils derived from different regions and different plant parts.

Isolated Compounds. Based on bioassay-guided fractionation (contact toxicity), three bioactive compounds were isolated and identified as (*R*)-carvone, elsholtzia ketone, and dehydroelsholtzia ketone by their spectroscopic data and comparison with literature data. Their chemical structures are given in the *Figure*.

Insecticidal Activity. The essential oil exhibited contact toxicity against *L. bostrychophila* with an LC_{50} value of 145.5 μg/cm^2 (*Table 2*). Based on the LC_{50} values, the essential oil exhibited eight times less acute toxicity than the positive control, pyrethrum extract (LC_{50} = 19.0 μg/cm^2) against the booklice. However, when compared with the other essential oils in previous studies using the same bioassay, the essential oil of *E. ciliate* aerial parts exhibited stronger or the same level of acute toxicity against the booklice, *e.g.*, essential oils of *Acorus calamus* [17], *Artemisia rupestris* and *A. frigida* [18][19], *Curcuma wenyujin* [20], *Foeniculum vulgare* [1], *Valeriana jatamansi* [21], but less toxic than the essential oils of *Ageratum houstonianum* [22], *Kaempferia galangal* [23], and *Illicium henryi* [24]. Based on LC_{50} values, among the three isolated constituents, (*R*)-carvone (LC_{50} = 57.0 μg/cm^2) exhibited the strongest acute toxicity against the booklice. (*R*)-Carvone displayed 2.5 times stronger contact toxicity (no overlap in LC_{50} values) than crude essential oil against the booklice (*L. bostrychophila*) and dehydroelsholtzia ketone (LC_{50} = 151.5 μg/cm^2) showed the same level of toxicity as the essential oil. Thus, it seems that the contact toxicity of the essential oil may be mainly attributed to (*R*)-carvone and dehydroelsholtzia ketone.

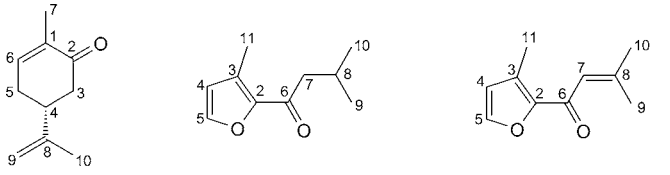

Figure. *Structure of the three biologically active constituents isolated from the essential oil of* Elsholtzia ciliate *aerial parts*

The essential oil also possessed fumigant toxicity against the booklice (*L. bostrychophila*) with an LC_{50} value of 475.2 μg/l (*Table 2*). Based on LC_{50} values, only one isolated constituent, (*R*)-carvone (LC_{50} = 417.4 μg/l) exhibited stronger fumigant toxicity (no overlap in LC_{50} values) than the crude essential oil against the booklice. Compared with dichlorvos (LC_{50} = 1.4 μg/l), the essential oil of *E. ciliate* and (*R*)-carvone showed only 352 times and 309 times less toxicity (based on LC_{50} values) against *L. bostrychophila*, respectively. When compared with the other essential oils in the previous studies using the same bioassay, the essential oil of *E. ciliate* exhibited stronger fumigant toxicity against the booklice, *e.g.* essential oils of *A. rupestris* (LC_{50} = 6.7 mg/l air) [18], *A. frigida* (LC_{50} = 1.3 mg/l air) [19], *C. wenyujin* (LC_{50} = 2.8 mg/l) [20], *A. calamus* (LC_{50} = 392.1 μg/l) [17], *K. galangal* (LC_{50} = 1.5 mg/l air) [23], and *V. jatamansi* (LC_{50} = 6.0 mg/l) [21], but was less toxic than the essential oils of *Allium chinense* (LC_{50} = 186.5 μg/l) [25] and *F. vulgare* fruits (LC_{50} = 34.1 μg/l) [1]. However, considering that the commercial fumigants are synthetic insecticides and the most effective fumigants (*e.g.* phosphine and MeBr) are also highly toxic to humans and other non-target organisms, fumigant activity of the essential oil of *E. ciliate* and the three major constituents, especially (*R*)-carvone, is quite promising.

In the previous studies, (*R*)-carvone has been demonstrated to possess contact and fumigant toxicity as well as repellency against many species of insects and mites, *e.g.* Japanese termite (*Reticulitermes speratus*) [26], sciarid fly (*Lycoriella ingenua*) [27], mosquitoes (*Aedes aegypti* and *Culex pipiens*) [28], German cockroaches (*Blattella germanica*) [29], Southern corn rootworm (*Diabrotica undecimpunctata*) and house flies (*Musca domestica*) [30], and several stored product insects, *e.g.* the cigarette beetle

Table 2. *Contact Toxicity* ([μg/cm^2]) *and Fumigant Toxicity* ([μg/l]) *of the Essential Oil of* Elsholtzia ciliate *and Its Isolated Compounds against* Liposcelis bostrychophila

Treatments		Toxicity (LC_{50})	95% Fiducial limits	Slope ± SE	χ^2
Contact	*E. ciliata*	145.5	135.3 – 157.0	8.9 ± 0.8	13.2
	(*R*)-Carvone	57.0	53.1 – 60.7	5.7 ± 0.5	10.6
	Dehydroelsholtzia ketone	151.5	142.5 – 161.8	7.0 ± 0.7	10.1
	Elsholtzia ketone	194.1	181.3 – 205.8	6.4 ± 0.6	11.8
	Pyrethrum extract[a])	19.0	17.3 – 20.6	7.6 ± 0.7	9.4
Fumigant	*E. ciliata*	475.2	449.6 – 495.7	4.9 ± 0.4	15.4
	(*R*)-Carvone	417.4	388.9 – 438.9	4.8 ± 0.5	9.8
	Dehydroelsholtzia ketone	658.2	622.5 – 684.6	8.4 ± 0.8	14.3
	Elsholtzia ketone	547.3	509.6 – 581.5	5.9 ± 0.5	14.8
	Dichlorvos[a])	1.35	1.25 – 1.47	6.9 ± 0.6	10.4

[a]) Positive control.

(*Lasioderma serricorne*) [31], red flour beetle (*T. castaneum*) and maize weevil (*S. zeamais*) [32], rice weevil (*S. oryzae*) [33], lesser grain borer (*R. dominica*), and flat grain beetle (*Cryptolestes pusillus*) [34], as well as the two-spotted spider mite (*Tetranychus urticae*) [35] and mould mites (*Tyrophagus putrescentiae*) [36]. There is no report on insecticidal activity of the other two constituents, dehydroelsholtzia ketone and elsholtzia ketone so far. The above findings suggest that the essential oil of *E. ciliate* aerial parts and the three isolated constituents, especially (*R*)-carvone show potential to be developed as possible natural insecticides/fumigants for the control of grain storage insects.

E. ciliate aerial parts are well known for its common use in the traditional Chinese medicine [8][9]. It seems that this medicinal herb is quite safe to human consumption because it has been used as a medicinal herb and a spice for hundreds of years. However, there is no experimental data on toxicity of the essential oil of *E. ciliate* to human is available, to the best of our knowledge. Thus, to develop a practical application for the essential oil and its constituents as novel insecticides, further research into the safety of the essential oil/compounds to humans is needed. Additional studies on the development of formulations are also necessary to improve the efficacy and stability and to reduce cost. Moreover, field evaluation and further investigations on the effects of the essential oil and its constituent compounds on non-target organisms are necessary.

Conclusions. – The present work indicated that the essential oil of *E. ciliate* aerial parts demonstrates contact and fumigant toxicity against the adults of *L. bostrychophila*. (*R*)-Carvone exhibited strong contact and fumigant toxicity against *L. bostrychophila*. Our results suggested that the essential oil of *E. ciliate* aerial parts and the three isolated constituents, especially (*R*)-carvone, may be recommended effectively in grain storage insects control program but needs to be further evaluated for safety in humans and to enhance their activity.

Experimental Part

General. ^{1}H- and ^{13}C-NMR spectra: *Bruker Avance DRX 500* instrument; in $CDCl_3$; δ in ppm rel. to Me_4Si as internal standard, *J* in Hz.

GC/MS Analysis of Essential Oil. The essential oil of *E. ciliate* aerial parts was subjected to GC/MS analysis on an *Agilent* (Santa Clara, CA, USA) system consisting of a model *6890N* gas chromatograph, a model *5973N* mass selective detector (EI-MS; electron energy, 70 eV), and an *Agilent* ChemStation data system. The GC column was an *HP-5ms* (Santa Clara, CA, USA) 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 temp. was held at 60° for 1 min and ramped at 10°/min to 180°, held for 1 min, and then increased at 20°/min to 280° and held for 15 min. The injector temp. was maintained at 270°. The sample (1 μl, 1:100 (*v*/*v*) in acetone) was injected, with a split ratio of 1:10. The carrier gas was He at a flow rate of 1.0 ml/min. Spectra were scanned from *m*/*z* 20 to 550 at 2 scans/s. Most constituents were identified by GC by comparison of their retention indices with those found in the literature 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 08 and Wiley 275 libraries or with mass spectra in the literature [37]. Component relative percentages were calculated based on the normalization method without using correction factors.

Plant Material and Essential-Oil Extraction. Fresh aerial parts of *E. ciliate* (10 kg) at flowering stage were harvested in August 2013 from Xiaolongmeng National Forest Park (38.24° N and 115.20° E, Mentougou District, Beijing, P. R. China). The plant was identified by Dr. *Q. R. Liu* (College of Life Sciences, Beijing Normal University, Beijing, P. R. China), and a voucher specimen (No. ENTCAU-Labiatae-Xiangru-109) was deposited with the Museum of Department of Entomology, China Agricultural University. The sample was cut into small pieces and subjected to hydrodistillation using a modified *Clevenger-type* apparatus for 6 h. Anh. Na_2SO_4 was used to remove H_2O after extraction. The essential oil was stored in airtight containers in a refrigerator at 4° for subsequent experiments.

Isolation of Active Compounds. The crude essential oil (25 ml) was chromatographed on a SiO_2 column (90 mm i.d., 600 mm length; *Qingdao Marine Chemical Plant*, Shandong Province, P. R. China) by gradient elution with hexane first, then with hexane/AcOEt, and finally with AcOEt to obtain 15 fractions. Based on contact toxicity, *Fr. 3*, *Fr. 6*, and *Fr. 10* were chosen for further fractionation. They were separated by repeated CC (SiO_2) and prep. TLC to afford three pure compounds determined as (*R*)-carvone (67 mg), elsholtzia ketone (51 mg), and dehydroelsholtzia ketone (47 mg). The structures of the isolated compounds were elucidated based on their NMR spectra.

*(*R*)-Carvone* (*=(5*R*)-2-Methyl-5-(prop-1-en-2-yl)cyclohex-2-en-1-one*). Colorless oil. ^{1}H-NMR (500 MHz, $CDCl_3$): 1.77 (*s*, Me(7)); 1.80 (*s*, Me(10)); 2.27 – 2.73 (*m*, CH_2(3), H–C(4), CH_2(5)); 4.77 (*s*, H_a–C(9)); 4.82 (*s*, H_b–C(9)); 6.77 – 6.78 (*m*, H–C(6)). ^{13}C-NMR (125 MHz, $CDCl_3$): 150.1 (C(8)); 133.7 (C(1)); 120.8 (C(2)); 108.5 (C(10)); 41.2 (C(4)); 30.9 (C(6)); 30.7 (C(3)); 28.1 (C(5)); 23.5 (C(7)); 20.8 (C(9)). The spectral data were in agreement with those reported in [32].

Elsholtzia Ketone (*=3-Methyl-1-(3-methylfuran-2-yl)butan-1-one*). Colorless oil. ^{1}H-NMR (500 MHz, $CDCl_3$): 0.98 (*d*, *J* = 7, Me(9), Me(10)); 2.30 – 2.32 (*m*, H–C(8)); 2.48 (*s*, Me(11)); 2.69 – 2.71 (*m*, CH_2(7)); 6.35 (*d*, *J* = 2.0, H–C(4)); 7.35 (*d*, *J* = 2.0, H–C(5)). ^{13}C-NMR ($CDCl_3$, 125 MHz): 11.2 (C(11)); 20.5 (C(9)); 27.5 (C(10)); 115.5 (C(4)); 120.3 (C(7)); 129.3 (C(8)); 143.0 (C(5)); 148.5 (C(3)); 156.0 (C(2)); 181.2 (C(6)). The spectral data were in agreement with those reported in [10][38].

Dehydroelsholtzia Ketone (*= Naginata Ketone = 3-Methyl-1-(3-methylfuran-2-yl)but-2-en-1-one*). Colorless oil. ^{1}H-NMR (500 MHz, $CDCl_3$): 1.96 (*s*, Me(9)); 2.21 (*s*, Me(10)); 2.41 (*s*, Me(11)); 6.31 (*d*, *J* = 2.0, H–C(4)); 6.71 – 6.73 (*m*, H–C(7)); 7.38 (*d*, *J* = 2.0, H–C(5)). ^{13}C-NMR ($CDCl_3$, 125 MHz): 11.0 (C(11)); 20.5 (C(9)); 27.5 (C(10)); 28.2 (C(8)); 47.9 (C(7)); 114.6 (C(4)); 143.0 (C(5)); 147.5 (C(3)); 157.0 (C(2)); 191.0 (C(6)). These values are in agreement with those of dehydroelsholtzia ketone reported in the literature [10][38].

Insects. The booklice (*L. bostrychophila*) were obtained from laboratory cultures in the dark in incubators at 28 – 30° and 70 – 80% relative humidity and was reared on a 1:1:1 mixture, by mass, of milk powder, active yeast, and flour. All the containers housing insects and the *Petri* dishes used in experiments were made escape proof with a coating of polytetrafluoroethylene (*Fluon*®, *Blades Biological*, Edenbridge, UK). Laboratory bioassays were done within one week after adult collections.

Contact Toxicity. Range-finding studies were run to determine the appropriate testing concentrations of the essential oil/compounds. The filter paper with 3.5 cm in diameter (*Whatman*) was treated with 150 μl of the soln. (2.0, 2.4, 2.9, 3.5, 4.2, and 5.0% in acetone). The treated

filter paper after treated with solid glue (*Glue Stick*, *Jong Ie Nara Co., Ltd.*, Hong Kong) was placed in a *Petri* dish (3.5 cm in diameter) and ten booklice were put on the filter paper. The plastic cover with holes was put and all the *Petri* dishes were kept in incubators at 27 – 29°, 70 – 80% relative humidity for 24 h and mortality of insects was observed. Acetone was used as controls and pyrethrum extract was used as a positive control. Pyrethrum extract (25% pyrethrin I and pyrethrin II) was purchased from *Fluka Chemie* (*Buchs*, Switzerland).

Fumigant Toxicity. A filter paper strip (3.5 cm × 1.5 cm) treated with 10 µl of an appropriate concentration (3.1, 3.3, 3.5, 3.6, 3.8, and 4.0% in acetone) of the essential oil/compounds. The impregnated filter paper was then placed in the bottom cover of a glass bottle of 250 ml. The insects, ten adults with undefined sex in a small glass bottle (8 ml), were exposed for 24 h at 27 – 29° and 70 – 80% relative humidity and each concentration with five replicates. Mortality of insects was observed. Dichlorvos (99.9%) was purchased from *Aladdin-reagent Company* (Shanghai, P. R. China) and used as a positive control.

Data Analysis. The observed mortality data were corrected for control mortality using *Abbott*'s formula [39]. The results from all replicates in fumigant and contact toxicity were subjected to *Probit* analysis using PriProbit Program V1. 6.3 to determine LC_{50} values and their 95% confidence intervals [40]. Samples for which the 95% fiducial limits did not overlap were considered to be significantly different.

This work was funded by the *Hi-Tech Research and Development of China 2011AA10A202*. We thank Dr. *Q. R. Liu* from College of Life Sciences, Beijing Normal University, Beijing, P. R. China for the identification of the investigated plant.

REFERENCES

[1] N. N. Zhao, L. Zhou, Z. L. Liu, S. S. Du, Z. W. Deng, *Food Control* **2012**, *26*, 486.
[2] T. W. Phillips, J. E. Throne, *Annu. Rev. Entomol.* **2010**, *55*, 375.
[3] S. Rajendran, V. Srianjini, *J. Stored Prod. Res.* **2008**, *44*, 126.
[4] S. S. Chu, G. H. Jiang, Z. L. Liu, *Pest Manag. Sci.* **2011**, *67*, 1253.
[5] Z. L. Liu, S. S. Du, *E-J. Chem.* **2011**, *8*, 1937.
[6] J. S. Zhang, N. N. Zhao, Q. Z. Liu, Z. L. Liu, S. S. Du, L. Zhou, Z. W. Deng, *J. Agric. Food Chem.* **2011**, *59*, 9910.
[7] Y. Liang, J. L. Li, S. Xu, N. N. Zhao, L. Zhou, J. Cheng, Z. L. Liu, *J. Econ. Entomol.* **2013**, *106*, 513.
[8] 'Flora Reipublicae Popularis Sinicae', Editorial Committee of Flora of China of Chinese Academy of Sciences, Science Press, Beijing, China, 1977, Vol. 66, p. 346.
[9] G. H. Tian, *Chin. Herb. Med.* **2013**, *5*, 104.
[10] R. K. Thappa, S. G. Agarwal, B. K. Kapahi, T. N. Srivastava, *J. Essent. Oil Res.* **1999**, *11*, 97.
[11] E. A. Korolyuk, W. Koenig, A. V. Tkachev, *Khimiya Rastitel'nogo Syr'ya* **2002**, *1*, 31.
[12] J.-H. Kim, D.-H. Jung, *J. Plant Biol.* **2003**, *46*, 287.
[13] H. C. Li, X. Tian, *Chin. J. Spectroscopy Lab.* **2006**, *23*, 906.
[14] G. Liu, H. Wang, B. H. Zhou, J. C. Song, *Chin. J. Exp. Tradit. Med. Formulae* **2006**, *12*, 18.
[15] X. D. Zheng, H. B. Hu, *Chin. J. Spectroscopy Lab.* **2005**, *22*, 179.
[16] N. X. Dung, L. H. Han, L. H. Hai, P. A. Leclercq, *J. Essent. Oil Res.* **1996**, *8*, 107.
[17] X. C. Liu, L. Zhou, Z. L. Liu, S. S. Du, *Molecules* **2013**, *18*, 5684.
[18] X. C. Liu, X. P. Li, H. Q. Li, Z. W. Deng, L. Zhou, L. Z. Liu, S. S. Du, *Molecules* **2013**, *18*, 10733.
[19] X. C. Liu, Y. L. Li, T. J. Wang, Q. Wang, Z. L. Liu, *Trop. J. Pharm. Res.* **2014**, *13*, 587.
[20] Z. L. Liu, N. N. Zhao, C. M. Liu, L. Zhou, G. H. Jiang, *Molecules* **2012**, *17*, 12049.
[21] X. C. Liu, L. Zhou, Z. L. Liu, *J. Chem.* **2013**, doi:10. 1155/2013/853912.
[22] X. N. Lu, X. C. Liu, Q. Z. Liu, Z. L. Liu, *J. Chem.* **2014**, doi:10. 1155/2014/645687.
[23] X. C. Liu, Y. Liang, W. P. Shi, Q. Z. Liu, L. Zhou, Z. L. Liu, *J. Econ. Entomol.* **2014**, *107*, 1706.
[24] X. C. Liu, Z. L. Liu, *J. Food Prot.* **2015**, *78*, 772.
[25] X. C. Liu, X. N. Lu, Q. Z. Liu, Z. L. Liu, *J. Asia-Pacific Entomol.* **2014**, *17*, 853.
[26] S.-M. Seo, J. Kim, S.-G. Lee, C.-H. Shin, S.-C. Shin, I.-K. Park, *J. Agric. Food Chem.* **2009**, *57*, 6596.
[27] I.-K. Park, J. N. Kim, Y.-S. Lee, S.-G. Lee, Y.-J. Ahn, S.-C. Shin, *J. Econ. Entomol.* **2008**, *101*, 139.
[28] H. E.-D. M. Zahran, S. A. M. Abdelgaleil, *J. Asia-Pacific Entomol.* **2011**, *14*, 46.
[29] Y.-S. Jang, Y.-C. Yang, D.-S. Choi, Y.-J. Ahn, *J. Agric. Food Chem.* **2005**, *53*, 7892.
[30] S. Lee, C. J. Peterson, J. R. Coats, *J. Stored Prod. Res.* **2003**, *39*, 77.
[31] C.-X. You, Y. Wang, W.-J. Zhang, K. Yang, Y. Wu, Z.-F. Geng, H.-P. Chen, H.-Y. Jiang, S.-S. Du, Z.-W. Deng, Z.-L. Liu, *Ind. Crop. Prod.* **2014**, *61*, 331.
[32] R. Fang, C. H. Jiang, X. Y. Wang, H. M. Zhang, Z. L. Liu, L. Zhou, S. S. Du, Z. W. Deng, *Molecules* **2010**, *15*, 9391.
[33] S.-W. Kim, J. Kang, I.-K. Park, *J. Asia-Pacific Entomol.* **2013**, *16*, 443.
[34] A. K. Tripathi, V. Prajapati, S. Kumar, *J. Econ. Entomol.* **2003**, *96*, 1594.
[35] M. E. I. Badawy, S. A. A. El-Arami, S. A. M. Abdelgaleil, *Exp. Appl. Acarol.* **2010**, *52*, 261.
[36] C.-H. Lee, B.-K. Sung, H.-S. Lee, *J. Stored Prod. Res.* **2006**, *42*, 8.
[37] R. P. Adams, 'Identification of Essential Oil Components by Gas chromatography/Mass Spectrometry', Allured Publishing Corporation, Carol Stream, IL, 2007.
[38] A. D. Dembitskii, G. I. Kalinkina, E. S. Berhliev, *Chem. Nat. Compds.* **1993**, *29*, 733.
[39] D. J. Finney, 'Probit Analysis', Cambridge University, London, UK. 1971
[40] M. Sakuma, *Appl. Entomol. Zool.* **1998**, *33*, 339.

Received July 6, 2015
Accepted August 14, 2015

