Industrial Crops & Products 110 (2017) 65–74

Contents lists available at ScienceDirect

Industrial Crops & Products

journal homepage: www.elsevier.com/locate/indcrop

Residual contact toxicity and repellence of *Cupressus lusitanica* Miller and *Eucalyptus saligna* Smith essential oils against major stored product insect pests

Philip K. Bett[a,*], Arop L. Deng[b], Joshua O. Ogendo[c], Samuel T. Kariuki[a], Maud Kamatenesi-Mugisha[d], Joel M. Mihale[e], Baldwyn Torto[f]

[a] Department of Biological Sciences, Egerton University, P.O. Box 536-20115, Egerton, Kenya
[b] College of Natural Resources and Environmental Studies, University of Juba, P.O. Box 82, Juba, South Sudan
[c] Department of Crops, Horticulture and Soils, Egerton University, P.O. Box 536-20115, Egerton, Kenya
[d] Bishop Stuart University, P.O. Box 09, Mbarara Municipality, Mbarara District, Uganda
[e] Department of Physical Sciences, Open University of Tanzania, P.O. Box 31608, Dares Salaam, Tanzania
[f] Behavioural and Chemical Ecology Department, icipe, P.O. Box 30772-00100, Nairobi, Kenya

ARTICLE INFO

Keywords:
Cupressus lusitanica
Essential oil
Eucalyptus saligna
Insect pest
Mortality
Repellence

ABSTRACT

In an effort to find eco-friendly alternatives to synthetic pesticides in grain storage, residual contact toxicity and repellence of *Cupressus lusitanica* and *Eucalyptus saligna* leaf essential oils were evaluated against adult *Tribolium castaneum, Acanthoscelides obtectus* and *Sitophilus zeamais*. In bioassays, oil was applied at 0.00, 0.05, 0.10, 0.15 and 0.20% v/w to wheat and bean grains and stored for 30–120 days after which test insects were introduced into sub-samples of treated grains. Both oils at 0.20% v/w and 120 days grain storage duration caused a mortality of 5.0–65.0% in test insects whereas in the repellence bioassay, at same doses and grain storage duration produced percent repellence values of 34–52.4% of test insects. Considering other pesticidal properties of *C. lusitanica* and *E. saligna* oils, current results point oils as potential residual contact toxicants and repellents for possible integration into insect pest management practices.

1. Introduction

Insect pests cause 5–10 and 20–30% damage to stored grains in the temperate and tropical countries, respectively (Philips and Throne, 2010). Post-harvest losses can include not only loss of the crop itself, but also lack of return on the resources needed to produce the crop, and a decrease in the livelihood of individuals involved in the production process (Bett, 2015). In addition, stored product pests also contaminate milled grains including presence of insect fragments in flour (Campolo et al., 2012). Several species of insects attributed to these losses and identified as the major insect pests of stored cereal and legume grains globally include, maize weevil, *Sitophilus zeamais* Motch. (Coleoptera: Curculionidae), Angoumois grain moth, *Sitotroga cerealella* Olivier (Lepidoptera: Gelechiidae), bostrichid beetles, *Prostephanus truncatus* Horn and *Rhyzopertha dominica* F. (Coleoptera: Bostrichidae), bean bruchid, *Acanthoscelides obtectus* Say (Coleoptera: Bruchidae), cowpea beetles, *Callosobruchus chinensis* F. (Coleoptera: Chrysomelidae), the rust-red flour beetle *Tribolium castaneum* Herbst (Coleoptera: Tenebrionidae) (Rajendran and Sriranjini, 2008; Deng et al., 2009; Nukenine, 2010; Ogendo et al., 2012; Bett, 2015).

The bean bruchid, *A. obtectus*, together with cowpea beetles (*C. chinensis* and *C. maculatus*) are destructors of stored legume grains. The bean bruchid is a major pest of beans in temperate to subtropical regions worldwide. The potential damage to stored grains by this pest is great owing to its ability to infest grains both pre- and post-harvest, and several larvae can develop in one seed (Ogendo et al., 2012; Bett et al., 2016). *Sitophilus zeamais* larvae damage maize crops by developing within an individual grain, eating it away from the inside out until it matures, and then reproducing, releasing more crop-damaging larvae. The maize weevil is a danger to both growing standing crops and stored maize (Ogendo et al., 2012). On the other hand, *T. castaneum* is a cosmopolitan stored product insect pest that can be found in warehouses, pet food stores, and grain processing facilities such as rice and flour mills. It is considered a secondary insect pest species and is frequently one of the least susceptible stored product beetle pest species to insecticides (Bett et al., 2016).

Stored product insect pest control is based mainly on the use of highly effective synthetic fumigants and contact toxicants. However,

* Corresponding author.
E-mail address: pkkbett@yahoo.co.uk (P.K. Bett).

http://dx.doi.org/10.1016/j.indcrop.2017.09.046
Received 14 April 2017; Received in revised form 18 September 2017; Accepted 20 September 2017
Available online 28 September 2017

increased health, environmental and socio-economic concerns and the consequent demand for pesticide-free food, have necessitated the development of non-chemical strategies for stored pest management (Ayvaz et al., 2010). Among the natural products, plant essential oils and their constituents have the potential to control storage insect pests and preserve food commodities.

Many studies have demonstrated that essential oils extracted from different plants showed a broad spectrum of activity against insect pests of stored grains, including ovicidal, larvicidal, adulticidal, antifeedant, repellent, and growth regulatory activities (Abay et al., 2012; Bett et al., 2013, 2016). A number of essential oils and constituents have been classified as contact toxicants (Rosman et al., 2007; Ogendo et al., 2011; Abay et al., 2012; Bett et al., 2013) and repellents (Nerio et al., 2010; Ogendo et al., 2012; Bett et al., 2016). Studies on the biological activity of *Eucalyptus* species extracts and constituents have good promise as fumigants and contact toxicants, repellents (Nivea et al., 2013) against major pests of stored products. Additionally, Tapondjou et al. (2005) found essential oils extracted from *E. saligna* leaves to have toxic effects on *S. zeamais* (LD_{50} = 0.36 ml cm^{-2}) and *Tribolium confusum* Jacquelin du Val (Coleoptera: Tenebrionidae) (0.48 ml cm^{-2}). Probit analysis showed that *T. confusum* was comparatively more susceptible (LD_{50} = 0.96 ml cm^{-2}) to the toxic effect of cymol (*p*-cymene), a major constituent of *E. saligna* oil than *S. zeamais* (LD_{50} = 1.35 ml cm^{-2}).

The repellent ability of essential oils and constituents from these plant species has already been reported (Nerio et al., 2010; Mossi et al., 2011; Liu et al., 2011; Regnault-Roger et al., 2012). Demonstrated that essential oils and constituents obtained from *Lantana camara* L. (Verbenaceae), *Ocimum americanum* L. (Lamiaceae), and *Tephrosia vogelii* Hook (Fabaceae) were effective repellents against *Sitophilus oryzae* L. (Coleoptera: Curculionidae) *T. castaneum*, *C. chinensis* and *R. dominica.* In other related studies, Liang et al. (2013) showed that the essential oils of *Curcuma longa* L. (Zingiberaceae), *Epimedium pubescens* L. (Berberidaceae), *Lindera aggregate* Sims (Lauraceae), *Nardostachys chinensis* Don (Caprifoliaceae), *Schizonepeta tenuifolia* Siebold & Zucc, *Zanthoxylum schinifolium*, and *Z. officinale* Roscoe (Zingiberaceae) exhibited strong repellent action against *T. castaneum*. The repellent action of the different essential oils against *T. castaneum* were reported to decrease in the order of *Cymbopogon martini*, *C. flexuosus* Roxb (poaceae) and *Lippia origanoides* L. (Verbenaceae) (Caballero-Gallardo et al., 2012).

The highly repellent effects of the main constituents of plant essential oils such as 1, 8-cineole, terpineol and α-pinene have also been demonstrated by other researchers (Tapondjou et al., 2005; Toloza et al., 2006; Nivea et al., 2013). Toloza et al. (2006) demonstrated strong repellent activity of essential oil from *Eucalyptus cinerea*, *E. viminalis Eucalyptus cinerea* Muel (Myrtaceae) and *E. saligna*, against permethrin-resistant human head lice. The repellent effect was associated with α-pinene 1, 8-cineole, citronellol, eugenol and camphor. Similarly, *Eucalyptus citriodora* Hook (Myrtaceae) and *Cymbopogon winterianus* L. (poaceae) oils are repellent to adult *C. maculatus* and repellence was associated with compounds like citronellal, 1, 8-cineole, limonene, geranial, neral, (*E*)-anethole, and α-pinene (Nivea et al., 2013). Synthetic chemicals are still more frequently used as repellents than essential oils. However, these natural products have the potential to provide efficient and safer repellents to humans and the environment. Furthermore, natural repellents may be included in stored product pest management where chemical residues and insects in produce may not be tolerated by consumers.

Essential oil-based insecticides are, therefore, very important of the control stored product insects pests owing to their activity against a variety of insects, fast penetration and reduced toxic residues in the treated products (Mbata and Payton, 2013). However, setbacks of using essential oil include volatility, solubility and oxidation, which play an important role in the essential oil activity, application and persistence. The aim, therefore, of the current study was to evaluate residual contact toxicity and repellence of essential oils obtained from leaves of *C. lusitanica* and *E. saligna* against *T. castaneum, A. obtectus* and *S. zeamais*.

2. Materials and methods

2.1. Experimental conditions and test insects

The rearing of test insects and bioassays were carried out at the Integrated Biotechnology Laboratory, Egerton University, Kenya at controlled conditions of temperature (28 ± 2 °C) and relative humidity (65 ± 5%) in continuous darkness. *Tribolium castaneum* was reared in wheat flour and 5% brewers' yeast (wt:wt). *Sitophilus zeamais* and *A. obtectus* were reared on whole wheat and bean grains, respectively. One- five day old (*T. castaneum* and *S. zeamais*) and 1–2 day old (*A. obtectus*) emerging adult insects were used for bioassays. The grains used for the bioassays, were untreated, clean and infestation-free obtained from Kenya Agricultural and Livestock Research Organization, Njoro, Kenya.

2.2. Essential oils

The *C. lusitanica* and *E. saligna* leaf essential oils were provided by the Integrated Biotechnology Laboratory Egerton, Kenya. The hydrodistilled essential oils were previously subjected to Gas chromatography-Mass spectrometry analysis and results are already reported (Bett et al., 2016) (Table 1, Figs. 1 & 2). *C lusitanica* oil was dominated by oxygenated monoterpenes whereas *E. saligna* oil was mainly monoterpene hydrocarbons. The major components found in *C. luistanica* oil were umbellulone (18.38%), α-pinene (9.97%), sabinene (8.16%) and limonene (7.91%) whereas *E. saligna* oil was dominated by 1, 8-cineole (24.26%), o-cymene (9.92%) and α-terpineol (8.81%) (Bett et al., 2016) (Table 1).

2.3. Residual toxicity bioassay

Residual effects of essential oils of *C. lusitanica* and *E. saligna* on adult *A. obtectus, S. zeamais* and *T. castaneum* were evaluated according to the method of Asawalam et al. (2006) with modifications. The oils dissolved in acetone AR (99.8% GC) were applied to 50 g wheat (or 100 g beans) grain samples in self-sealing polythene bags (20 cm × 25 cm; 2 l capacity) at rates of 0.0, 0.05, 0.10, 0.15 and 0.20% v/w. The grains were shaken thoroughly to ensure uniform distribution oil in grains. The solvent was allowed to evaporate for 1 h before polythene bags were sealed. The negative control consisted of untreated grains whereas Actelic Super ™ (0.056% v/w) and crude soya oil (1.0% v/w) served as positive controls. The bags were then sealed and transferred to an experimental room for long-term storage (120 days). A random sub-sample (10 g wheat and 20 g bean grains) was then drawn from each experimental unit at 30, 60, 90 and 120 days post- treatment. Into each sub-sample in 100 ml jars, 20 unsexed test insects (N_T) were introduced and the number of dead insects (N_D) recorded 24, 72, 120 and 168 h post- introduction of insects to estimate adult insect mortality. The percentage adult mortality was computed according to Asawalam et al. (2006) and corrected for natural mortality using Abbott's formula (Abbott, 1925), respectively in Eqs. (1) and (2)

$$\text{Actual Mortality } (\%) = \frac{N_D}{N_T} \times 100 \quad (1)$$

$$\text{Corrected Mortality } (\%) = \frac{(P_O - P_C)}{(100 - P_C)} \times 100 \quad (2)$$

where P_O represent observed and P_C control percent mortalities; N_D and N_T represent number of dead and total number of test insects per jar.

2.4. Residual repellency

Each test essential oil was applied to 20 g wheat or 40 g bean grain samples in special self-sealing polythene bags (20 cm × 25 cm; 2 l capacity) at five concentrations (0, 0.05, 0.10, 0.15 and 0.20% v/w).

Table 1
Retention index & time (min) and percent concentration of chemical constituents of leaf essential oils obtained from *Eucalyptus saligna* and *C. lusitanica*.
(Source: Bett et al., 2016).

No[a]	Rt (min)	Compound Name	RI[b]	% *E. saligna*	% *C. lusitanica*
1	6.87	2,4-Dimethyl-3-pentanone	804	0.09	–
2	8.35	Isovaleric acid	861	0.24	–
3	8.53	2-Methylbutanoic acid	868	0.05	–
4	8.63	(*Z*)-3-Hexenol	873	0.02	–
5	8.82	(*E*)-2-Hexen-1-ol	880	0.02	–
6	9.24	1,2-Dimethyl-1,4-cyclohexadiene	896	0.02	–
7	9.76	2-Methylpropyl-2-methylpropanoate	918	–	0.12
8	9.85	Tricyclene	922	–	0.06
9	9.99	α-Phellandrene	928	0.03	0.99
10	10.12	α-Pinene	935	24.40	9.97
11	10.43	α-Fenchene	948	1.58	0.51
12	10.55	Thuja-2,4(10)-diene	954	0.11	0.06
13	10.72	Benzaldehyde	962	0.05	0.01
14	10.90	3-Methylbutyl propanoate	969	0.12	–
15	10.95	Sabinene	972	0.31	8.16
16	11.29	Myrcene	987	–	2.29
17	11.34	(*E*)-Dehydroxylinalool oxide	989	0.14	–
18	11.58	β-Phellandrene	1000	0.16	0.48
19	11.66	δ-3-Carene	1005	–	6.93
20	11.72	Isoamyl isobutyrate	1009	0.14	–
21	11.80	δ-2-Carene	1013	–	0.53
22	11.95	o-Cymene	1023	9.92	5.81
23	12.02	Limonene	1027	–	7.91
24	12.10	1,8-Cineole	1031	24.26	–
25	12.17	(*Z*)-β-Ocimene	1036	0.16	0.23
26	12.31	Phenylactealdehyde	1045	0.12	–
27	12.55	γ-Terpinene	1059	0.31	0.24
28	12.71	(*E*)-Sabinene hydrate(IPP vs OH)	1069	–	0.47
29	13.01	p-Cymenene	1092	–	1.33
30	13.25	Linalool	1101	–	3.91
31	13.32	Isopentyl isovalerate	1106	0.33	–
32	13.48	p-1,3,8-Menthatriene	1115	–	0.16
33	13.52	endo-Fenchol	1117	2.35	–
34	13.56	α-Thujone	1120	–	0.23
35	13.64	*p*-(*Z*)-Menth-2-en-1-ol	1124	–	0.61
36	13.72	α-Campholenal	1129	1.81	–
37	13.96	[1S-(1α,3α,5α)]-6,6-dimethyl-2-methylenebicyclo[3.1.1]heptan-3-ol	1143	7.13	–
38	14.03	Camphor	1147	–	0.62
39	14.11	Camphene hydrate	1152	0.53	–
40	14.24	Sabina ketone	1159	–	0.22
41	14.33	Pinocarvone	1165	3.02	–
42	14.39	Borneol	1168	4.57	–
43	14.51	Umbellulone	1175	–	18.38
44	14.54	Terpinen-4-ol	1177	1.52	6.12
45	14.66	[α,α],4-Trimethyl-benzenemethanol	1184	–	1.25
46	14.75	α-Terpineol	1189	8.81	1.98
47	14.86	γ-Terpinen-7-al	1207	–	0.19
48	15.06	Verbenone	1208	0.53	–
49	15.24	Eucarvone	1220	–	0.37
50	15.33	Terpinolene	1227	1.43	–
51	15.51	Cumin aldehyde	1238		0.31
52	15.71	Piperitone	1252	0.28	1.19
53	16.19	Thymol	1284	0.27	0.76
54	16.30	Benzyl isobutanoate	1291	0.08	–
55	16.33	Terpinolene	1293	–	0.66
56	17.02	α-Terpinene	1342	–	2.60
57	17.10	2,2,5,5-Tetramethyl-3-cyclopenten-1-one	1348	0.15	–
58	17.44	α-Copaene	1373	0.11	–
59	17.62	Phenylethyl butyrate	1386	0.19	–
60	17.71	(*E*)-Jasmone	1392	0.13	–
61	17.78	3-Isopropylbenzaldehyde	1397	–	0.16
62	17.90	Premnaspirodiene	1407	–	0.09
63	17.98	α-Cedrene	1412	–	0.09
64	18.06	(*E*)-Caryophyllene	1418	–	0.18
65	18.31	Germacrene B	1438	0.08	–
66	18.37	(*E*)-Muurola-3,5-diene	1443	–	0.54
67	18.59	α-Guaiene	1459	0.15	–
68	18.61	(*E*)-Muurola-4(14),5-diene	1461	–	3.40
69	18.69	α-Macrocarpene	1467	–	0.19
70	18.76	α-Curcumene	1473	–	0.21
71	19.02	Viridiflorene	1492	0.09	0.00
72	19.01	Epizonarene	1492	–	0.73
73	19.07	β-Macrocarpene	1497	–	0.18
74	19.14	β-Vetivenene	1502	–	0.11
75	19.24	Durohydroquinone	1510	0.09	–
76	19.33	(*Z*)-Calamenene	1518	–	1.98
77	19.51	α-Dehydro-ar-himachalene	1533	–	0.35
78	19.59	β-Calacorene	1539	–	0.43
79	19.82	γ-Gurjunene	1558	0.05	–
80	19.83	α-Calacorene	1559	–	0.12
81	19.93	Pogostol	1567	0.08	–
82	20.03	Spathulenol	1576	0.43	0.05
83	20.11	Caryophyllene oxide	1582	–	0.23
84	20.11	Globulol	1582	0.17	–
85	20.43	iso-Leptospermone	1608	3.23	–
86	20.45	1,10-di-epi-Cubenol	1611	–	0.35
87	20.51	α-Colocalene	1616	–	0.08
88	20.65	β-Gurjunene	1628	0.08	–
89	20.65	β-Acoradiene	1628	–	0.36
90	20.75	(*Z*)-Cadina-1(6),4-diene	1637	–	0.26
91	20.91	β-Eudesmol	1650	–	0.43
92	21.13	Cadalene	1670	–	0.14
93	21.33	(*Z*)-14-nor-Muurol-5-en-4-one	1688	–	1.89
94	21.46	10-nor-Calamenen-10-one	1699	–	0.17
95	22.55	(Z)-5-Hydroxy-calamenene	1823	–	0.08
96	23.61	Isopimara-9(11),15-diene	1926	–	0.14
97	23.94	Kaur-15-ene	1961	–	0.03
98	24.26	Sandaracopimara-8(14),15-diene	1996	–	0.22
99	24.52	13-epi-Manool oxide	2024	–	0.27
100	25.37	Abietadiene	2115	–	0.12
101	25.81	Nezukol	2163	–	0.68
102	27.35	(*E*)-Totarol	2342	–	0.08

– = Absent.
[a] No = Peak numbers as indicated in Figs. 1 & 2.
[b] RI = Retention Index.

DEET treated and untreated grains were used as positive and negative controls, respectively. The treated grains were transferred to the experimental room for long-term storage (120 days). A random subsample of 2 g wheat and 4 g beans were then drawn from each experimental unit at 30, 60, 90 and 120 days post-treatment. The base of a 14-cm diameter glass Petri-dish was lined with aluminum foil, divided into four equal parts and treated grain samples placed in each quarter equidistant to the center in an alternate untreated (control)-treated arrangement with four replicates per concentration. Twenty (20) unsexed adult stages of *A. obtectus, S. zeamais*, and *T. castaneum* were then released at the center of petri-dish and the top secured by its cover. The number of insects present in the control (N_C) and treated (N_T) grains were recorded 1, 3, 5 and 24 h post- exposure. Percent repellence (PR) values were computed according to Asawalam et al. (2006)

$$\text{Percent repellence (PR)} = \frac{(N_C - N_T)}{(N_C + N_T)} \times 100 \quad (3)$$

2.5. Statistical data analysis

The experimental design used was a completely randomized design (CRD) with four replicates per concentration in all bioassays. The concentrations of essential oils used were 0.0, 0.05, 0.10, 0.15 and 0.20% v/w and these were replicated four times. Adult insects were picked at random and placed in glass jars and petri-dishes accordingly.

The data on insect mortality were corrected for natural mortality using Abbott's formula (Abbott, 1925). In addition, the data on

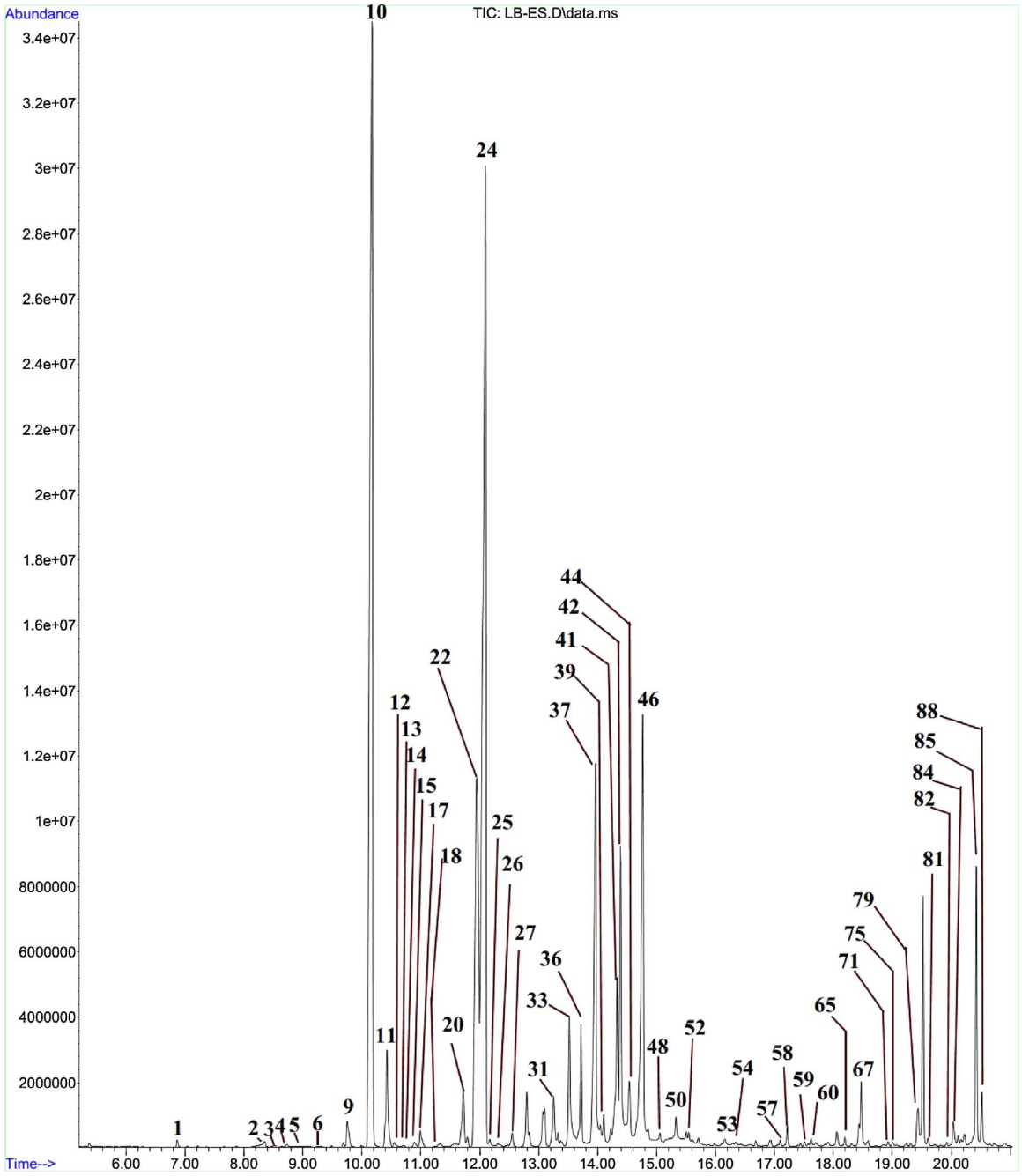

Fig. 1. Chromatogram of the leaf essential oil of *Eucalyptus saligna*. Peaks 1–88 show the essential oil components identified. (Source: Bett et al., 2016).

corrected percentage insect mortality and percentage repellence were homogenized using angular transformation (Sokal and Rohlf, 1995), before being subjected to two-way Analysis of Variance (multiple comparisons) (SPSS, 2010). The means were separated using Tukey Honestly Significant Difference (HSD) test at the 5% significance level (Sokal and Rohlf, 1995). For the relationship between doses of essential oils used and insect mortality, the lethal concentration that killed 50% (LC_{50}) of test insects was determined using Probit Regression Analysis (SPSS, 2010). In a column, any two calculated LC_{50} values whose 95% feducial limits did not overlap were considered as significantly different (Finney, 1971).

3. Results

3.1. Residual contact toxicity

The *C. lusitanica* leaf essential oils produced dose-, insect species- and storage duration-dependent residual contact toxicity against adult *T. castaneum*, *A. obtectus* and *S. zeamais*. At 0.20% v/w and treated grain storage period of 30 days, *C lusitanica* leaf essential oils caused 6.3, 25.0 and 85.0% kill of adult *T. castaneum*, *S. zeamais* and *A. obtectus*, respectively, 168 h post-introduction of test insects (Fig. 3a). The *C. lusitanica* oil treated grains stored for 30 days was toxic to adult *S. zeamais* and *A. obtectus* with LC_{50} values of 0.07 and 0.12% v/w, respectively 168 h post-introduction of test insects. On the other hand, the oil at the same concentration was less toxic to *T. castaneum* with LC_{50} of 0.79% v/

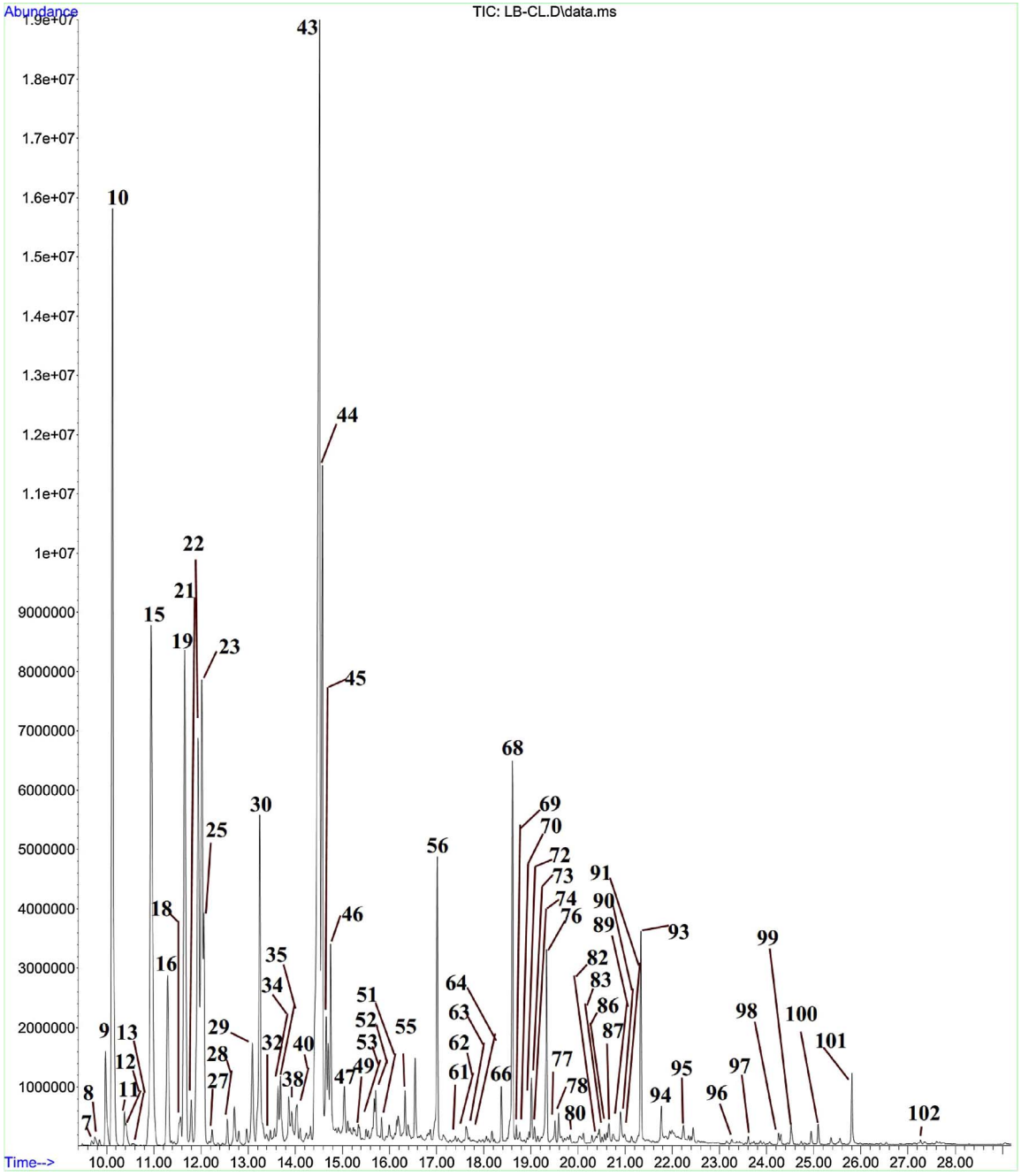

Fig. 2. Chromatogram of the leaf essential oil of *Cupressus lusitanica*. Peaks 7–102 show the essential oil components identified. (Source: Bett et al., 2016).

w, 168 h post-introduction of test insects (Table 2). However, at the same concentration and 120 days grain storage duration, *C lusitanica* oils caused a mortality of 5.0, 17.5 and 65.0% in adult *S. zeamais, T. castaneum* and *A. obtectus*, respectively 90 h post-introduction of test insects (Fig. 4a). At the longest storage duration of 120 days, *C. lusitanica* oil was also toxic to *T. castaneum* and *A. obtectus* and *S. zeamais,* with LC_{50} values of 0.12, 0.13 and 0.38% v/w, respectively 168 h post introduction of test insects (Table 2).

The *E. saligna* oils also produced dose-, insect species- and storage duration-dependent residual contact toxicity against *T. castaneum*, *A. obtectus* and *S. zeamais*. Results also indicated that at a dose of 0.20% v/w, *E. saligna* oil was highly efficacious over treated grain storage period of 30 days causing 32.5, 90.0 and 93.0% mortality against adult *T. castaneum*, *S. zeamais* and *A. obtectus*, respectively 168 h post-introduction of test insects (Fig. 3b). *E. saligna* oil treated grain storage period of 30 days had similarly high toxicity levels with LC_{50} values of 0.003 and 0.005% v/w for *A. obtectus* and *S. zeamais* respectively 168 h post-introduction of test insects. *T. castaneum* was more tolerant, with LC_{50} values of 0.51% v/w 168 h post-introduction of test insects (Table 3).

The same results trend was observed at same concentration and 120 days grain storage duration where *S. zeamais* and *A. obtectus* were most susceptible to *E. saligna* oil causing mortalities of 90 and 93%, respectively 168 h post-introduction of test insects (Fig. 4b). However, at the same concentration and 120 days grain storage duration, *E. saligna* oils caused a mortality of 5.0, 60.0 and 64.2.0% in *T. castaneum*, *S. zeamais* and *A. obtectus*, respectively 168 h post-introduction of test insects. Similar LC_{50} values were observed for *E. saligna* oil after 120 days grain storage duration with *T. castaneum, S. zeamais* and *A. obtectus* recording LC_{50} values of 0.04, 0.10 and 0.70% v/w respectively

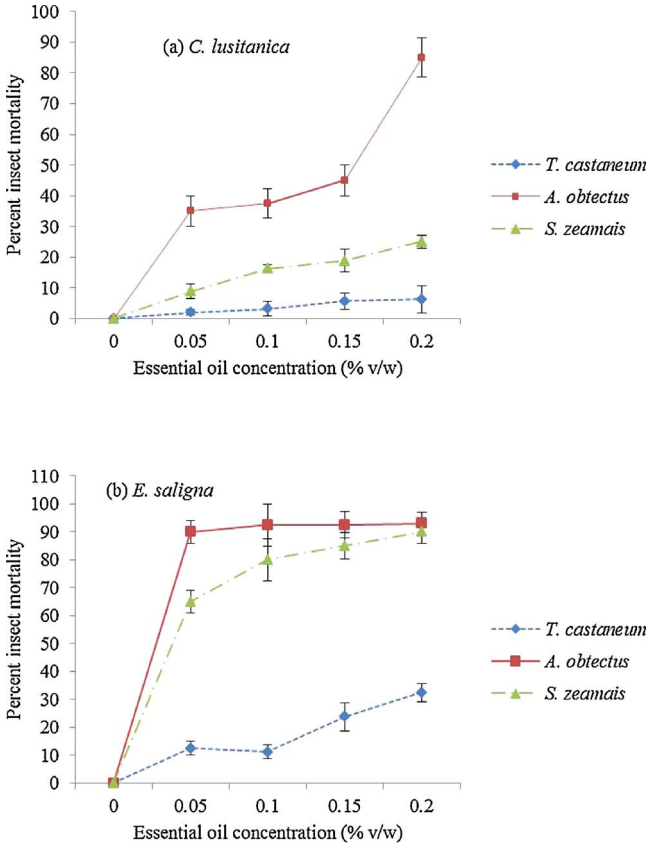

Fig. 3. Percent mortality (Mean ± SE, n = 4) of *T. castaneum, A. obtectus* and *S. zeamais* after 30 days contact with five concentrations (v/w) of (a) *C. lusitanica* and (b) *E. saligna* leaf essential oils.

168 h post-introduction of test insects (Table 3). By comparison, *A. obtectus* was the most susceptible whereas *T. castaneum* was the most tolerant of the three insect species to *C lusitanica* oils.

3.2. Residual repellence of essential oils

3.2.1. C. lusitanica essential oil

Results of residual repellence of *C. lusitanica* leaf essential oils against *S. zeamais, T. castaneum* and *A. obtectus* after 30–120 days of grain storage are presented in Fig. 5. The *C. lusitanica* leaf essential oils produced a dose-, grain storage duration- and exposure time-dependent percent residual repellence against adult *T. castaneum*, *A. obtectus* and *S. zeamais*. Data also showed that, at the highest concentration of 0.20% v/w and 30 days grain storage duration, *C. lusitanica* leaf essential oil was moderately repellent to *S. zeamais* (49.3%) but produced low PR values against *T. castaneum* (13.2%) and A. *obtectus* (32.2%) 12 h post-introduction of test insects (Fig. 3). At the same concentration and 120 days grain storage duration, the oil was moderately repellent with PR values of 37.9, 47.6 and 51.1% against adult *A. obtectus. S. zeamais* and *T. castaneum*, respectively 12 h post-introduction of test insects (Fig. 5).

3.2.2. E. saligna essential oil

Data on residual repellence of *E. saligna* leaf essential oils against *S. zeamais, T. castaneum* and *A. obtectus* after 30–120 days grain storage duration are presented in Fig. 6. The *E. saligna* leaf essential oils produced dose-, grain storage duration- and exposure time-dependent residual PR against adult *T. castaneum* and *S. zeamais* except *A. obtectus* in which all factors were insignificant. The PR values for *E. saligna* essential oils, at 0.20% v/w and 30 days grain storage duration, against adult *A. obtectus, S. zeamais* and *T. castaneum* were 17.8%, 22.9% and 33.6%, respectively, 12 h post-introduction of test insects (Fig. 4). Similarly, at the same concentration and 120 days grain storage duration; oil was moderately repellent with a PR value of 52.4% in *T. castaneum* but weakly repellent to *A. obtectus* (34.0%) and *S. zeamais* (36.6%), 12 h post-introduction of test insects (Fig. 6).

4. Discussion

From the results of residual contact toxicity, *C. lusitanica* and *E. saligna* essential oils exhibited concentration- and storage and contact duration-dependent toxicity against *S. zeamais, T. castaneum* and *A. obtectus.* The fact that oils at a concentration of 0.20% v/w and storage duration of 30–120 days caused moderate to high mortalities of *S.*

Table 2
LC_{50} values (% v/w) of *C. lusitanica* essential oils after 30–120 days storage duration and 24–168 h contact with test insect pests (*Tribolium castaneum, Acanthoscelides obtectus* and *Sitophilus zeamais*).

	Grain Storage Duration (Days)			
[a]Insect/Time (h)	30	60	90	120
T. castaneum				
24	0.29(0.20, 2.59[a])	22.5(−)[b]	3.05(−)[b]	0.78(−)[b]
72	0.29(0.18, 6.51)[c]	0.35(0.22, 14.1)[c]	4.7(−)[b]	3.22(−)[b]
120	0.29(0.18, 6.51)[c]	2.57(0.19,0.78)[c]	0.18(0.12,1.50)[c]	0.84(−)[b]
168	0.79(−)[b]	0.26(0.19,0.79)[c]	0.18(0.12,1.51)[c]	0.12(0.09,0.16)[c]
A. obtectus				
24	1.43(−)[b]	1.22(0.14,2.60)[c]	0.36(0.20,398.0)[c]	0.49(0.23,8649)[c]
72	0.44(0.28,1.40)[c]	0.71(0.35,9.85)[c]	0.61(−)[b]	0.72(0.35,11.50)[c]
120	0.19(−)[b]	0.26(−)[b]	0.19(−)[b]	0.26(−)[b]
168	0.12(−)[b]	0.03(0.0, 0.051)[c]	0.28(−)[b]	0.13(−)[b]
S. zeamais				
24	1.72(−)[b]	0.24(0.16,0.96)[c]	0.35(0.23,114.7)[c]	0.28(0.21,1.60)[c]
72	0.29(0.19,1.50)[c]	0.17(0.12,0.51)[c]	0.31(0.20, 2.30)[c]	0.41(0.24,63.10)[c]
120	0.18(0.12,1.0)[c]	0.07(0.04, 0.09)[c]	0.30(0.21, 13.4)[c]	0.24(0.15, 143.8)[c]
168	0.07(0.04,0.10)[c]	0.06(0.04, 0.07)[c]	0.30(0.21, 13.4)[c]	0.38(0.24,10.27)[c]

Probit Pearson Goodness -of –fit (χ^2 = 3.5–4.8, df = 2, P < 0.27).
[a] Figures in parentheses represent the lower and upper 95% confidence limits for the LC_{50} values.
[b] Significant response in Probit Regression Analysis at P < 0.05.
[c] Insignificant responses.

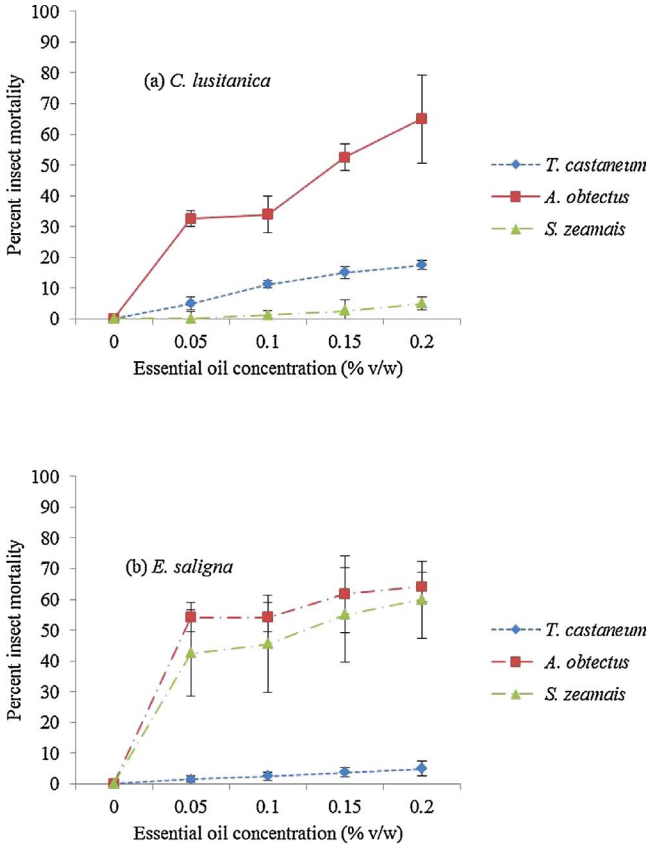

Fig. 4. Percent mortality (Mean ± SE, n = 4) of *T. castaneum, A. obtectus* and *S. zeamais* after 120 days contact with five concentrations (v/w) of (a) *C. lusitanica* and (b) *E. saligna* leaf essential oils.

zeamais and *A. obtectus* demonstrates the potential of oils in the control of stored product insect pests during long-term storage of products. However, *T. castaneum* was clearly tolerant to *C. lusitanica* and *E. saligna* essential oils for storage durations of 30–120 days at concentrations of 0. 20% v/w.

The same trend is observed in other studies where essential oils have exhibited different toxicities against coleopteran and lepidopteran insect pests of stored cereals and legumes. In short-term residual bioactivity studies with crude powders and extracts, significant adult insect mortalities and reproductive inhibitory effects against coleopteran pests of stored food commodities have also been reported (Al-Jabr, 2006; Ogendo et al., 2008a; Nivea et al., 2013). For instance, in local residual contact toxicity studies for 4-month storage duration, *T. vogelii* fruit essential oil had stronger residual toxicity (31–47% kill) than leaf oil (18–21% kill) against *S. oryzae*. The converse was true for *O. americanum* leaf oil that caused 58–75% kill of *C. chinensis* compared to 37–53% mortality rates by *T. vogelii* leaf oil (Ogendo et al., 2011). Nivea et al. (2013) reported that *O. americanum* essential oil was strongly toxic against *C. maculatus* adults (LC_{50} = 0.23 µl l^{-1} air) while the oils from *Hyptis suaveolens* L. (Lamiaceae), *H. spicigera* L. (Lamiaceae) and *Lippia multiflora* Moldenke (Lamiaceae) exhibited higher LC_{50} values of 1.30, 5.53 and 6.44 µl l^{-1} air, respectively. The persistence of the biological activity of the four oils was variable and that from *O. americanum* was most persistent. In addition, Al-Jabr (2006) was able to demonstrate that complete mortality of *Oryzaephilus surinamensis* L. (Coleoptera: Silvanidae) could be achieved by *Mentha viridis* L. (Lamiaceae), *Matricaria chamomilla* L. (Asterales: Asteracae) and *Cinnamomum camphora* L. (Lauraceae) at concentration more than 0.5%. Although, 1% of *Prunus amygdalus* L. (Rosaceae) and *Cymbopogon winterianus* L. (poaceae) gave complete mortality of *T. castaneum* after two weeks of exposure. Conversely, *Rosmarinus officinalis* L. (Lamiales: Lamiaceae) was the least toxic to both insect species.

The observed differential toxicity effects of *C. lusitanica* and *E. saligna* essential oils against four coleopteran pests of stored food grains could be explained by individual and/or synergistic bioactivity of major chemical constituents and differential responses by test insect species (Arriaga et al., 2005; Ogendo et al., 2013). In the current study, it was clear that *A. obtectus* were more susceptible to test essential oils compared to *S. zeamais* and *T. castaneum.* The possible explanation for this variation is the fact that adult stages of *A. obtectus* do not feed, hence become progressively weaker making them more susceptible to toxic effects of test oils. Contact toxicity of essential oils against insect pests has been associated previously to presence of 1, 8-cineole, eugenol, methyl eugenol, and limonene and α-pinene among other bioactive essential oil constituents (Ilboudo et al., 2010; Abd-Elhady, 2012; Olivero-Verbel et al., 2013.). The insecticidal activity of eucalyptus oils

Table 3
LC_{50} values (% v/w) of *E. saligna* essential oils after 30–120 days storage duration and 24–168 h contact with test insect pests (*Tribolium castaneum, Acanthoscelides obtectus* and *Sitophilus zeamais*).

	Grain Storage Duration (Days)			
Insect/Time (h)	30	60	90	120
T. castaneum				
24	0.17(0.14,25.00[a])[c]	0.36(−)[b]	0.36(−)[b]	0.36(−)[b]
72	0.11(0.08, 0.13)[c]	0.42(−)[b]	0.42(−)[b]	0.48(−)[b]
120	0.106(−)[b]	0.43(0.22,2126)[c]	0.62(−)[b]	0.73(−)[b]
168	0.51(0.30, 2.21)[c]	0.26(0.18,1.60)[c]	0.62(−)[b]	0.7(−)[b]
A. obtectus				
24	0.16(−)[b]	0.22(0.19,0.42)[c]	0.58(−)[b]	0.58(−)[b]
72	0.12(0.09,0.14)[c]	0.16(−)[b]	0.27(0.17, 3.90)[c]	0.59(−)[b]
120	0.06(0.05,0.08)[c]	0.06(−)[b]	0.12(0.09,0.17)[c]	0.12(0.09,0.17)[c]
168	0.003(−)[b]	0.38(0.22, 4.77)[c]	0.04(−)[b]	0.04(−)[b]
S. zeamais				
24	0.21(−)[b]	0.07(−)[b]	0.09(−)[b]	0.25(−)[b]
72	0.20(0.16,0.33)[c]	1.7(0.49,2687.7)[c]	0.11(0.08, 0.15)[c]	0.12(0.99,0.15)[c]
120	1.79(0.49, 2688.7)[c]	0.06(0.03,0.08)[c]	0.06(0.03,0.08)[c]	0.12(0.99,0.15)[c]
168	0.005(−)[b]	0.38(0.22, 4.77)[c]	0.09(0.05,0.14)[c]	0.10(0.05, 0.17)[c]

Probit Pearson Goodness -of –fit (χ^2 = 0.001-2.8, df = 2, $P < 0.99$).

[a] Figures in parentheses represent the lower and upper 95% confidence limits for the LC_{50} values.

[b] Significant response in Probit Regression Analysis at $P < 0.05$.

[c] Insignificant responses.

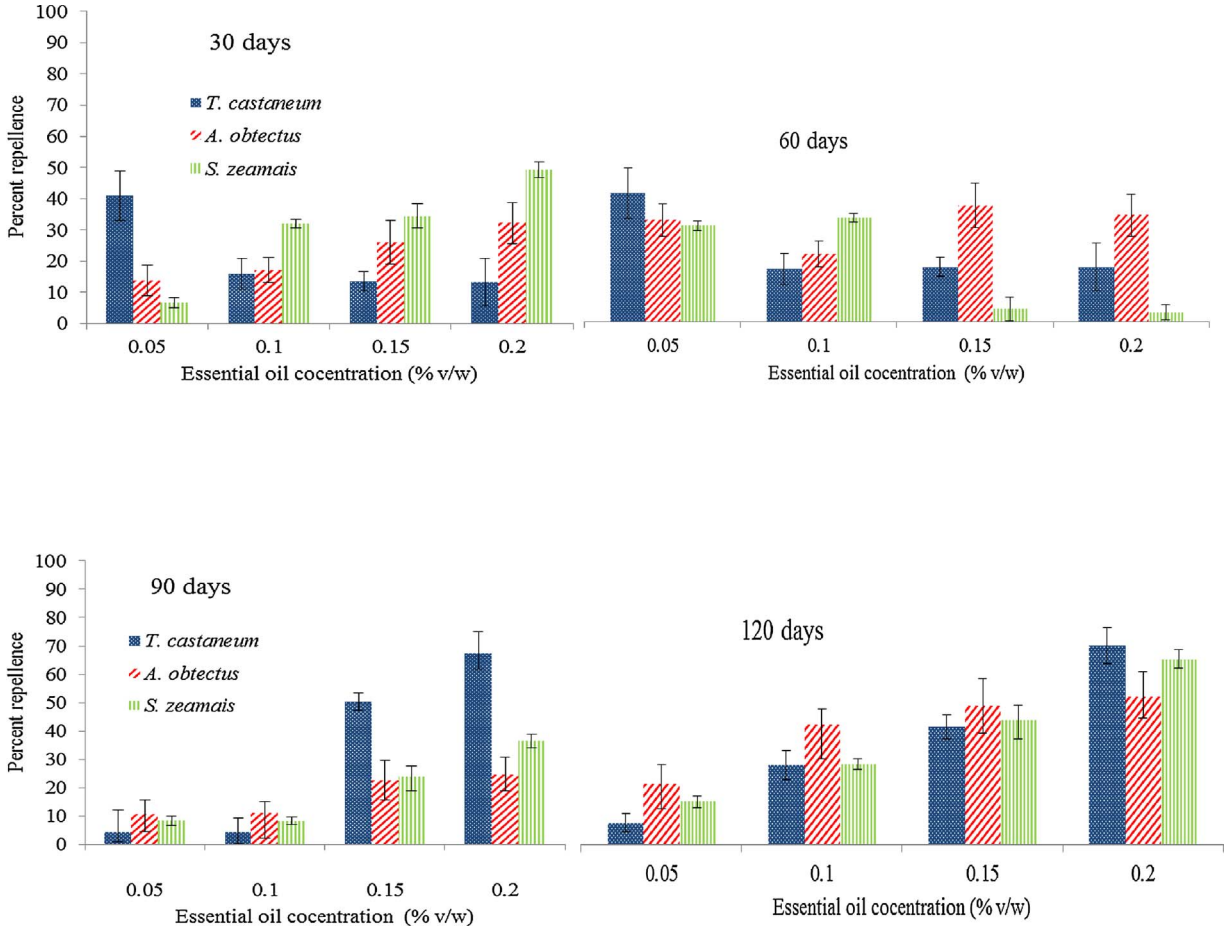

Fig. 5. Percent repellence (Mean ± SE, n = 4) of *C. lusitanica* essential oils against adult *T. castaneum, A. obtectus* and *S. zeamais* 12 h post-exposure of test insects and in (a) 30 days (b) 60 days (c) 90 days and (d)120 days grain storage duration.

has been associated with components such as 1, 8-cineole, citronellal, citronellol, citronellyl acetate, *p*-cymene, eucamalol, limonene, linalool, α-pinene, γ-terpinene, α-terpineol, alloocimene, and aromadendrene (Batish et al., 2008; Su et al., 2006; Liu et al., 2008; Bett et al., 2016).

The results of residual repellency assays of leaf essential oils of *C. lusitanica* and *E. saligna* against test insects showed variable responses. However, repellence was influenced by insect and plant species, concentration of oil exposure time and storage duration. Results on instant repellence have shown clearly that *C. lusitanica* essential oil was a strong repellent against *T. castaneum* at a concentration of 0.20% v/w after 24 h of exposure and moderately repellent against *S. zeamais*. *E. saligna* oil was a poor repellent in all test insects even at higher concentrations and longer exposure periods. The *C. lusitanica* essential oil main constituents umbellulone, α-pinene could have contributed to its repellent activity against *T. castaneum*. However, minor essential constituents such as camphor, α-terpineol, limonene (Table 1) may have contribute synergistically to the overall repellent activity of the major constituents (Mossi et al., 2011).

These results are in agreement with previous local studies in which instant repellency depended on inter-plant species, intra-plant variations, concentration, insect species. Essential oils obtained from *L. camara*, *O. americanum*, and *T. vogelii* were effective repellents against *S. oryzae*, *T. castaneum*, *C. chinensis* and *R. dominica* with PR values in the range of 60–83% (Ogendo et al., 2008b). Chebet et al. (2013) demonstrated that grains treated with crude powders of *T. vogelii* and *Azadirachta indica* Juss (Meliaceae) were equally the most repellent (PR values: 88–90%) against adult *P. truncatus* followed by *L. camara* (PR 73%). Tapondjou et al. (2005) reported essential oils and cymol (*p*-cymene) obtained from *E. saligna* and *C. sempervirens*, to have repellent and toxic effects on *S. zeamais* and *T. castaneum*. The observed variable repellent activity could partly be attributed to the presence of volatile constituents such as monoterpenes and sesquiterpenes which are well-known repellents of phytophagous (biting) insects by acting in the vapour form on the olfactory receptors (Lee et al., 2003; Wang et al., 2006). The highly repellent effects of the main constituents of essential oils such as 1, 8-cineole, terpineol and a-pinene have been demonstrated (Tapondjou et al., 2005; Bett et al., 2016).

The results indicate also that repellence decreased with dosage and even negative repellence (attraction) observed. It was also observed that in residual repellence assay percent repellence increased with exposure time in all test insects. The insecticidal constituents of many plant extracts and essential oils are monoterpenoids. Due to their high volatility they may be lost after long exposure periods (Regnault-Roger et al., 2012). However high volatility can be overcome by mixing essential oils with Kaolin powder (clay) and diatomaceous earths (Keita et al., 2000, 2001; Campolo et al., 2014). Fumigation of *Callosobruchus maculatus* L. (Coleoptera: Chrysomelidae) with *Ocimum basilicum* L. (Lamiaceae) and *O. gratissimum* essential oils mixed with kaolin powder caused 70–99% mortality of adults and adult emergence reduced by 0–4% (Keita et al., 2000, 2001) compared to pure oils. In similar studies, Campolo et al. (2014) found *Citrus sinensis* L. (Rutaceae) essential oil showed a synergistic effect on the mortality of *R. dominica*, if combined with kaolin, and antagonistic effect when admixed with diatomaceous earth.

Similar results trend were also observed by Wambua et al. (2011) who reported a dose- and exposure time-dependent negative repellence (attraction) of *Helicoverpa armigera* Hubner (Lepidoptera: Noctuidae) larvae to chickpea leaves treated with aqueous extracts of *T. vogelii*.

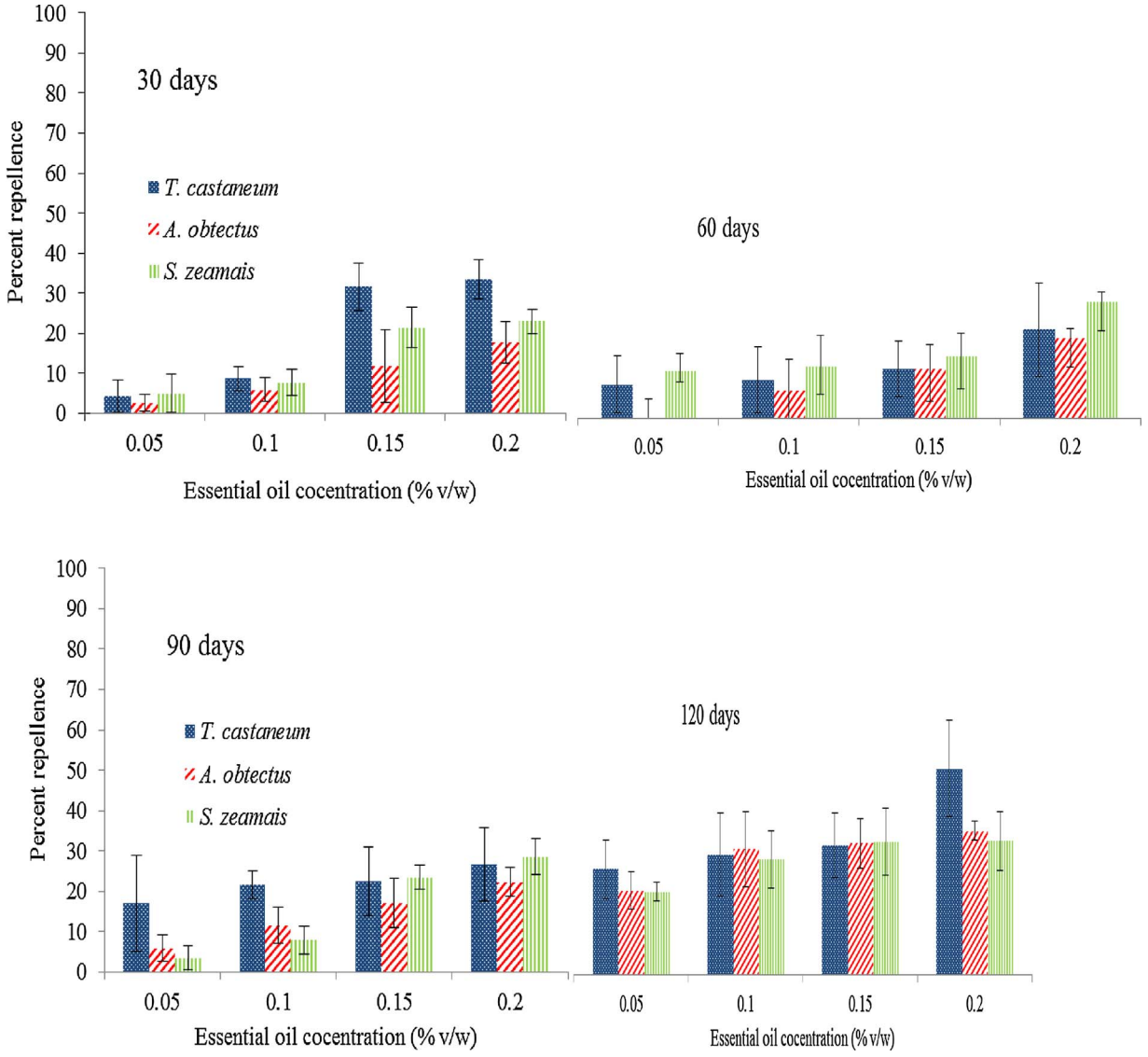

Fig. 6. Percent repellence (Mean ± SE, n = 4) of *E. saligna* essential oils against adult *T. castaneum, A. obtectus* and *S. zeamais* 12 h post-exposure of test insects and in (a) 30 days (b) 60 days (c) 90 days and (d)120 days grain storage duration.

Ogendo et al. (2003) reported that maize grains admixed with Actellic Super™ 2% (Pirimiphos-methyl + Permethrin) dust registered negative PR values against *S. zeamais* due to the arrestment of test insect by the chemical. In similar studies, Ogendo et al. (2008b) reported eugenol produced PR values that decreased with dosage of *C. chinensis* on treated grains. The major cause of the negative PR values was possibly due to the high contact toxicity of eugenol (Ogendo et al., 2008b) against *C. chinensis*.

Essential oil-based insecticides are very important for the control of stored insects because they are active against a variety of insects, fast penetrating and no toxic residues in the treated products (Mbata and Payton, 2013). However, setbacks of using essential oils include volatility, solubility and oxidation, which plays an important role in the their activity, application and persistence. Plant-insecticides as compared to synthetics have manifold effects on insect pests of stored products: insecticidal, repellent and reproduction inhibition (Nerio et al., 2010; Alzogaray et al., 2011; Caballero-Gallardo et al., 2012). The combination of all these methods used simultaneously or alternately, would certainly decrease the undesirable and secondary effects of and also reduce the amounts of insecticide employed. Therefore, the moderate residual toxicity and repellence *C. lusitanica* and *E. saligna* essential oils against *T. castaneum*, *A. obtectus*, and *S. zeamais* can still contribute to the management of these insect pests considering also the other adverse effects of the essential oils.

Conflict of interest/disclosure statement

The above mentioned authors of this manuscript are not aware of any individual and institutional affiliations or financial support or membership that may be considered as a conflict of interest.

Acknowledgements

The authors of this manuscript wish to acknowledge the Lake Victoria Research Initiative (VicRes), Egerton University and National Council for Science, Technology and Innovation for providing funds in support of this study. We are also grateful to the International Centre of Insect Physiology and Ecology (*icipe*), Nairobi for providing analysis equipment. Finally Mr. Xavier Cheseto and Mr Onesmus K. Wanyama both of *icpe* for providing technical support during analysis process and Ms. Ann J. Kiplagat of Egerton University for assisting in bioassays.

References

Abay, G., Karakoç, Ö.C., Tüfekçi, A.R., Koldas, S., Demirtas, I., 2012. Insecticidal activity of *Hypnum cupressiforme* (Bryophyta) against *Sitophilus granarius* (Coleoptera: Curculionidae). J. Stored Prod. Res. 51, 6–10.

Abbott, W.S., 1925. A method of computing the effectiveness of an insecticide. J. Econ. Entomol. 18, 265–267.

Abd-Elhady, H.K., 2012. Insecticidal activity and chemical composition of essential oil from *Artemisia judaica* L. against *Callosobruchus maculatus* (F.) (coleoptera: bruchidae). J. Plant Prot. Res. 52, 347–352.

Al-Jabr, A.M., 2006. Toxicity and repellency of seven plant essential oils to *Oryzaephilus surinamensis* (Coleoptera: Silvanidae) and *Tribolium castaneum* (Coleoptera: Tenebrioidae). Sci. J. King Faisal Univ. (Basic Appl. Sci.) 7, 49–58.

Arriaga, A.M.C., Magalhães, F.E.A., Feitosa, E.M.A., Malcher, G.T., Andrade-Neto, M., Nascimento, R.F., 2005. Composition of the essential oil of *Tephrosia egregia* Sandw. J. Essent. Oils Res. 17, 451–452.

Asawalam, E.F., Emosairue, S.O., Hassanali, A., 2006. Bioactivity of *Xylopia aetiopica* (Dunal) a rich essential oil constituents on maize weevil *Sitophilus zeamais* Motch. (Coleoptera: Curculionidae). Electron. J. Environ. Agric. Food Chem. 5, 1195–1204.

Ayvaz, A., Sagdic, O., Karaborklu, S., Ozturk, I., 2010. Insecticidal activity of the essential oils from different plants against three stored-product insects. J. Insect Sci. 10, 21–23.

Batish, D.R., Singh, H.P., Kohli, R.K., Kaur, S., 2008. Eucalyptus essential oil as a natural pesticide. For. Ecol. Manag. 256, 2166–2174.

Bett, P.K., Deng, A.L., Ogendo, J.O., Kamatenesi-Mughisha, M., Mihale, J.M., 2013. Toxic and repellent properties of *Cupressus lusitanica* and *Eucalyptus saligna* essential oils against *Callosobrochus chinensis* and *Sitophilus zeamais*. In: Ogendo, J.O., Lukhoba, C.W., Bett, P.K., Machocho, A.K. (Eds.), Proceedings of the First International Conference on Pesticidal Plants Volume 1 (August, 2013). ADAPPT-Network: Egerton University. pp. 121–123.

Bett, P.K., Deng, A.L., Ogendo, J.O., Kariuki, S.T., Kamatenesi-Mugisha, M., Mihale, J.M., Torto, B., 2016. Chemical composition of *Cupressus lusitanica* and *Eucalyptus saligna* leaf essential oils and bioactivity against major insect pests of stored food grains. Ind. Crops Prod. 82, 51–62.

Bett, P.K., 2015. Chemical Composition of *Cupressus lusitanica*, Miller and *Eucalyptus saligna*, Smith Essential Oils and Bioactivity Against Lepidopteran and Coleopteran Pests of Stored Grains. Egerton University, Kenya PhD Thesis, 144pp.

Caballero-Gallardo, K., Olivero-Verbel, J., Stashenko, E.E., 2012. Repellency and toxicity of essential oils from *Cymbopogon martinii*, *Cymbopogon flexuosus* and *Lippia origanoides* cultivated in Colombia against *Tribolium castaneum*. J. Stored Prod. Res. 50, 62–65.

Campolo, O., Agatino, V.P., Verdone, M., Palmeri, V., 2012. Survey of solid impurities and active infestation in flours produced in Calabria (Italy). J. Stored Prod. Res. 50, 36–41.

Campolo, Romeo, F.V., Malacrinò, A., Laudani, F., Carpinteri, G., Fabroni, S., Rapisarda, P., Palmeri, V., 2014. Effects of inert dusts applied alone and in combination with sweet orange essential oil against *Rhyzopertha dominica* (Coleoptera: Bostrichidae) and wheat microbial population. Ind. Crops Prod. 61, 361–369.

Chebet, F., Deng, A.L., Ogendo, J.O., Kamau, A.W., Bett, P.K., 2013. Bioactivity of selected plant powders against *Prostephanus truncatus* (Coleoptera: Bostrichidae) in stored maize grains. Plant Prot. Sci. 49, 34–43.

Deng, A.L., Ogendo, J.O., Owuor, G., Bett, P.K., Omolo, E.O., Kamatenesi-Mugisha, M., Mihale, J.M., 2009. Factors determining the use of botanical insect pest control methods by small-holder farmers in the Lake Victoria Basin, Kenya. Afr. J. Environ. Sci. Technol. 3, 108–115.

Finney, D.J., 1971. Probit Analysis, 3rd ed. Cambridge University Press, London.

Ilboudo, Z., Dabiré, L.C.B., Nébié, R.C.H., Dicko, I.O., Dugravot, S., Cortesero, A.M., Sanon, A., 2010. Biological activity and persistence of four essential oils towards the main pest of stored cowpeas, *Callosobruchus maculatus* (F.) (Coleoptera: Bruchidae). J. Stored Prod. Res. 46, 124–128.

Keita, S.M., Vincent, C., Schmit, J.P., Ramaswamy, S., Belanger, A., 2000. Effect of various essential oils on *Callosobruchus maculatus* (F.) (Coleoptera: Bruchidae). J. Stored Prod. Res. 36, 355–364.

Keita, S.M., Vincent, C., Schmit, J.P., Arnason, J.T., Belanger, A., 2001. Efficacy of essential oil of *Ocimum basilicum* L. and *O. gratissimum* L. applied as an insecticidal fumigant and powder to control *Callosobruchus maculatus* Fab. (Coleoptera: Bruchidae). J. Stored Prod. Res. 37 (4), 339–349.

Lee, S., Peterson, C.J., Coats, J.R., 2003. Fumigation toxicity of monoterpenoids to several stored product insects. J. Stored Prod. Res. 39, 77–85.

Liang, Y., Lu, J., Xu, S., Zhao, N.N., Zhou, L., Cheng, J., Liu, Z.L., 2013. Evaluation of repellency of some Chinese medicinal herbs essential oils against *Liposcelis bostrychophila* (Psocoptera: Liposcelidae) and *Tribolium castaneum* (Coleoptera: Tenebrionidae). J. Econ. Entomol. 106, 513–519.

Liu, X., Chen, Q., Wang, Z., Xie, L., Xu, Z., 2008. Allelopathic effects of essential oil from *Eucalyptus grandis* on pathogenic fungi and pest insects. For. China 3, 232–236.

Liu, Z.L., Yu, M., Li, X.M., Wan, T., Chu, S.S., 2011. Repellent activity of eight essential oils of Chinese medicinal herbs to *Blattella germanica* L. Rec. Nat. Prod. 5, 176–183.

Mbata, G.N., Payton, M.E., 2013. Effect of monoterpenoids on oviposition and mortality of Callosobruchus maculatus (F.) (Coleoptera: Bruchidae) under hermetic conditions. J. Stored Prod. Res. 53, 43–47.

Mossi, A.J., Astolfi, V., Kubiak, G., Lerin, L., Zanella, C., Toniazzo, G., de Oliveira, D., Treichel, H., Devilla, I.A., Cansiana, R., Restelloa, R., 2011. Insecticidal and repellency activity of essential oil of *Eucalyptus* sp. against *Sitophilus zeamais* Motschulsky (Coleoptera, Curculionidae). J. Sci. Food Agric. 91, 273–277.

Nerio, L.S., Olivero-Verbel, J., Stashenko, E., 2010. Repellent activity of essential oils: a review. Bioresour. Technol. 101, 372–378.

Nivea, M.S.G., de Oliveira, J.V., Navarro, D.M.A.F., Dutra, K.A., da Silva, W.A., 2013. Contact and fumigant toxicity and repellency of *Eucalyptus citriodora* Hook., *Eucalyptus staigeriana* F., *Cymbopogon winterianus* Jowitt and *Foeniculum vulgare* Mill. essential oils in the management of *Callosobruchus maculatus* (F.) (Coleoptera: Chrysomelidae, Bruchinae). J. Stored Prod. Res. 54, 41–47.

Nukenine, E.N., 2010. Stored product protection in Africa: past, present and future. In: Proceedings of the 10th International Working Conference on Stored Product Protection. Julius-Kuhn-Archiv 2010. pp. 26–41.

Ogendo, J.O., Belmain, S.R., Deng, A.L., Walker, D.J., 2003. Comparison of toxic and repellent effects of *Lantana camara* with *Tephrosia vogelii* Hook and a synthetic pesticide against *Sitophilus zeamais* Motsculsky (Coleoptera: Curculionidae) in stored maize grain. Insect Sci. Appl. 23, 127–135.

Ogendo, J.O., Kostyukovsky, M., Ravid, U., Matasyoh, J.C., Deng, A.L., Omolo, E.O., Kariuki, S.T., Shaaya, E., 2008a. Bioactivity of *Ocimum gratissimum* oil and two constituents against five insect pests attacking stored food products. J. Stored Prod. Res. 44, 328–334.

Ogendo, J.O., Kostyukovsky, M., Ravid, U., Matasyoh, J.C., Deng, A.L., Omolo, E.O., Kariuki, S.T., Shaaya, E., 2008b. Bioactivity of *Ocimum gratissimum* oil and two constituents against five insect pests attacking stored food products. J. Stored Prod. Res. 44, 328–334.

Ogendo, J.O., Deng, A.L., Kostyukovsky, M., Ravid, U., Matasyoh, J.C., Omolo, E.O., Kariuki, S.T., Kamau, A.W., Bett, P.K., Shaaya, E., 2011. Plant essential oils as potential toxicants and protectants of stored food commodities: is there hope for food security for smallholder farmers? In: African Crop Science Conference Proceedings 10: 229–236. African Crop Science Society.

Ogendo, J.O., Deng, A.L., Birech, R.J., Bett, P.K., 2012. Plant-based products as control agents of stored-product insect pests in the tropics. In: Bhat, R., Alias, A.K., Paliyath, G. (Eds.), Progress in Food Preservation. John Wiley & Sons, London, pp. 581–601.

Ogendo, J.O., Deng, A.L., Kostyukovsky, M., Ravid, U., Matasyoh, J.C., Omolo, E.O., Kariuki, S.T., Kamau, A.W., Bett, P.K., Shaaya, E., 2013. Residual bioactivity of *Ocimum americium* L. and *Tephrosia vogelii* Hook essential oils against coleopteran pests and inhibition of wheat seed germination. In: Ogendo, J.O., Lukhoba, C.W., Bett, P.K., Machocho, A.K. (Eds.), Proceedings of the First International Conference on Pesticidal Plants Volume 1 (August, 2013). ADAPPT-Network: Egerton University. pp. 91–94.

Olivero-Verbel, J., Tirado-Ballestas, I., Caballero-Gallardo, K., Stashenko, E.E., 2013. Essential oils applied to the food act as repellents toward *Tribolium castaneum*. J. Stored Prod. Res. 55, 145–147.

Philips, T.W., Throne, J.E., 2010. Biorational approaches to managing stored product insects. Annu. Rev. Entomol. 55, 375–397.

Rajendran, S., Sriranjini, V., 2008. Plant products as fumigants for stored-product insect control (Review). J. Stored Prod. Res. 44, 126–135.

Regnault-Roger, C., Vincent, C., Arnason, J.T., 2012. Essential oils in insect control: low-risk products in a high-stakes world. Rev. Entomol. 57, 405–424.

Rosman, V., Kalinovic, I., Korunic, Z., 2007. Toxicity of naturally occurring compounds of Lamiaceae and Lauraceae to three stored-product insects. J. Stored Prod. Res. 43, 349–355.

SPSS, 2010. SPSS Version 19. IBM Inc.

Sokal, R.R., Rohlf, F.J., 1995. Biometry, 3rd ed. Freedman and Company, New York.

Su, Y.C., Ho, C.L., Wang, I.C., Chang, S.T., 2006. Antifungal activities and chemical compositions of essential oils from leaves of four eucalyptus. Taiwan J. For. Sci. 21, 49–61.

Tapondjou, A.L., Adler, C., Fontem, D.A., Bouda, H., Reichmuth, C., 2005. Bioctivities of cymol and essential oils of *Cupressus sempervirens* and *Euacalyptus saligna* against *Sitophilus zeamais* Motschulsky and *Tribolium confusum* du Val. J. Stored Prod. Res. 41, 91–102.

Toloza, A., Czygadlo, J., Cueto, G.M., Biurrun, F., Zerba, E., Picollo1, M., 2006. Fumigant and repellent properties of essential oils and component compounds against permethrin-resistant *Pediculus humanus capitis* (Anoplura: Pediculidae) from Argentina. J. Med. Entomol. 43, 889–895.

Wambua, L.M., Deng, A.L., Ogendo, J.O., Owuoche, J., Bett, P.K., 2011. Toxic, antifeedant and repellent activity of aqueous crude extracts of *Tephrosia vogelii* Hook on the larval stages of *Helicoverpa armigera* Hubner. Baraton Interdiscip. Res. J. 1, 19–29.

Wang, J., Zhu, F., Zhou, X.M., Niu, C.Y., Lei, C.L., 2006. Repellent and fumigant activity of essential oil from *Artemisia vulgaris* to *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae). J. Stored Prod. Res. 42, 339–347.