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Phytoconstituents and Insecticidal Activity of Different Solvent Leaf Extracts of *Chromolaena odorata* L., against *Sitophilus zeamais* (*Coleoptera*: *Curculionidae*)

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Authors' contributions

This work was carried out in collaboration between all authors. Author OAL designed the study, isolation of the oil, performed the insecticidal activity as well as the statistical analysis. Author IAO managed the literature searches and wrote the final draft of the manuscript. Author ARO managed the analyses of the GC and GC/MS. All authors read and approved the final manuscript.

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ABSTRACT

The chemical profiles of volatile compounds obtained from different leaf extracts of *Chromolaena odorata* L, were determined by gas chromatography (GC) and gas chromatography couples with mass spectrometry (GC/MS). The hexane extract was characterized by abundance of phytol (23.1%), caryophyllene oxide (12.7%), germacrene D (9.0%) and (7.8%) and β-caryophyllene (8.2%). The major constituents of the chloroform extract were dodecyl acetate (13.6%), oleic acid methyl ester (11.2%), *di*-n-octyl phthalate (11.1%) and hexadecanoic acid methyl ester (6.6%).

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Phytol (11.1%), caryophyllene oxide (9.9%), γ-muurolene (6.4%) and hexadecanoic acid (5.4%) were the main compounds of the ethyl acetate fraction. The main components of the methanol extract were hexadecanoic acid (11.2%), caryophyllene oxide (8.5%), α-terpineol (7.8%) and α-cubebene (7.7%). Overall, ubiquitous terpenes are less common when compared with previous investigation. However, fatty acids, aromatic compounds and diterpenoids contributed significantly to the major volatile fractions. The insecticidal sensitivity of different solvent leaf extracts of *C. odorata* towards the adults of *S. zeamais* after 96h exposure was found in the order: methanol > ethyl acetate > chloroform > hexane.

Aims: The aim of this research work was to investigate the chemical constituents of the volatile oils from different solvent extracts of *Chromolaena odorara* and evaluation of their insecticidal properties.

Study Design: Extraction of *C. odorata* with different solvents by Soxhlet apparatus and the insecticidal property.

Place and Duration of Study: Fresh leaves of *C. odorata* were collected from Mowe-Ofada, Obafemi-Owode Local Government Area, Ogun State, Nigeria.

Methodology: Air-dried and pulverized leaves were extracted with hexane, methanol, chloroform and ethyl acetate and their chemical constituents were analyzed by GC and GC/MS. The oils were then evaluated for their insecticidal activity.

Results: A total of sixty-seven compounds were identified, amounting to 96.4%, 91.2%, 91.1% and 93.4% of the total constituents of hexane, chloroform, ethyl acetate and methanol fraction, respectively. The major were compounds phytol, caryophyllene oxide, dodecyl acetate, oleic acid, hexadecanoic acid, di-n-octyl phthalate. The insecticidal sensitivity of different solvent extracts of *C. odorata* towards the adults of *S. zeamais* depicts its potential as an insecticide.

Conclusion: The chemical composition of the volatile compounds differed from each other and the extracts displayed potent insecticidal activities.

Keywords: Chromolaena odorata; solvent; terpenes insecticidal.

1. INTRODUCTION

Chromolaena odorata (L.) R.M. King and H. Robinson (formerly *Eupatorium odoratum*) is one of the world's worst tropical weeds. The weed goes by many common names including Siam weed, Devil weed, French weed and known as 'Akintola Taku' by the Yorubas of Nigeria. It is a rapidly growing and strongly scenting perennial shrub. Native to South America and Central America, it has been introduced into the tropical regions of Asia, Africa and the Pacific, where it is an invasive weed [1]. *C. odorata* is being used traditionally for its many medicinal properties, especially for external uses as in wounds, skin infections, inflammation etc. Studies have demonstrated that the leaf extract has antioxidant [2], anti-inflammatory [3,4], allelopathic [5], cytoprotective, cytotoxicity and hepatoprotective [6], analgesic [1], antimicrobial [7], insecticidal [8],anti-diabetic and anti-cataract [9], anti-schistosomiasis properties [10] and platelet-activating factor (PAF) receptor binding antagonist activity [11].

Previous phytochemical studies showed that this plant contains essential oils, flavonones and chalcolnes with cytotoxicity and anticancer properties [12,13], antimicrobial flavonoids [14], antioxidant phenolic compounds [15,16], anti-inflammatory fatty acids [3], potent α-glucosidase inhibitory and antibacterial active kaurane-type diterpenoids [17] and alkaloids [18].

Essential oils from the plant have shown antifungal and antimicrobial activities and antiradical potential [19], mortality of the maize grain weevil, *Sitophilus zeamais* [20], in vitro potential cytotoxic activity on human epidermic cell line [21], toxic on *Rhipicephalus lunalatus*, ectoparasite of the dwarf goat [22], exploited as insecticide [23], ovicide and larvicide [19]. The chemical constituents of its volatile oils are always dominated by ubiquitous mono-and sesquiterpenes of diverse structural pattern. Some major compounds of its oils so defined include β-caryophyllene, germacrene-D, bicyclogermacrene, geijerene, (*Z*)-β-farnesene, α-pinene, pregeijerene, camphor and limonene [19-37].

Previous volatile compositions were obtained from hydrodistilled samples. In the present study, we report the volatile compounds obtained by extraction with different solvents, from the air-dried samples of *C. odorata*. This procedure

which is quite different from the conventional method such as hydrodistillation has been previously described [38,39]. This is part of our growing interests on the analysis of chemical composition and biological activities of essential oils from Nigerian flora.

2. MATERIALS AND METHODS

2.1 Plant Material

Fresh leaves of *C. odorata* were collected from Mowe-Ofada (6.8N, 3.43E), Obafemi-Owode Local Government Area, Ogun State, Nigeria. Botanical identification of the plant material was carried out at the Herbarium of Department of Botany, University of Lagos, Akoka-Yaba, Nigeria. A voucher specimen (LUH 5098) was deposited at the University Herbarium.

2.2 Preparation of extracts

The air-dried and pulverised leaves (150g) of *C. odorata* were separately extracted with hexane, chloroform, ethyl acetate and methanol using Soxhlet apparatus for 8h. The resultant filtrates were distilled off at low pressure to give a pale to dark-brown color solid residues. Extracts were preserved in a sealed sample tube and stored at 4℃ until analysis. One milligram of each extract was re-dissolved in n-hexane (10mL) prior to analysis.

2.3 Gas Chromatography (GC) Analyses

GC analyses of the different extracts were carried out on a Hewlett Packard HP 6820 Gas Chromatograph equipped with a FID detector and HP-5MS capillary column (30mx0.25mm i.d, film thickness 0.25µm) and the split ratio was 1:25. The oven temperature was programmed from 50℃ (after 2min) to 240℃ at 5℃/min and the final temperature was held for 10 min. Injection and detector temperatures were maintained at 200℃ and 240℃, respectively. Hydrogen was the carrier gas at a flow rate of 1mL/min. 0.5µL of the individual oil was injected into the GC. Peaks were measured by electronic integration. A homologous series of n-alkanes were run under the same conditions for determination of retention indices.

2.4 Gas Chromatography-Mass Spectrometry (GC/MS) Analyses

GC/MS analyses of the different extracts were performed with a Shimadzu GCMS-QP 2010 model gas chromatograph-mass spectrometer equipped with an AOC-20i auto injector (Shimadzu, Kyoto, Japan). Column: HP-5MS, 30mx0.25mm i.d, 0.25µm film thickness. Temperature program: from 50℃ (2min) to 250℃ (10min.) at 3℃/min. Injection temperature: 250℃; injection volume: 1.0µL; Inlet pressure: 37.1kPa; Carrier gas: He (1mL/min); Injection mode: split (10:1); MS interface temp: 250℃; MS mode: EI; detector voltage: 0.9kV; mass range: 40-400amu; scan speed: 769u/s; interval: 0.50s (2 Hz). 1.0µL of each extract in hexane was injected into the GC/MS.

Identification of the constituents

The identification of constituents was performed on the basis of retention indices (RI) determined by co-injection with reference to a homologous series of n-alkanes, under identical experimental conditions. Further identification was performed by comparison of their mass spectra with those from NIST 08 Libraries (on ChemStation HP) and Wiley 9th Version and the home-made MS library built up from pure substances and components of known volatile substances, as well as by comparison of their retention indices with literature values [40,41].

2.5 Insecticidal Activity

Adult insects of mixed sex, 7-14 days old of *Sitophilus zeamais* reared on maize at 25±1℃ and 65%±5% relative humidity (R.H.) were used for the bioassay. The insecticidal toxicity of the different *C. odorata* extracts against *S. zeamais* was carried out according to an established procedure [42]. Filter paper (Whatman No. 1, cut into 2cm diameter pieces) was impregnated with different solvent extracts of *C. odorata* (10-250mg/L air) at doses calculated to give equivalent insecticidal concentrations. The impregnated filter paper was then attached to the undersurface of the Petri dishes (90mm) containing 10 adults of *S. zeamais* subjected to different concentrations of *C. odorata* extracts. Each concentration and the controls were replicated three times. Mortality was determined after 24, 48, 72 and 96h from the commencement of exposure. When no leg movement was observed, insects were considered dead. The percentage insect mortality was calculated using Abbott's formula for natural mortality in untreated controls [43].

2.6 Statistical Analysis

The mean and standard deviation of three experiments were determined. Statistical analysis of the differences between mean values obtained for experimental groups were calculated as means ± standard deviation (SD) of three independent measurements using Microsoft excel program, 2003 and Origin 6.0 for IC_{50}. Data were subjected to one way analysis of variance (ANOVA). *P* values ≤ 0.05 were regarded as significant and *P* values ≤ 0.01 as very significant. The percentage of mortality and lethal concentrations (LC_{50}) values for insecticidal and larvicidal activities were determined using Abbott's formula and probit analysis program, version 1.5, respectively and reported as LC_{50} with 95% confidence intervals, representing the concentrations in µg/mL with 50% mortality rate in 96h.

3. RESULTS AND DISCUSSION

The yield of the extracts obtained from the Soxhlet extracts of the leaves of *C. odorata* were 1.2%, 1.1%, 0.8% and 0.9% (v/w), respectively, calculated on a dry weight basis, for the hexane, chloroform, ethyl acetate and methanol. Table 1 presents the components identified, their percentage compositions and retention indices based of their elution on the HP-5MS column. A total of sixty-seven compounds were identified, amounting to 96.4%, 91.2%, 91.1% and 93.4% of the total volatile constituents of hexane, chloroform, ethyl acetate and methanol fraction, respectively. The hexane fraction was characterized by larger amounts of sesquiterpenes (57.1%) and diterpenoid (23.1%) while, sesquiterpenes (43.4%) and fatty acid esters (27.0%) were the main classes of compounds in ethyl acetate extract. The chloroform extract was dominated by sesquiterpenes (20.6%) and fatty acid esters (54.6%). Aromatic compounds are also prominent (11.1%). However, the methanol extract had relative contents of fatty acids esters (31.7%), sesquiterpenes (23.0%) and monoterpenes (30.8%). Overall, monoterpene hydrocarbon compounds were present only in a small quantity in the ethyl acetate extract (2.2%) but lacking in the other extracts.

The hexane extract was characterized by abundance of phytol (23.1%), caryophyllene oxide (12.7%), germacrene D (9.0%), 2,5-bis-(1,1-dimethyl) phenol (7.8%) and β-caryophyllene (8.2%). Monoterpene compounds could not be identified in this hexane extract. The chloroform extract had its major compounds as dodecyl acetate (13.6%), oleic acid methyl ester (11.2%), di-*n*-octyl phthalate (11.1%), hexadecanoic acid methyl ester (6.6%) and stearic acid methyl ester (6.3%). Monoterpene hydrocarbon compounds could not be identified in the chloroform fraction. Phytol (11.1%), caryophyllene oxide (9.9%), γ-muurolene (6.4%) and hexadecanoic acid (5.4%) were the main compounds of the ethyl acetate fraction. The methanol extract was rich in hexadecanoic acid (11.2%), caryophyllene oxide (8.5%), α-terpineol (7.8%) and α-cubebene (7.7%). There are significant amounts of di-n-octyl phthalate (5.1%) and vanillin (5.0%).

Two forms of essential oils of *C. odorata* growing in Nigeria have been reported. One form contained a higher amount of camphor, limonene and cadinol, but devoid of geijerene and pregeijerene [33] and the second one with major constituents as α-pinene, β-pinene, germacrene D and β-copaen-4α-ol with low contents of geijerene and pregeijerene [32]. The present result though differs from the two previous reports above.

The insecticidal effects of different solvent extracts of *C. odorata* leaves against *S. zeamais* were evaluated by determining the percentage mortality and lethal concentrations (LC_{50}– standard measure of the toxicity of the extract that will kill 50% of the pest) after 96h exposure. Major volatiles of *C. odorata* according to the Literature are available in Table 2. Fig. 1 showed the cumulative percentage mortality of *S. zeamais* exposed to leaf extracts of *C. odorata* at different periods of time. Dose dependent percentage mortality was observed by all the extracts at 96h after treatment, although, there was no significant difference in mortality rates of both hexane and chloroform extracts for the whole exposure time. The order of effect was methanol (93.3%) > ethyl acetate (80%) > chloroform (43.3%) > hexane (33.3%). Table 3 summarizes the insecticidal activity of leaf extracts of *C. odorata* against *S. zeamais* after 96h exposure. The bioassays showed that all the extracts were toxic towards *S. zeamais* after 96h of exposure. The methanol extract was found to be most toxic with 96h-LC_{50} value of 39.13µg/mL air. Moderate insecticidal activity was observed for ethyl acetate (LC_{50} 46.63mg/mL air), while, chloroform and hexane extracts were the least toxic with LC_{50} values of 155.14 and 372.61mg/mL air, respectively, when compared

with the controls (allethrin and permethrin, LC_{50} of 7.45 and 11.10mg/mL air respectively).

Previously, the crude ethanol extract of [44] and essential oils [20] of *C. odorata* was shown to possessed insecticidal activity against *S. zeamais*, while its essential oils are also toxic to some other insect pests [19,22,23,35], this study revealed that methanol and ethyl acetate extracts of *C. odorata* leaf extracts (LC_{50}<51.00µg/mL) have remarkable insecticidal activity on the adult of *S. zeamais* and may be explored as a potential natural insecticidal on weevils.

Table 1. Volatile constituents of *Chromolaena odorata*

Compounds [a]	RI [b]	RI [c]	Percentage composition (%)			
			Hex	Chlor	Etace	Meth
β-Pinene	973	974	-	-	0.4	-
δ-3-Carene	1011	1008	-	-	1.4	-
Limonene	1029	1024	-	-	0.4	-
1, 8-Cineole	1032	1026	-	-	0.2	2.3
cis-Linalool oxide (furanoid)	1079	1067	-	0.8	-	1.5
m-cresol	1103	1072	-	-	-	1.1
trans-Pinocarveol	1136	1135	-	-	0.3	-
cis-Verbenol	1141	1137	-	0.2	0.2	1.2
Isopulegol	1158	1145	-	-	-	1.7
α-Terpineol	1191	1186	-	-	-	7.8
Myrtenol	1198	1194	-	-	-	0.7
β-Citronellol	1235	1223	-	-	-	1.6
Carvone	1239	1239	-	-	-	0.9
Piperitone	1263	1249	-	-	-	1.0
Undecanal	1305	1305	-	0.8	-	-
α-Cubebene	1343	1345	0.1	-	-	7.7
Citronellyl acetate	1352	1350	-	-	-	4.8
Eugenol	1363	1356	-	0.5	-	-
α-Copaene	1376	1373	5.2	3.4	-	1.9
β-Elemene	1389	1389	1.2	0.7	-	-
Vanillin	1409	1393	-	-	-	5.0
β-Caryophyllene	1418	1417	8.2	3.5	1.5	1.6
trans-α-Bergamotene	1429	1432	-	0.2	-	-
Thujopsene	1433	1429	0.8	-	2.3	-
γ-Elemene	1440	1434	0.3	-	-	-
α-Humulene	1456	1452	2.2	-	0.7	-
γ-Muurolene	1477	1478	0.4	-	6.4	-
Germacrene D	1481	1484	7.8	-	-	-
α-Amorphene	1485	1483	-	2.9	-	-
Viridiflorene	1489	1496	-	-	1.4	-
α-Bulnesene	1506	1509	-	-	1.5	-
α-Muurolene	1514	1500	-	-	1.4	-
trans- γ-Bisabolene	1517	1514	-	-	3.4	-
2,5-bis-(1,1-dimethyl) Phenol	1519	1516	7.9	-	-	-
γ-Cadinene	1521	1513	1.9	1.7	-	-
δ-Cadinene	1526	1522	5.2	2.3	-	-
α-Elemol	1543	1548	4.1	-	-	-
Palustrol	1569	1567	-	-	1.7	-
Spathulenol	1576	1577	2.2	0.8	1.1	-
Caryophyllene oxide	1581	1582	12.7	4.2	9.9	8.5
Viridiflorol	1589	1592	1.0	-	-	2.2
Widdrol	1592	1599	-	-	1.8	2.1
Carotol	1596	1594	-	-	2.3	-
Ledol	1600	1602	0.9	-	1.5	-
Dodecyl acetate	1610	1607	5.1	13.6	1.7	-
γ-Eudesmol	1632	1630	-	-	1.0	-
δ-Cadinol	1642	1640	3.4	-	0.9	-

Compounds [a]	RI [b]	RI [c]	Percentage composition (%)			
			Hex	Chlor	Etace	Meth
β-Eudesmol	1646	1649	-	-	4.1	-
α-Bulnesol	1666	1670	-	0.9	-	-
Heptadecene	1689	1689	-	-	-	2.7
Myristic acid	1768	1761	-	-	2.0	2.3
Pentadecanoic acid	1877	1877	-	-	1.0	-
Hexadecanoic acid methyl ester	1926	1927	-	6.6	3.0	2.8
Hexadecanoic acid	1970	1959	-	4.8	5.4	11.2
Hexadecanoic acid ethyl ester	1978	1976	-	-	-	4.2
Kaur-16-ene	2033	2043	-	-	2.2	-
Linoleic acid methyl ester	2095	2096	-	4.3	3.0	1.3
Oleic acid methyl ester	2105	2085	-	11.2	2.9	-
Linolenic acid methyl ester	2109	2108	-	-	0.9	-
Phytol	2114	1942	23.1	3.9	11.1	1.9
Stearic acid methyl ester	2134	2128	2.2	6.3	2.0	2.0
Linoleic acid ethyl ester	2163	2155	-	1.5	-	-
Oleic acid ethyl ester	2169	2180	-	1.3	0.6	1.6
Linolenic acid ethyl ester	2176	2185	-	2.3	0.7	-
Stearic acid ethyl ester	2193	2191	-	1.0	1.4	0.9
(Z)-9-Tricosene	2269	2271	-	0.4	4.4	2.7
di-n-octyl Phthalate	2547	2682	-	11.1	2.5	5.1
	Total		96.4	91.2	91.1	93.4
Monoterpene hydrocarbons			-	-	2.2	-
Oxygenated monoterpenes			-	1.5	2.7	-
Sesquiterpene hydrocarbons			33.2	14.7	18.6	10.2
Oxygenated sesquiterpenes			24.8	5.9	24.8	12.8
Diterpenes			23.1	3.9	13.3	1.9
Aromatic compounds			7.9	11.1	2.5	6.2
Fatty acids			7.3	54.6	27.0	31.7

[a] Elution order on HP-5MS capillary column; [b] Retention indices on HP-5MS capillary column; [c] Literature Retention indices (see Experimental); - Not identified; Hex =Hexane extract; Chlor = Chloroform extract; Etace = Ethyl acetate extract; Meth = Methanol extract

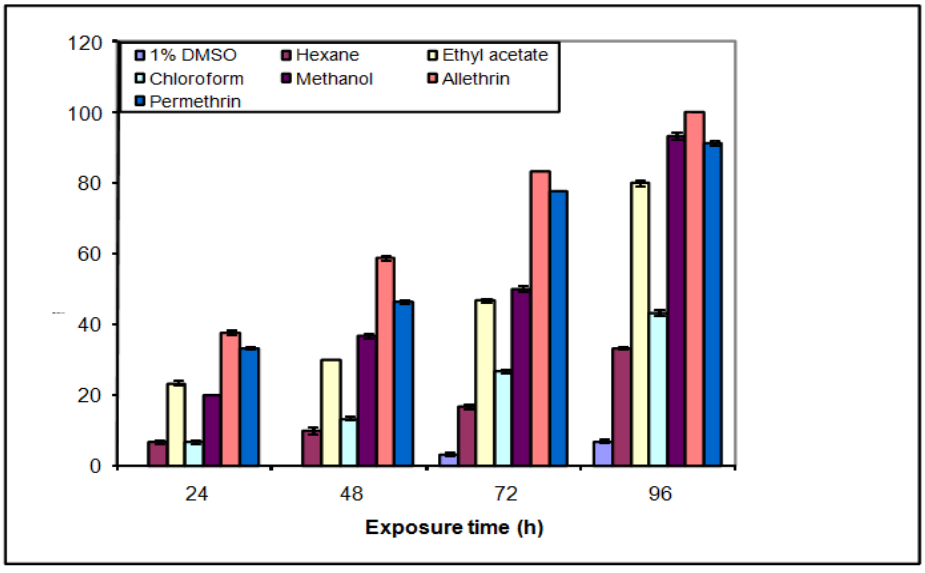

Fig. 1. Cumulative percentage mortality of *Sitophilus zeamais* exposed to leaf extracts of *C. odorata* at different periods of time

Table 2. Major volatiles of *C. odorata* according to the literature

Origin (Part)	Major Constituents	References
Benin (L)	pregeijerene (29.9%), germacrene D (21.6%), β-caryophyllene (14.3%), geijerene (10.1%), α-pinene (8.0%)	24
Benin (L)	α-pinene (20.7%), pregeijerene (14.6%), geijerene (12.0%), β-pinene (10.3%), germacrene-D (9.7%)	19
Benin (-)	β-caryophyllene (21%) and germacrene-D (15.3%)	19
Cameroon (L)	pregeijerene and geijerene represent 34.1%, α-pinene (14.3%) and γ-muurolene (9.8%)	25
Cameroon (L)	bicyclogermacrene (12.55%), geijerene (11.85%), (Z)-β-farnesene (9.98%) and α-pinene (9.36%)	22
Congo (L)	pregeijerene and geijerene represent 19%, p-cymene (22.2%) and thymol acetate (15.8%)	22
Togo (L)	β-caryophyllene (25.2%), germacrene D (18.8%) and linalool (7.9%)	21
India (Ap)	pregeijerene (14.2%), epi- cubebol (9.8%), cubebol (8.6%) and cis-sabinene hydrate (5.7%)	26
India (Fr)	germacrene D (24.8%), geijerene (12.6%), pregeijerene (12.5%) and cyperene (7.8%)	26
Ivory Coast (L)	α-pinene (21.15%), pregeijerene (11.68%), β-pinene (10.12%) and germecrene D (9.50%)	27
Ivory Coast (-)[a]	geijerene and pregeijerene	34
Ivory Coast (-)	pregeijerene (19.9%), α-pinene (17.9%), geijerene (11.4%), β-pinene (10.6%) and germacrène D (9.8%)	23
Ivory Coast (L)	germacrene D (14–13.9%), pregeijerene (9.7–8.1%), ß-caryophyllene (9.5–17.2%), caryophyllene oxide (8.3–11.3%) and geijerene (8.0–9.5%)	31
"	germacrene D (14.4%), β-caryophyllene (14.0%) and trans-longipinocarveol (14.0%)	"
"	β-caryophyllene (21.3%), germacrene D (18.9%), pregeijerene (11.2%) and γ-cadinene (8.2%)	"
"	β-caryophyllene (16.0%), germacrene D (15.4%), pregeijerene (13.0%) and geijerene (11.1%)	"
"	pregeijerene (20.8%), germacrene D (18.2%) and α-pinene (8.6%)	"
"	germacrene D (34.5%), β-caryophyllene (15.4%) and pregeijerene (14.6%)	"
Nigeria (L)	α-pinene (42.2%), β-pinene (10.6%), germacrene D (9.7%), β-copaen-4α-ol(9.4%), (*E*)-caryophyllene (5.4%) and geijerene/pregeijerene (7.5%)	32
"(L)[a]	camphor, limonene and cadinol, but devoid of geijerene and pregeijerene	32
Vietnam (L)	geijerene (42.5%) and β-cubebene (12.5%)	29
Thailand (-)	pregeijerene (17.6%), germacrene D (11.1%), α-pinene (8.4%),β-caryophyllene (7.3%), vestitenone (6.5%) and β-pinene (5.6%)	30
China (-)	*trans*-caryophyllene (16.22%), δ-cadinene (15.53%), α-copaene (11.32%) and caryophyllene oxide (9.42%)	35

[a] *Quantitative data not available; L = Leaf; Ap = Aerial Parts; Fr = Fruits; - Not known*

Table 3. Insecticidal activity of *C. odorata* leaf extracts on *Sitophilus zeamais* [a]

Extract	LC50(95%) CI)[b]	Slope ±SE	X^2 (df)	Regr. Line
Hexane	372.61(148.93-831.29)	1.01±0.30	1.09 (3)	y=8.65x-4.95
Chloroform	155.14(87.98-560.63)	1.16±0.27	0.66 (3)	y=12.32x-8.30
Ethyl acetate	46.63 (31.74-64.80)	1.64±0.33	2.81 (3)	y=18.68x-1.70
Methanol	39.13 (23.09-61.30)	1.41±0.30	1.83 (3)	y= 23.32x-8.30
Allethrin [d]	7.45 (2.01-14.65)	0.8±0.72	1.41(3)	y=21.15x+17.05
Permethrin [d]	11.10 (6.03-23.19)	1.01±0.00	1.97 (3)	y= 20.54x-10.08

[a] Mean ± SE (n = 3) at 96 h; [b] LC_{50} (95% CI)-Lethal concentrations with 50 % mortality rate and 95% confidence intervals (95% CI); [c] χ-Chi-Square for Heterogeneity (calculated at 0.05 Level); [d] Insecticidal activity (LC_{50}) of controls at 72 h. Regr. = Regression

The methanol and ethyl acetate extracts of *C. odorata* leaf (LC_{50} < 51.00 µg/mL) exhibited potent insecticidal activity against the adults of *S. zeamais*, using related bioassay, comparable with data from other plant extracts such as methanol extract of *Hyptis spicigera* (50% mortality) at 48h exposure [45], methanol extract of *Garcinia kolae* (70-100% mortality) [46], methanol extract of *Vitex cymosa* (70% mortality) [47] and the lethal concentrations (LC_{50}) of ethanol extracts of *Acorus calamus* and *Annona squamosa* were between 11.73 to 32.34% [48]. It is well known that the bioactivity of a plant extract could be due to the major compound or a synergy between the major and minor constituents. It is evident that one or more compounds present in the extracts may have been responsible for the observed insecticidal property. Plant extracts and volatile oils containing a large amount of phytol, n-hexadecanoic acids and caryophyllene oxide were known to exhibit insecticidal activities against Coleopteran stored products [38,39,49-51].

4. CONCLUSION

The present compositional pattern of the extracts of *C. odorata* is unusual due to the fact ubiquitous *monoterpene* and *sesquiterpene* compounds that are characteristics of previous reports are either absent or present in low amounts. Only β-caryophyllene, germacrene D, γ-muurolene and caryophyllene oxide were identified in significant proportions in the present study. In this study, caryophyllene oxide, phytol, dodecyl acetate, oleic acid methyl ester, di-n-octyl phthalate and hexadecanoic acid methyl ester were present in higher quantities. The insecticidal activity is in agreement with previous studies indicating any forms of the extracts of *C. odorata* should posses this property irrespective of the method of extraction.

CONSENT

Not applicable.

Ethical APPROVAL

All authors hereby declare that all experiments have been examined and approved by the appropriate ethics committee and have therefore been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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