



RESEARCH PAPER

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Chemical composition and bioefficacy of *Dennettia tripetala* and *Uvariadendron angustifolium* leaves essential oils against the angoumois grain moth, *Sitotroga cerealella*

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Key words: Fumigation, toxicity, essential oil, *D. tripetala*, *U. angustifolium*, *S. cerealella*.

<http://dx.doi.org/10.12692/ijb/5.8.161-172>

Article published on October 29, 2014

Abstract

The essential oils of the leaves of two aromatic species collected in Benin, *Dennettia tripetala* and *Uvariadendron angustifolium* were analyzed by GC and GC / MS. The major components of the *D. tripetala* oil were 2-Phylnitroethane (52.6%), linalol (26.8%) and methyl eugenol (5.6%). That *U. angustifolium* was dominated by geranial (44.9%), neral (32.1%) and geraniol (2.0%). The evaluation of the toxicity on *S. cerealella* was performed in the laboratory by a fumigation method in a closed glass jar at a temperature of 29 ± 2 °C and natural photoperiod with a relative humidity of $70 \pm 10\%$. The results show an insecticidal effect on the samples for the two dose $0.5\mu\text{l.ml}^{-1}$ 24 h after exposure, with an effect significantly higher in the case of *D. tripetala* ($\text{LC}_{50} = 0.253\mu\text{l.ml}^{-1}$ and $\text{LC}_{99} = 2.685\mu\text{l.ml}^{-1}$) efficiency. This toxicity of the essential oils was also illustrated by the significant inhibition of emergence of insects compared to control groups, without affecting the germination of rice seeds treated.

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Introduction

Rice is an important food product in the world economy. During the phase just prior to harvest and especially after harvest during storage, this product is attacked by insects stocks including Angoumois grain moth, *Sitotroga cerealella* (Olivier, 1789). It is considered a dangerous pest for stored grains and difficult to combat. Today, the infestation of rice stocks by Angoumois grain moth emerges as a serious problem in the rice-growing areas in Benin (Togola *et al.*, 2010). Under conditions of heavy infestation, the stored products can suffer 100% loss. *S. cerealella* attacks result in the reduction of the weight of products, lower germination of seeds and the loss of nutritional value and market value. Control of this pest of many grains revealed the use of ionizing radiation from gamma source of cobalt 60 and resistant varieties of grain in the case of rice, wheat or corn; parasites, pathogens or predators (*Trichogramma* spp *Blattisocius tarsalis* *Cotesia ruficrus*, and *Bracon hebetor* *Pteromalus cerealella*) as a biological insecticide on different developmental stages of *S. cerealella*; frequent use of synthetic chemicals such as deltamethrin, malathion and phosphine fumigation alone or combined treatments and mostly based powders or extracts of plants or insecticides potential repellents such as *Cymbopogon citratus*, *Chenopodium ambrosioides*, *Azadirachta indica*, and *Khaya ivorensis* were studied. (Adjalian *et al.*, 2014). However, unlike pests such as *Sitophilus* spp, *Rhizopertha dominica*, , *Tribolium* sp, little work is done on the fight against the Angoumois grain moth out of volatile extracts.

In Some essential oils are Traditionally Farming through fumigant or touch actions to protect grain storage pests Against, a suitable method to preserve products Stored in warehouses and on small farms (Shaaya *et al.*, 1997, Bell, 1994). According Alzouma *et al.* (1994) fumigation is the most cost effective tool for managing pests stored. Activities fumigation *S. cerealella* were evaluated from the essential oil of garlic (*Allium sativum*). Fumigation toxicity of the essential oil of neem seeds in doses of 25-200 pi caused 100% mortality of adults and larvae. The

toxicity of extracts of *Eugenia aromatica* (L) in the protection of six varieties of NERICA rice paddy infested *S. cerealella* showed that the extract produced a low adult emergence. Also these treatments did not affect the viability and capacity of water absorption of grains compared to the control treatment (Aringbangba, 2011). The toxicity of essential for stored product insects oils is influenced by the chemical composition of the oil and used part (Don-Pedro, 1966; Lee *et al.*, 2001).

The search for new biologically active molecules against populations *Sitotroga cerealella* has explored two Annonaceae flora of Benin. *Uvariodendron angustifolium*, syn. *Uvaria angustifolia*. This species, in the south find of Benin, is farming in traditional medicine to treat rheumatism and stomach ache malaria, or for flavoring local dishes (leaves) (Noudogbessi *et al.*, 2014). *Dennettia tripetala* (G.) (Baker f.) GE Schatz where the leaves and fruits are Farming in combination with herbs for the treatment other of cough, infantile convulsions, and worm infestation (Ejechi and Akpomedaye, 2005) *Dennettia tripetala* extracts have been reported about also to exhibit insecticidal properties (Egwunyenga *et al.*, 1998) and antifungal (Nwachukwu and Osuji, 2008). In the present study, the chemical constituents of essential oils from *Dennettia tripetala* and *Uvariodendron angustifolium* were determined, and the insecticidal activity of these essential oils was tested through toxicity fumigation against the adult stages and the emergence of the F1 generation of the stored-products pest, *Sitotroga cerealella* and also the effect of treatments on the germination of rice grains. No study has been reported previously concerning the activity of these compounds as fumigants against this stored product insect. The essential oils were applied primarily we adults to prevent prevention egg mass output and further damages from larvae and for the protection of rice grains.

Materials and methods

Plant material and extraction of essential oils The leaves of *Dennettia tripetala* Baker f. and

Uvariadendron angustifolium (Engl. & Diels) RE Fries, family Annonaceae, were harvested in the Ewe-Adakplamè locality in the municipality of Ketou in the south of Benin in 2013. They were identified and certified at the National Herbarium of the University of Abomey-Calavi. In the laboratory, they were spread on the bench away from the light at 20 ° C. Essential oils were obtained by hydro distillation from the leaves (200 to 250g) for 3 hours using Clevenger-type extractor. The less dense than water species are collected by simple decantation and dried over anhydrous sodium sulfate. The extracted oils were stored at 4 ° C and protected from light in amber vials. Oil yields were calculated using the following formula:

$$\text{Yield (\%)} = \frac{\text{weight of oil (g)}}{\text{Mass of plant materiel (g)}} \times 100$$

Insects

Strains *Sitotroga cerealella* used for mass rearing for this study came from the reserve West Africa Rice Development Association, ex International Institute of Tropical Agriculture (Benin). They were reared in the laboratory at the temperature of 29 ± 2° C with relative humidity of 70 ± 10% and natural photoperiod in glass jars or plastic on paddy rice as a substrate.

Analysis of the volatile constituents

GC/MS

The essential oils were analyzed on a Hewlett-Packard gas chromatograph Model 7890, coupled with a Hewlett-Packard MS model 5875, equipped with a DB5 MS column (30m x 0.25mm; 0.25µm), programming from 50°C (5 min) to 300°C at 5°C/min, 5 min hold. Helium as carrier gas (1.0 ml/min); injection in split mode (1:30); injector and detector temperature: 250 and 280°C respectively. The MS working in electron impact mode at 70 eV; electron multiplier: 2500eV; ion source temperature: 180°C; mass spectra data were acquired in the scan mode in *m/z* range 33-450.

GC/FID

The essential oils were analyzed on a Hewlett-Packard gas chromatograph Model 6890, equipped

with a DB5 MS column (30m x 0.25mm; 0.25µm), programming from 50°C (5min) to 300°C at 5°C/min, 5min hold. Hydrogen was used as carrier gas (1.0 ml/min); injection in split mode (1:60); injector and detector temperature, 280 and 300°C respectively. The essential oil is diluted in hexane: 1/30. The compounds assayed by GC in the different essential oils were identified by comparing their retention indices with those of reference compounds in the literature and confirmed by GC-MS by comparison of their mass spectra with those of reference substances (Rösch *et al.*, 1999; Adams, 1989); Swigar and Silverstein, 1981).

TEST

All tests were performed in the laboratory at a temperature of 29 ± 2 ° C and natural photoperiod with relative humidity of 70 ± 10%.

Fumigation toxicity

The device used consists of glass jars containing 50g capacity of 1 liter of paddy rice (*Oryza sativa* L.) variety of IR841, cotton mass was suspended in 0.3 g using a wire attached to the inner face of the lid of the jars. Concentrations (0, 0.2, 0.5, 1 and 3 µl.ml⁻¹) were selected after several preliminary tests of each essential oil dissolved in absolute ethanol were tested. The control was carried out with pure 96% ethanol. A 50µl volume of each solution thus prepared was taken and applied onto the cotton. Three replicates were performed for each dose and were introduced into each jar containing ten (10) couples of *Sitotroga cerealella* adult's aged 0 to 24 or 10 males and 20 females from the breeding ground, all sealed. Mortality in populations of *S. cerealella* exposed to the insecticidal activity by fumigation with different treatments was observed for 24h, 48h, 72h and 96h. The number of dead individuals was counted after each exposure time. If no movement of the wings or legs is observed, the insect is considered dead. There are, in fact in any population treated natural mortality adds to the mortality caused by this toxic, the mortality percentages were corrected by Abbott's formula. The experimental units were then observed

at regular intervals of time (24 h) for the emergence of young insects from the 20th day until the 45th day after treatment.

Effect of treatments on seed germination of rice paddy

The effect of essential oils on seed germination of paddy rice was evaluated. Paddy was treated with different concentrations of essential oils mentioned before. After 11 days of treatment (sufficient for mortality of all insects exposed time) with different concentrations of the tested oil, rice seeds were transferred to plates containing kneaded wet cotton with water to obtain their seeds. The percentage of germination was computed Ogendo *et al.* (2004) as follows:

$$\text{Germinations (\%)} = (\text{number of seed germinated}) / (\text{Total grain sampled}) \times 100\%$$

Statistical analysis

The raw data from the experiments performed were processed statistically by the method of analysis of variance (ANOVA) using SAS software (Statistical Analysis System) Version 9.1 (Dagnelie, 1975). They underwent the following changes: $2\text{Arcsin}(\sqrt{X/n})$, X being the number of dead insects under the effect of the essential oil and n denotes the total number of insects added to each jar. $\sqrt{X+0,5}$ (X is the number of young *S. cerealella* having emerged from the

substrate).

The masses of being attacked quantitative and continuous data, observing the conditions of normalization and equal variance seeds have undergone no statistical transformation.

Finally, it was performed a structuring medium using the Newman-Keuls test (Dagnelie, 1975). Statistical results were considered significantly different when the null hypothesis probability is less than or equal to 5%. For more accurate results, the effectiveness of the toxicity of these oils was assessed, and the LC₅₀ and LC₉₉ calculated. They were deduced from the plot of the regression by the method of Finney. For this, the corrected mortality percentages are converted into probit.

Results

Average yields of essential oil were obtained on three replicates. The essential oil yield of *D. tripetala* is relatively better (0.95% ± 0.03) than *U. angustifolium* and respectively (0.92 ± 0.02%). Fifty-one (51) compounds have been identified in the leaves essential oil *Dennettia tripetala*, representing 98% of the oil. These main components are 1-phenyl-2-nitro-ethane (52.6%), methyl eugenol (5.6%) and linalool (26.8%). The essential oil of *U. angustifolium* leaves consists of forty-three (43) compounds dominated by geranial (44.9%), neral (32.1%) and geraniol (2.0%).

Table 1. Yields and chemical composition of the essential oil from *D. tripetala* leaves of *D. tripetala*.

N°	Names of the compound	RI	(%)
1	Cis-3-Hexenol	846	t
2	2E-Hexenol	857	0.1
3	Hexanol	860	0.1
4	α-Thujene	918	t
5	α-Pinene	926	0.8
6	Camphene	942	0.2
7	Isomer triethylbenzene	953	t
8	Isomer triethylbenzene	956	t
9	Sabinene	965	1.3
10	β-Pinene	970	0.6
11	6-Methyl-5-Heptene-2-ol	978	t
12	Myrcene	982	0.2
13	α-Phellandrene	998	t
14	δ-3-Carene	999	t
15	α-terpinene	1009	t

16	Para-Cymene	1017	0.3
17	Limonene	1022	0.5
18	β -Phellandrene	1023	0.1
19	Eucalyptol	1025	0.2
20	(Z)- β -Ocimene	1029	0.1
21	(E)- β -Ocimene	1039	1.0
22	δ -Terpinene	1051	0.2
23	Cis Oxide de Linalol	1064	0.1
24	Terpinolene	1078	t
25	Trans Oxide de Linalol	1080	0.1
26	Linalol	1095	26.8
27	cis-para-Menth-2-en-1-ol	1118	0.1
28	Benzeneacetonitrile	1131	0.1
29	Borneol	1166	0.3
30	Terpinen-4-ol	1174	0.4
31	Naphthalene	1178	0.2
32	α -Terpineol	1188	1.0
33	Bornyl acetate	1277	0.1
34	2-Phenylnitroethane	1296	52.6
35	α -Cubebene	1339	t
36	Eugenol	1343	1.0
37	α -Copaene	1369	0.1
38	β -Elemene	1382	0.8
39	Methyl eugenol	1390	5.6
40	β -Caryophyllene	1414	0.5
41	Neryl acetone	1438	0.1
42	α -humulene	1450	0.1
43	Germacrene D	1475	0.7
44	α -selinene	1489	0.2
45	E,E- α -Farnesene	1490	0.1
46	Germacrene A	1501	0.1
47	Elemol	1541	0.2
48	Germacrene B	1553	t
49	Spathulenol	1571	0.2
50	Oxide de Caryophyllene	1577	0.3
51	Guaiol	1589	0.1
Total			97.6%
Essential oil yield (%)			0.95%
Monoterpenic hydrocarbons			5.5%
Oxygenated monoterpenes			30.1%
Sesquiterpenic hydrocarbons			2.6%
Oxygenated sesquiterpenes			0.8%
Aromatic oxygenated compounds			58.2%
oxygenated aliphatic compounds			0.4%
t (traces) \leq 0.1% ; RI = Retention Index			

Effectiveness of essential oils tested against S. cerealella

Mean mortality caused by the influence of two different concentrations of essential oils tested on adult *S. cerealella* populations are shown in Tables 3 and 4. Toxicity of essential oils depends on the concentration and duration exposure. They caused a highly significant mortality ($P < 0.001$) of adult *S. cerealella* from the lowest dose. It reached 100%

mortality at a dose of $0.2 \mu\text{l.ml}^{-1}$ essential oil *D. tripetala* whereas this rate is reached after 48 hours at a dose of $0.5 \mu\text{l.ml}^{-1}$. This toxicity has been also shown 45 days after infestation by the total inhibition of the emergence of young insects from *S. cerealella* $0.5 \mu\text{l.ml}^{-1}$ for both tested unlike in the controls oils. Thus, a highly significant difference ($p < 0.001$) was also noted for medium emergences regarding both treatments.

Table 2. Yield and chemical composition of the essential oil from *U. angustifolium* leaves.

N°	Names of the compound	RI	(%)
1	α -pinene	934	1.4
2	camphene	949	t
3	β -pinene	978	0.8
4	6-methylhept-5-en-2-one	984	1.3
5	myrcene	989	0.4
6	dehydro-1,8-cineole	990	t
7	δ -3-carene	1007	t
8	α -terpinene	1018	t
9	p-cymene	1024	0.2
10	limonene	1028	0.2
11	δ -terpinene	1057	0.2
12	<i>cis</i> -linalool oxide	1069	t
13	<i>trans</i> -linalool oxide	1073	t
14	linalool	1098	1.7
15	(Z)-isocitral	1162	0.2
16	p-mentha-1,5-dien-8-ol	1166	0.2
17	(E)-Isocitral	1180	1.4
18	<i>trans</i> -carveol	1191	0.1
19	<i>cis</i> -carveol	1226	0.1
20	neral	1244	32.1
21	geraniol	1253	2.0
22	geranial	1274	44.9
23	geranic acid	1353	0.8
24	α -copaene	1379	0.5
25	β -elemene	1390	0.1
26	β -caryophyllene	1425	1.8
27	α -humulene	1459	0.3
28	δ -muurolene	1480	0.1
29	germacrene-D	1486	1.1
30	β -selinene	1492	1.3
31	α -selinene	1499	0.5
32	δ -cadinene	1526	0.2
33	elemol	1552	0.3
34	germacrene-B	1564	0.5
35	spathulenol	1586	0.1
36	caryophyllene oxide	1591	1.2
37	δ -eudesmol	1637	0.1
38	epi- α -muurolol	1647	0.1
39	β -eudesmol	1658	1.0
40	selin-11-en-4- α -ol	1668	1.9
41	intermedeol	1667	0.3
42	(2Z, 6Z)-farnesol	1676	0.1
43	(2E, 6Z)-farnesal	1719	0.1
Total			99.6%
Essential oil yield (%)			0.92%
Monoterpenic hydrocarbons			3.2%
Oxygenated monoterpenes			84.8%
Sesquiterpenic hydrocarbons			6.4%
Oxygenated sesquiterpenes			5.2%
t (traces) \leq 0.1% ; RI = Retention Index			

Table 3. Rate of *S. cerealella* death provoked by *D. tripetala* essential oil in fumigation method.

Treatment ($\mu\text{l.ml}^{-1}$)	Mean (\pm SE) mortality of <i>S. cerealella</i>			
	24h	48h	72h	96h
0	0.47 \pm 0.05(1.15)e	0.52 \pm 0.00(1.15)b	0.56 \pm 0.04(2.34)b	0.67 \pm 0.03(3.62)b
0.2	1.43 \pm 0.03(42.67)d	3.14 \pm 0.00(100)a	3.14 \pm 0.00(100)a	3.14 \pm 0.00(100)a
0.5	2.01 \pm 0.04(70.78)c	3.14 \pm 0.00(100)a	3.14 \pm 0.00(100)a	3.14 \pm 0.00(100)a
1	2.80 \pm 0.18(95.50)b	3.14 \pm 0.00(100)a	3.14 \pm 0.00(100)a	3.14 \pm 0.00(100)a
3	3.14 \pm 0.00(100)a	3.14 \pm 0.00(100)a	3.14 \pm 0.00(100)a	3.14 \pm 0.00(100)a
Probability	<0.0001***	<0.0001***	<0.0001***	<0.0001***
CV(%)	7.15	1.42	1.79	1.43

0: ethanol treatment corrected with the control without treatment; *** = very highly significant difference (0.1%). The averages in brackets arose from raw data. The averages followed by the same letter were not significantly different at the beginning of 5% (Newman and Keuls test).

To further assess the effectiveness of the toxicity of these oils, we calculated LC_{50} and LC_{99} . It is clear from this table that the essential oil of *D. tripetala* appears to have a relatively higher efficiency. These results are confirmed by the values of LC_{50} and LC_{99}

obtained from a function of the regression line and which correspond to 0.253 $\mu\text{l.ml}^{-1}$ and 2.685 $\mu\text{l.ml}^{-1}$ (Table 6). Indeed, the LC_{50} is very close to the first dose, so that the LC_{99} is between the second and third doses.

Table 4. Rate of *S. cerealella* death provoked by *U. angustifolium* essential oil in fumigation method.

Treatment ($\mu\text{l.ml}^{-1}$)	Mean (\pm SE) mortality of <i>S. cerealella</i>			
	24h	48h	72h	96h
0	0.47 \pm 0.05(1.15)e	0.52 \pm 0.00(1.15)c	0.56 \pm 0.04(2.34)b	0.67 \pm 0.03(3.62)b
0.2	1.39 \pm 0.04(40.43)d	2.84 \pm 0.15(96.63)b	3.14 \pm 0.00(100)a	3.14 \pm 0.00(100)a
0.5	1.88 \pm 0.06(65.15)c	3.14 \pm 0.00(100)a	3.14 \pm 0.00(100)a	3.14 \pm 0.00(100)a
1	2.36 \pm 0.08(85.39)b	3.14 \pm 0.00(100)a	3.14 \pm 0.00(100)a	3.14 \pm 0.00(100)a
3	2.96 \pm 0.17(97.75)a	3.14 \pm 0.00(100)a	3.14 \pm 0.00(100)a	3.14 \pm 0.00(100)a
Probability	<0.001***	<0.001***	<0.001***	<0.001***
CV(%)	7.97	4.56	1.79	1.43

0: ethanol treatment corrected with the control without treatment; *** = very highly significant difference (0.1%). The averages enter brackets arise raw data. The averages followed by the same letter were not significantly different at the beginning of 5% (Newman and Keuls test).

Table 5. Efficacy of *Dennettia tripetala* or *Uvariadendron angustifolium* essential oil on *Sitotroga cerealella* progeny emergence at 45 days post treatment.

Dose ($\mu\text{l.ml}^{-1}$)	F1 progeny emergence (mean \pm SE)	
	<i>D. tripetala</i>	<i>U. angustifolium</i>
0	2.60 \pm 0.04 (401.33)a	2.60 \pm 0.04 (401.33)a
0.2	0.10 \pm 0.10(0.33)b	0.41 \pm 0.06(1.66)b
0.5	0.00 \pm 0.00(0.00)b	0.00 \pm 0.00(0.00)c
1	0.00 \pm 0.00(0.00)b	0.00 \pm 0.00(0.00)c
3	0.00 \pm 0.00(0.00)b	0.00 \pm 0.00(0.00)c
Probability	<0.001***	<0.001***
CV(%)	9.67	6.38

0: ethanol treatment corrected with the control without treatment; *** = very highly significant difference (0.1%). The averages in brackets arose from raw data. The averages followed by the same letter were not significantly different at the beginning of 5% (Newman and Keuls test).

Effect on the germination of seeds of paddy rice. Fig 1 shows the graph illustrating the percentage of sprouted grains of paddy rice according to two different doses of the essential oils. Both essential oils have greatly reduced adult populations *S.cerealella* by fumigation without affecting the germination of grains of paddy rice processed. The germination rate change from 90% to 100% from the low dose of essential oil while it is less than 80% in the treated only with ethanol. In fact, according to statistical analysis of data there is a highly significant difference ($p < 0.001$) for both treatments as regards the percentages of seed germination after the paddy rice test.

Discussion

Uvariadendron angustifolium, syn. *Uvaria angustifolia* (Annonaceae) is a tree in the forests of West Africa that can reach 15-40 m high (Hutchinson *et al.*, 1954). This species, found in the south of Benin, is used in traditional medicine to treat rheumatism and stomach ache, malaria, or for flavoring local dishes (leaves) (Analytical Flora of Benin, 2006). The essential oils obtained from the leaves of *U. angustifolium* were characterized by a high proportion of oxygenated monoterpenes (84.8%). Essential oils have been mainly dominated by citral (geranial: 44.9% neral and 32.1%). These results are similar with the only chemical study reported by Noudogbessi *et al.* (2014). For essential oil of *D. tripetala*, the present results are different to those obtained by Adeoti *et al.* (2000) and Gbolade *et al.* (2009) on the same plant harvested respectively in Benin and Nigeria. Variability levels recorded could be related to the importance of the secretory cells in the leaves of our sample, their physiology, the place or the harvest period. *Dennettia tripetala* (G.) (Baker f.) GE Schatz (Annonaceae) is a woody spicy vegetable and forest, where the leaves and fruits are used in combination with other herbs for the treatment of cough, infantile convulsion, and worm infestation (Ejechi and Akpomedaye, 2005). *Dennettia tripetala* extracts have also been reported to exhibit insecticidal properties (Egwenyenga *et al.*, 1998) and antifungal (Nwachukwu and Osuji, 2008). The

essential oils obtained from leaves of *D. tripetala* harvested in Benin in this study were characterized by a high proportion of aromatic oxygen compounds (58.2%) and oxygenated monoterpenes (30.1%).

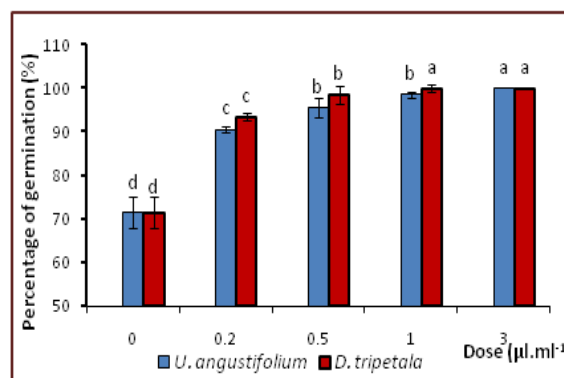


Fig. 1. Percentage of germination of rice according to the essential oil doses: 0: ethanol treatment corrected with the control without treatment. The averages followed by the same letter were not significantly different at the beginning of 5% (Newman and Keuls test).

Regarding the insecticidal activities of two essential oils, our findings corroborate the work of several researchers who have demonstrated the toxicity of essential oils by inhalation or fumigation against stored product pests (Keita *et al.*, 2001; Lee *et al.*, 2001; Kim *et al.*, 2003; Shaaya *et al.*, 1997). The major advantage of fumigation is to facilitate the penetration of gases inside the grain and thus destroy eggs, larvae and pupae that develop (Benayad, 2008). The toxicity of the fumigant components of essential oils of plants against adult insects were significantly ($P < 0.001$) influenced by the dose and time. The cumulative rate of insect mortality was highest 48 hours after the treatment. Levels of fumigant activities observed could be explained by variations in the structure of the complex relationships of insecticidal activity that influenced their degree of penetration into the insect cuticle and neurotoxicity (Ogendo *et al.*, 2010). Insects undergoing treatment with a dose of 0.2 µl.ml⁻¹ showed a small resistance which did not last more than a day since mortality could reach over 90% after the second day. The very low mortality level indicator shows that our test remains reliable for the study of the insecticidal effect of essential oils tested. The insecticidal activity of the

essential oil of *D. tripetala* did not need much time to occur, since the maximum 100% mortality was recorded the first day post treatment with a dose of 3 $\mu\text{L.mL}^{-1}$. In addition, LC_{50} and LC_{99} values (0.253 $\mu\text{L.mL}^{-1}$ and 2.685 $\mu\text{L.mL}^{-1}$) confirm its high toxicity to insects in respect of fumigation method. Indeed, the remarkable presence of high aromatic oxygen compounds (58.2%) and monoterpene oxygenates (30.1%) could explain its pronounced insecticidal effect. Reducing the emergence of F1 progeny in the treated groups could be due to increased adult mortality, ovicidal and larvicidal properties of essential oils confirming the findings of Selase and Getu (2009); Bamaïyi *et al.* (2007); Tapondjou *et al.* (2002). The emergence of high levels recorded in the control plots also confirm the effectiveness of essential oils tested. Note that it does not exist in the literature work on insecticidal activities of essential oil *Uvariadendron angustifolium*. Germination tests showed that the plant materials tested against *S. cerealella* showed no visible adverse effects on the germination capacity of seeds. Also, according to Ketoh *et al.* (2002) and Glitho *et al.* (2008), the presence of residues in treated seeds does not affect their ability to germination. Unlike the current results, Paranagama *et al.* (2003) study showed that the treatment of *C. citratus* oil reduced the germination capacity of paddy compared to non-infested lot. All tests have confirmed that the treatment of food with essential oil of the two aromatic and medicinal plants of Benin can be very effective against the pest of these commodities Regnault-Roger *et al.* (2008); Philogène *et al.* (2008); Vincent *et al.* (2000); Vincent *et al.* (2003); Foua Bi (1993).

Conclusion

The present study assessed the insecticidal properties of essential oils of *Dennettia tripetala* and *Uvariadendron angustifolium* leaves. Volatile extracts from two Annonaceae studied proved to be very effective by way of fumigation and do not alter the germination quality of paddy grains developed. The use of these two essential oils appears to be a promising method for the protection of stored rice

against *Sitotroga cerealella*. However, since the plant products evaporate quickly in the environment and do not persist longer unlike synthetic pesticides, pesticide efficacy of herbs could be enhanced when dissolved or mixed with a material fixer for slow release or carrier such as starch or liquid paraffin, and incorporated as part of the integrated pest management especially at the small-scale farmers.

Acknowledgments

Authors are grateful to Polytechnic School of Abomey-Calavi (Benin) for financial Support. They are also thankful to Professor Paul YEDOMOHAN from National Herbarium of Benin for plants identification.

References

- Abbott WS.** 1925. A method for computing the effectiveness of an insecticide. *Journal of Economic Entomology* **18**, 265-267.
- Adeoti SB, Ayedoun MA, Leclercq PA.** 2000. Essential Oil of *Dennettia tripetala* Leaves from Benin. *Journal of Essential Oil Research* - 01/2000; **12(4)**, 412-414.
<http://dx.doi.org/10.1080/10412905.2000.9699551>
- Adjalien E, Noudogbessi JP, Kossou D, Sohounhloue D.** 2014. État et perspectives de lutte contre *Sitotroga cerealella* (Olivier, 1789), déprédateur des céréales au Bénin: synthèse bibliographique. *Journal of Applied Biosciences* **79**, 6955-6967.
<http://dx.doi.org/10.4314/jab.v79i1.16>.
- Alzouma I, Huignard J, Lenga A.** 1994. Les coléoptères Bruchidae et les autres insectes ravageurs des légumineuses alimentaires en zone tropicale. In: Post-récolte, principes et application en zone tropicale. ESTEM/AUPELF Verstraeten *et al.* Eds. 79-103 p.
- Aringbangba RO.** 2011. Toxicity of extract from *Eugenia aromatica* (L.) on *Sitotroga cerealella* (olivier) (lepidoptera: gelechiidae) infesting different varieties

of paddy rice. A thesis submitted to the school of post graduate studies in partial fulfilment of the requirements for the award of master of technology degree in food storage technology. Department of Biology, Federal University of Technology, Akure. Pages 91.

Bamaiyi LJ, Ndams IS, Toro WA, and Odekina S. 2007. Laboratory evaluation of mahogany (*Khaya senegalensis*(Desv.) seed oil and seed powder for the control of *Callosobruchus maculatus* (Fab.) (Coleoptera: Bruchidae) on stored cowpea, Journal of Entomology **4**, 237-242.

<http://dx.doi.org/10.3923/je.2007.237.242>.

Bekele J, Hasanali A. 2001. Blend effects in the toxicity of the essential oil constituents of *Ocimum Kilimands* and *Ocimum kenyense* (Labiatae) on two post-harvest insects pests. Phytochemistry **57**, 385-391.

Bell A. 1994. Emploi des substances végétales comme produits de protection des stocks contre le grand capucin du grain (*Prostephanus truncatus*) et autres ravageurs. GTZ, Eschborn, Allemagne.

Benayad N. 2008. Les huiles essentielles extraites des plantes médicinales marocaines: Moyen efficace de lutte contre les ravageurs des denrées alimentaires stockées. Thèse de doctorat, Université Mohammed V – Agdal Faculté des Sciences de Rabat.

Dagnelie P. 1975. Théorie et Méthodes statistiques. Applications agronomiques. Les Presses Agronomiques de Gembloux A.S.B.L. Avenue de la faculté, 22-5800 Gembloux (Belgique).

Don-Pedro KN. 1996. Fumigant toxicity of citrus peel oils against adult and immature stages of storage insect pests. Pesticide Science **47**, 213–223.

Egwunyenga OA, Alo EB, Nmorsi PG. 1998. Laboratory evaluation of the repellency of *Dennettia tripetala* Baker (Annonaceae) to *Dermestes maculatus* (F.) (Coleoptera: Dermestidae). Journal of Stored

Products Research **34**, 195-199.

Ejechi BO, Akpomedaye DE. 2005. Activity of essential oil and phenolic acid extracts of pepper fruit (*Dennettia tripetala* G. Baker) against some food-borne microorganisms. African Journal of Biotechnology **4**, 258-261.

Flore Analytique du Bénin 2006. A Akoègninou, WJ Van Der Burg, LLG Van Der Maessen (Eds.), Backhuys Publishers, 1034 p.

Foua-Bi K. 1993. Produits naturels utilisés dans la protection des stocks en Afrique noire. In Thiam, A., Ducommun, G. Eds : Protection naturelle des végétaux en Afrique. Enda, Dakar, 85-100 o.

Gbolade AA, Arcoraci T, D'Arrigo M, Olorunmola FO, Biondi DM, Ruberto G. 2009. Essential oils of *Dennettia tripetala* Bak. f. stem bark and leaf. Constituents and biological activities. Planta Medica; **75** – I32 p.

<http://dx.doi.org/10.1055/s-0029-1234796>

Glitho AI. 2002. Post-récolte et biopesticides en Afrique, Annexe. In: Biopesticides d'origine végétale. Regnault Roger C., Philogène B.J.R. & Vincent C. Eds. Paris, 313-321.

Glitho IA, Ketoh KG, Nuto PY, Amevoin SK, Huignard L. 2008. Approches non toxiques et non polluantes pour le contrôle des populations d'insectes nuisibles en Afrique du Centre et de l'Ouest. 207-217. In Regnault-Roger, C, Philogène, B.J.R. et Vincent, C (Eds). Biopesticide d'origine Végétale 2 édition. Lavoisier, TEC & DOC, Paris, 550 p.

Hassanali A, Lwande W, Sitayo O, Moreaka L, Nokoe S, Chapaya A. 1990. Weevil repellent constituents of *Ocimum kilimandscharicum* (Labiatae) as post harvest protectant against infestation of three major stored product insect pests. Bulletin of Entomological Research **85**, 361-367.

Ho SH, Koh L, Ma Y, Huang Y, Sim KY. 1996.

The oil of the garlic *Alium sativum* L. (Amaryllidaceae) as a potential grain protectant against *Tribolium castaneum* (Herbst) and *Sitophilus zeamais* Motsch. Post harvest Biology and Technology **4**, 179-183.

Hutchinson J, Dalziel JM. 1954. Flora of West Tropical Africa. 2nd ed. London: Crown Agents.

Jirovetz L, Buchbauer G, Ngassoum MB, Geissler M. 2002. Aroma compound analysis of *Piper nigrum* and *Piper guineense* essential oils from Cameroon using solid-phase micro-extraction-gas chromatography, solid-phase microextraction-gas chromatography-mass spectrometry and olfactometry. Journal of Chromatography **976**, 1-2, 265-275.

Kéita SM, Vincent C, Schmit JP, Arnason JT, Bélanger 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). Journal of Stored Product research **37**, 339-349.

Ketoh GK, Glitho IA, Huignard J. 2002. Susceptibility of the bruchid *Callosobruchus maculatus* (Coleoptera: Bruchidae) and its parasitoid *Dinarmus basalis* (Hymenoptera: Pteromalidae) to three essential oils. Journal of Economic Entomology, **95**(1), 174-182

Kim S, Roh L, Kim D, Lee H, Ahn Y. 2003. «Insecticidal activities of aromatic plant extract and essential oils against *Sitophilus oryzae* Land *Callosobruchus chinensis* Fab». Journal of Stored Products Research **39**, 293-303.

Kossou KD, Aho N. 1993. Stockage et conservation des grains alimentaires tropicaux. Principes et pratiques. Flamboyant Ed. Cotonou, 125 p.

Lee SE, Lee BH, Choi WS, Park BS, Kim JG, Campbell BC. 2001. Fumigant toxicity of volatile

natural products from Korean spices and medicinal plants towards the rice weevil *Sitophilus oryzae* L. Pest Management Science **57**(6), 548-553.

Noudogbessi JP, Gary-Bobo M, Adomou A, Adjalian E, Alitonou GA, Avlessi F, Garcia M, Sohounhloue DC, Menut C. 2014. Comparative chemical study and cytotoxic activity of *Uvariadendron angustifolium* essential oils from Benin. Natural product communications **02**/2014; **9**(2), 261-4.

Nwachukwu EO, Osuji JO. 2008. Evaluation of plant extracts for antifungal activity against *Sclerotium rolfsii* causing cocoyam cormel rot in storage. Research Journal of Agricultural and Biological Sciences **4**, 784-787

Ogendo JO, Deng AL, Belmain SR, Walker DJ, Musandu AO, Obura RK. 2004. Pest status of *Sitophilus zeamais* Motschulsky, control methods and constraints to safe maize grain storage in Western Kenya, Egerton. Journal of Science and Technology **5**, 175-193

Ogendo JO, Deng AL, Kostyukovsky M, Ravid U, Matasyoh JC, Omolo EO, Kariuki ST, Bett PK, Kamau EAW. 2010. Fumigant toxicity of five essential oil constituents against major stored-product insect pests of food grains. Second RUFORUM Biennial Meeting 20 - 24 September 2010, Entebbe, Uganda Research Application Summary.

Paranagama P, Abeysekera T, Nugaliyadde L, Abeywickrama K. 2003. Effects of the essential oils of *Cymbopogon citratus*, *C. nardus* and *Cinnamomum zeylancium* on pest incidence and grain quality of rough rice (paddy) stored in an enclosed seed box, Journal of Food Agriculture and Environment **134**, 134-136

Philogène BJR, Regnault-Roger C, Vincent C. 2008. Biopesticides d'origine végétale: bilan et perspectives, I-24P. In Regnault-Roger, c., Philogène,

B.JR., Vincent, C. (éds) *Biopesticides d'origine végétale*, 2^{ème} éd., Lavoisier, Paris.

Regnault-Roger C. 2002. De nouveaux phyto-insecticides pour le troisième millénaire. In : Philogène B.J.R, Ragnault-Roger C. et Vincent C., coord. *Biopesticide d'origine végétale*. Paris : Lavoisier-Editions Tec et Doc, 19-39 p.

Regnault-Roger C, Philogène, BJR, Vincent C. 2008. *Biopesticides d'origine végétale*, 2^{ème} édition, Lavoisier, Paris. édition, 550p.

Rösch P, Popp J, Kiefer W. 1999. Raman, Investigations on Lamiaceae Plants, *Journal of Molecular Structure* **121**, 480 – 481.

Selase AG, Getu E. 2009. Evaluation of botanical plants powders against *Zabrotes subfasciatus* (Boheman) (Coleoptera: Bruchidae) in stored haricot beans under laboratory condition, *African Journal of Agricultural Research* **4**, 1073-1079

Shaya E, Kostjukovski M, Eilberg J, Sukprakarn C. 1997. Plant oil as fumigant and contact insecticides for the control of stored product insects. *Journal of Stored Products Research* **33**, 7-15.

Tapondjou LA, Adler C, Bouda H, Fontem DA. 2002. Efficacy of powder and essential oil from *Chenopodium ambrosioides* leaves as post-harvest grain protectants against six-stored product beetles, *Journal of Stored Products Research* **38(4)**, 395-402.

[http://dx.doi.org/10.1016/S0022-474X\(01\)00044-3](http://dx.doi.org/10.1016/S0022-474X(01)00044-3)

Togola A, Nwilene F, Chougourou CD, Agunbiade TA. 2010. Présence, population et dégâts de l'alucite des céréales *Sitotroga cerealella* sur les stocks de riz au Bénin 63 p.

Vincent C, Panneton B, Fleurat-Lessard F. 2000. La lutte physique en phytoprotection. INRA, Paris. 347 141 p.

Vincent C, Hallman G, Panneton B, Fleurat-Lessard F. 2003. Management of Agricultural Insects with Physical Control Methods». *Annual Review of Entomology* **48**, 261 281.