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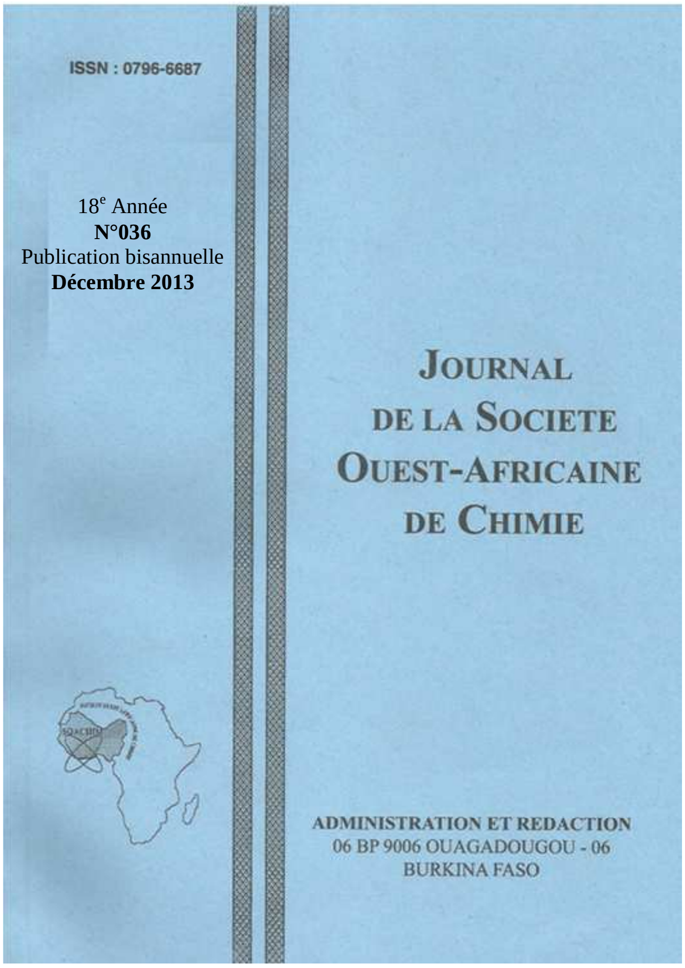

Curative and persistent activities of essential oils extracted from two varieties leaves of *Tephrosia* species from Benin on *Callosobruchus maculatus*

Jean-Pierre Noudogbessi[1], Fidèle Paul Tchobo[1], Théophile Odjo[2], Elvis Adjalian[1], Léonce Dovonon[1], Gbêdossou Sophie Bogninou-Agbidinoukoun[1], Gilles Figueredo[3], Félicien Avlessi[1], Kossou Dansou[2], Jean-Claude Chalchat[4], Dominique Codjo Koko Sohounhloué[1*]

[1] *Laboratoire d'Etude et de Recherche en Chimie appliquée (LERCA), Ecole Polytechnique d'Abomey-Calavi, Université d'Abomey-Calavi 01 BP 2009 Cotonou, Rép. du Bénin*
[2] *Département de Production Végétale ; Faculté des Sciences Agronomiques /Université d'Abomey-Calavi ; 01 BP 526 Cotonou Rép. du Bénin*
[3] *Laboratoire d'Analyse des Extraits Végétaux et des Arômes (LEXVA Analytique)*
[4] *Laboratoire de Chimie des Huiles Essentielles, Université Blaise-Pascal, Clermont-Ferrand II, Campus des Cézeaux, 63177 Aubière cedex, France*

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Abstract : The *T. vogelii* essential oil was dominated by high levels (≥ 5.0%) of caryophyllene oxide (5.0%), *cis*-calamenene (6.0%), epi-α-bisabolol (8.3%), γ-eudesmol (12.8%), α-muurolol (13.7%) and germacrene-B (16.5%). On the other hand the volatile extract from *T. densiflora* leaves was remarkably dominated by caryophyllene oxide (63.5%) followed by humulene epoxide II (5.0%). The ovicidal, larvicidal and insecticidal performances of both essential oils, at concentrations 0, 5, 10, 15 and 20 µL/mL, were evaluated by testing their curative and persistent effects on *C. maculatus* (Fabricius) living on cowpea (Temperatures: between 22 and 31°C; Relative Humidity: 80%) of local variety "Chawe". The damage caused by *C. maculatus* and its larvae on the cowpea quality in determining the percentages of attacked seeds (A) and weight loss (B) of these did not allow to conclude an obvious use of these plants as biopesticides post-harvests.

Key words: *Tephrosia*, caryophyllene oxide, germacrene-B, biopesticides, *C. maculatus*

Résumé : l'huile essentielle de *Tephrosia vogelei* est dominée par des taux élevés (≥ 5,0 %) d'oxyde de caryophyllène (5,0 %), cis-calamenène (6,0 %), épi-α-bisabolol (8,3 %), γ-eudesmol (12,8 %), α-muurolol (13,7 %) et germacrène-B (16,5 %). Par ailleurs, l'extrait volatil des feuilles de *T.densiflora* est remarquablement dominé par l'oxyde de caryophyllène (63,5 %) suivi de l'époxyde II de Humulène (5,0 %). Les performances ovicide, larvicide et insecticide des deux huiles essentielles aux concentrations 0,5,10,15 et 20 µl/ml sont évaluées en testant leurs effets curatifs et persistants sur *C.maculatus* (Fabricius) séjournant sur une variété locale de niébé ''Chawé''(température entre 22 et 31 °C ; humidité relative 80 %). Les dommages causés par *C.maculatus* et ses larves sur la qualité du niébé dans la détermination des pourcentages des graines attaquées (A) et de la perte de poids (B) de celles-ci ne permettent pas de conclure à une utilisation évidente de ces plantes comme des bio pesticides post-récoltes.

Mots clés : *Tephrosia*, oxyde de caryophyllène, germacrène-B, bio pesticides, *C.maculatus*.

[*] **Corresponding Author** : dominique.sohounhloue@uac.bj

1. Introduction

The foodstuffs are very precious products for which the storage conditions for the welfare of human health and his environment are the object of several studies. In the African countries situated in the South of Sahara and particularly in Benin, the cowpea is one of the main food legumes [1]. With a protein content around 25%, it appears as one of the cheapest sources of proteins for the deprived populations. In West Africa where more than 70% of this foodstuff is produced, the cowpea has gradually become an integral part of farming systems [2]. However, in farms situated in the rural zones of West Africa including those of Benin, the farmers continue to feel enormous difficulties for the protection of cowpea seeds after harvest. In this respect, several strategies implemented to improve the yields through the use of synthetic pesticides and insecticides are usually failed in the post-harvest by *Callosobrocus maculatus* which is indeed the most devastating pest of cowpea stocks (*Vigna unguiculata* L.) in tropical Africa. It grows at the expense of the pods and seeds of cowpea (*Vigna unguiculata*) [3, 4]. The pods infestations by this insect begin in the cultures in the beginning of the plant fruiting. The damage which it causes can reach 100 % in a few months [5]. Moreover, the presence in foodstuffs stored by toxic residues and the appearance of resistant strains of *C. maculatus* to pesticides and to chemical insecticides are becoming increasingly worrying and appealing to alternative solutions. In tropical Africa and particularly in Benin, there are biodegradable aromatic plants (leaves, flowers, fruits or seeds) possessing insecticidal properties whose populations are traditionally used to protect crops in storage and for other medical needs [6] since the ancient times. These plant species are a potential reserve of natural biopesticides for the benefit of agriculture [7]. Today, farmers make recourse to the herbal medicine by the use of these available and plentiful plants in our countrysides, producers less expensive essential and generating no consequence and no major impacts on human health, the biodiversity and the environment. The essential oils have been the object of important works, especially those of Delobel and Malonga in Congo [8], Kayitare and Ntezurubanza in Rwanda [9] or Kétoh in Togo [10]. Other investigations led in Uganda [11] and Zimbabwe [12] brought back the acaricidal properties of *T. vogelii* extracts. This study which joins whithin the framework of the protection of the stocks of cowpea under its two forms (seed in pod or not), evaluates on *C. maculatus* the curative and persistent effects of essential oils extracted from two aromatic plants of Leguminosae-Papilionoideae family.

2. Experimental

2.1. Plant material

T. vogelii (Hook. f.) and *T. densiflora* (Hook. f.) leaves were collected in 2008 to Kpedekpo in Zou Department of Benin. They were identified and certified at the Abomey-Calavi University National Herbarium then stored in the laboratory between 18 and 20°C in the shade of the sunlight during all the extraction period. Pods of cowpea, having the same size (approximately 10 cm) and containing each one on average eleven grains were selected to Dannou Tokpli (Benin). In the laboratory, these pods were preserved at 2°C to get rid of any form of infestations known from the fields. After this disinfestation period (5 days), the cowpea seeds, well dried in the shade (29°C) for 72 h, were arranged under two forms namely: with pod and without pod.

2.2. Test organisms

Callosobruchus maculatus (Fabricius) adults were selected with the support of a postharvest insects specialist of a peasant reserve of cowpea to Dannou Tokpli. In the laboratory, they were put in breeding in the optimal conditions of reproduction and obtaining new generations (T: 27.5 ± 0.2°C, RH: 80.3 ± 1.6%).

2.3. Phytochemical analysis

The essential oils were obtained by water distillation of the leaves and fruit (300 g) for 3 hours in a Clevenger according to the method used in britanic pharmacopoeia [13]. They were dried over anhydrous sodium sulphate and analysed by GC/MS.

GC/MS: The essential oils were analysed on a Hewlett-Packard gas chromatograph Model 7890, coupled to a Hewlett-Packad MS model 5875, equipped whith a DB5 MS column (30m X 0.25mm; 0.25µm), programming from 50°C (5 min) to 300°C at 5°C/mn, 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, 2500 V; 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 analysed on a Hewlett-Packard gas chromatograph Model 6890, equipped whith a DB5 MS column (30m X 0.25mm; 0.25µm), programming from 50°C (5 min) to 300°C at 5°C/mn, 5 min hold. Hydrogen 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 [14 - 16].

2.4. Insecticidal, larvicidal and ovicidal evaluations

The following tests were realized according to the methodology described by Noudogbessi and *al.* in 2009 [17]. They are, on the whole, arranged in a device of complete random block per mode of application with five (05) repetitions.

2.5. Curative effect

The temperature of the experimental framework varied between 22 °C and 31 °C and relative humidity was fixed at 80%. The adults insects (two females and one male) of the same age (0-48 hours), taken at random from the mass rearing were filed according to the infestation mode on the plant material (cowpea seeds in pod or without pod) contained in glass jars. A total of $5 \times 3 \times 2 = 30$ experimental units were established. The assembly thus prepared was placed in observation at regular time intervals (24 h) for the follow-up of the oviposition of females. During the observation period, an insect that died was replaced immediately and recorded. After six (6) days, all insects (female and male) were recovered and only the cowpea so infested was immediately treated with essential oil. Five concentrations (0, 5, 10, 15, 20 µL/mL) selected according to the chemical profile of each essential oil dissolved in absolute ethanol were tested. The glass jar containing the cowpea sample untreated with essential oil has also been infested by *C. maculatus* adult (control). The experimental units were afterward observed for 20 days to follow in regular time interval (24 h), the rhythm of young insects emergence. During this period, there was a strict monitoring of the various manifestations of the appearance of young insects at the level of the substratum and the following parameters were collected:

- Number of insects dead and recorded during six days;
- Number of emergent insects of the various forms of cowpea infested and recorded every day during 20 days;
- Number of the seeds infested and the weight loss of cowpea.

2.6. Persistent effect

The persistence was evaluated by infesting both forms of the cowpea treated with essential oil 15 days ago. On the substrata beforehand treated, the insects (two females and one male) were deposited and put in observation. The parameters measured during this essay were:

- Number of insects dead at the end of 16 days;
- Number of emergent insects at the end of 20 days;
- Number of the seeds infested and the weight loss of cowpea.

2.7. Estimated losses after treatment

To better assess *C. maculatus* damages, the cowpea seeds were recovered, manually counted and weighed. The rate of the weight loss of the seeds was determined according to the method of counting and weighing (MCP). Two criteria were used for assessing the *C. maculatus* damages on the cowpea seeds: the seeds attack percentage (A%) and the rate in weight loss (B%).

$$A\% = \frac{N_a}{N_a + N_s} \times 100$$

and

$$B\% = \frac{P_s N_a - P_a N_s}{N_a + N_s} \times 100$$

Na = number of attacked grains;
Ns = number of healthy grains;
Pa = weight of damaged grains;
Ps = weight of healthy grains

2.8. Statistical analysis

The results from the observations were treated statistically by analysis of variance method (ANOVA) using S A S software (Statistical Analysis System) Version 9.1 [19]. The raw data underwent the following transformations: Arcsin ($\sqrt{X/n}$), X being the number of insects died under the influence of the essential oil and n indicating the total number of insects introduced into each bottle.

$\sqrt{X+0,5}$ (X is the number of young *C. maculatus* having emerged from the substrate). The masses of grains affected being continuous quantitative data and respecting the conditions of normalization and equal variance were no statistical transformation. Finally, it was preceded to a structuralization of the averages by means of the test of Newman and Keuls [19]. The results of statistical tests were considered significantly different when the probability of the null hypothesis was less than or equal to 5%.

3. Results and discussion

The essential oils studied were characterized by very low yields. The values of these yields were 0.052% for *T. vogelii* and 0.038% for *T. densiflora*. They were mainly rich in sesquiterpenes hydrocarbons (18.5 - 37.7%) and oxygenated sesquiterpenes (50.7 - 69.6%). *T. vogelii* essential oil contains 32 compounds representing 98% of the weight of this volatile extract. In that extracted from *T. densiflora* leaves, 31 compounds corresponding to 94.1 % of the weight essential oil have been identified. Volatile extract of *T. vogelii* was rich in germacrene-B (16.5%), α-muurolol (13.7%), γ-eudesmol (12.8%), epi-α-bisabolol (8.3%), *cis*-calamenene (6.0%), caryophyllene oxide (5.0%), β-elemene (4.8%) and β-pinene (3.2%). That of *T. densiflora* was mainly composed of caryophyllene oxide (63.5%), humulene epoxide II (5.0%) and β-caryophyllene (4.9%) (**table I**). The major compounds of the samples studied during the current works really differ in proportion to those works published by Noudogbessi *and al.* [20]. Compared with the *T. densiflora* essential oil studied in the current works, it was reported a high rate of caryophyllene oxide (63.9%) accompanied by humulene epoxide II (12.6%) in *Tephrosia cinerea* Pers. leaves harvested in Brazil [21].

The mortality average provoked by the influence of different concentrations (0 to 20 µL/mL) of the two essential oils tested on *C. maculatus* adults, the larvae newly formed, the rate of seeds attacked and the corresponding losses of weight were shown in **Tables II to V**. In curative method, the mortality rates recorded before the cowpea seeds treatments were low and statistically identical at the level of each form conservation.

In the **Table II**, it was presented the evolution of mortality, emergence, number of seeds attacked and weight loss caused by *C. maculatus* in presence of *T. vogelii* essential oil.

In the **Table II**, the values recorded showed after statistical analysis similar and important of emergence rates of young *C. maculatus* at the concentrations ≥ 5 µL/mL for both forms of cowpea conservation. These emergences, low compared with that of control (0 µL/mL) showed that *T. vogelii* essential oil would have demonstrated by means the compounds it contained ovicidal or larvicidal slightly effect so delaying the flow of young insects appearance. It results from these observations that the rates of seeds attacked (A) and the weight loss proportions were not statistically significant at the level of both forms of cowpea conservation.

Table I : Chemical composition of the of *T. vogelii* and *T. densiflora* leaves essential oils

Compounds	RI_{exp}	RI_{th}	*Tv*	*Td*
			(%)	
α-pinene	936	932	2.0	0.2
sabinene	973	969	1.2	0.5
β-pinene	**978**	**974**	**3.2**	1.0
limonene	1031	1024	1.4	-
linalool	1100	1095	0.2	0.3
α-terpineol	1193	1186	0.5	0.5
δ-elemene	1338	1335	1.0	1.3
α-copaene	1379	1374	-	0.3
β-bourbonene	1391	1387	-	1.1
β-elemene	**1394**	**1389**	**4.8**	1.5
α-*cis*-bergamotene	1414	1411	1.3	1.1
β-caryophyllene	**1423**	**1417**	0.1	**4.9**
β-copaene	1432	1430	1.0	0.4
α-*trans*-bergamotene	1435	1432	0.3	0.7
(Z)-β-farnesene	1442	1440	0.5	2.0
α-humulene	1454	1452	1.3	0.5
germacrene-D	1487	1484	1.1	0.1
α-zingiberene	1496	1493	1.6	1.0
α-bulnesene	1518	1509	0.5	0.3
γ-cadinene	1521	1513	-	0.5
δ-cadinene	1528	1522	1.7	0.9
***cis*-calamenene**	**1534**	**1528**	**6.0**	0.4
elemol	1551	1548	2.7	1.4
germacrene-B	**1562**	**1559**	**16.5**	1.5
(E)-nerolidol	1564	1561	1.1	1.9
caryophyllene oxide	**1588**	**1582**	**5.0**	**63.5**
5-epi-7-epi-α-eudesmol	1612	1607	1.8	1.2
humulene epoxide II	**1614**	**1608**	1.4	**5.0**
γ-eudesmol	**1632**	**1630**	**12.8**	-
β-acorenol	1637	1636	2.7	-
α-muurolol	**1647**	**1644**	**13.7**	1.6
epi-α-bisabolol	**1684**	**1683**	**8.3**	-
(Z)-*trans*-α-bergamotol	1693	1690	1.2	-
hexadecan-1-ol	1876	1874	0.1	0.1
phytol	1943	1942	1.0	2.2
isophytol	1948	1946	-	1.2
Monoterpene hydrocarbons			**7.8**	**1.7**
Oxygenated monoterpenes			**0.7**	**0.8**
Sesquiterpene hydrocarbons			**37.7**	**18.5**
Oxygenated sesquiterpenes			**50.7**	**69.6**
Aliphatic alcohols			**0.1**	**0.1**
Diterpenic compounds			**1.0**	**3.4**
Total			**98.0**	**94.1**

Tv = *Tephrosia vogelii*, **Td** = *Tephrosia densiflora*,
RI = Retention index, **exp** = experimental, **th** = theoric

Table II : Rate of *C. maculatus* dead, of emergence, of number of cowpea attacked (A) and of weight loss (B) provoked by *T. vogelii* essential oil in curative method.

Doses (µL.mL^{-1})	cowpea grain (without pod)			
	Dead	**Emerged**	**A**	**B**
0	0.11±0.00(17.01)b	9.10±0.11(86.22)a	0.81±0.20(69.38)a	3.50±0.01(+0.21)a
5	0.90±0.02(23.00)b	7.81±0.22(51.01)b	0.73±0.22(65.02)a	3.99±0.10(+0.11)a
10	0.15±0.04(25.33)b	7.86±0.99(55.33)b	0.77±0.01(68.11)a	3.01±0.40(-1.01)a
15	0.11±0.02(48.15)b	7.87±0.73(55.00)b	0.69±0.10(50.01)a	1.81±0.01(-6.03)a
20	0.14±0.03(55.55)b	7.08±0.50(54.31)b	0.75±0.44(69.01)a	1.90±0.01(-0.21)a
Probability	0.0073ns	0.871**	0.9991ns	0.9013ns
CV (%)	10.03	19.23	22.22	12.01
Doses (µL.mL^{-1})	**cowpea grain (in pod)**			
	Dead	**Emerged**	**A**	**B**
0	0.03±0.00(0.62)a	8.41±0.11(67.99)a	0.69±0.12(49.03)b	4.01±0.00(+1.11)a
5	0.09±0.11(01.21)a	8.22±0.01(16.01)b	0.71±0.00(21.00)b	3.98±0.10(+0.02)a
10	0.03±0.01(08.43)a	5.20±0.33(15.17)b	0.66±0.10(47.22)b	4.00±0.00(+0.01)a
15	0.05±0.33(04.17)a	5.03±1.06(14.99)b	0.64±0.01(15.88)b	3.70±0.01(+0.03)a
20	0.05±0.18(06.00)a	4.13±1.21(17.00)b	0.33±0.33(10.12)bc	3.80±0.01(-1.05)a
Probability	0.1303ns	< 0.0001***	0.0007*	0.1614ns
CV (%)	29.11	41.00	38.71	18.61

ns = not significant at 5%; *** = very highly significant difference (0.1%); * = very highly significant difference (5%). 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 III : Rate of *C. maculatus* dead, of emergence, of number of cowpea attacked (A) and of weight loss (B) provoked by *T. densiflora* essential oil in curative method

Doses (µL.mL^{-1})	cowpea grain (without pod)			
	Dead	**Emerged**	**A**	**B**
0	0.15±0.10(01.05)a	8.10±0.11(06.01)b	0.56±0.41(7.09)a	3.09±0.11(-0.10)a
5	0.09±0.12(18.09)a	8.00±0.31(33.44)b	0.56±0.10(6.41)a	2.83±0.10(-0.10)a
10	0.10±0.12(26.03)a	7.94±0.39(14.21)b	0.48±0.23(4.13)b	3.00±0.00(0.10)a
15	0.07±0.55(27.01)a	6.98±0.33(43.01)c	0.50±0.02(0.10)b	1.42±0.99(-0.01)a
20	0.16±0.22(18.17)a	7.02±0.00(49.01)c	0.32±0.66(10.31)c	1.61±0.08(-0.22)a
Probability	0. 9050ns	< 0.1041 **	0.1002***	0.8011ns
CV (%)	12.01	31.75	31.47	29.12
Doses (µL.mL^{-1})	**cowpea grain (in pod)**			
	Dead	**Emerged**	**A**	**B**
0	0.08±0.05(0.01)a	10.25±0.01(13.95)a	0.81±0.33(25.32)a	3.01±0.15(-0.20)a
5	0.03±0.00(11.47)a	10.04±0.11(19.99)a	0.75±0.17(16.18)a	2.98±0.05(-0.01)a
10	0.00±0.88(10.01)a	9.89±0.00(49.32)a	0.75±0.09(06.23)a	2.78±0.22(-0.22)a
15	0.03±0.89(12.82)a	9.61±0.88(12.99)a	0.67±0.78(11.02)b	2.51±0.33(0.54)a
20	0.05±0.00(09.00)a	9.33±0.27(25.30)a	0.41±0.99(14.01)b	1.99±0.55(-0.06)a
Probability	0.9002ns	0.9991ns	0.1102***	0.0003ns
CV (%)	11.90	42.29	91. 06	55. 18

ns = not significant at 5%; *** = 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)

The results in **Table III** showed the activity of *T. densiflora* essential oil applied to different concentrations in curative method for cowpea conservation. At the all concentrations, the averages of emergence were important. At the level of the seeds without pod, the ovicidal or larvicidal character of the volatile extract was observed only from 15 µL/mL generating emergence averages statistically identical until 20 µL/mL. However, they were different from that of the control (0 µL/mL). Moreover, in the case of the seeds with pods, the emergence rates recorded were important and were not significantly different. The eggs deposited by *C. maculatus* adult before the seeds treatment with *T. densiflora* essential oil and the newly formed larvae have been certainly developed inside the pods preventing the essential oil manifestation. In this way, the development stages of eggs and larvae have escaped the possible repressive effects of the volatile extract used. The rates of seeds attacked (A) (in pod or not) decreased progressively as the concentrations of essential oil increased. At 10, 15 and 20 µl/ml, the values of (A) were slightly lower contrary to that of the control. This slight decrease of the rates of seeds attacked would result from the essential oil effect which would have reduced the young *C. maculatus* performance just after their emergence. Overall, in both forms preserved, the weight loss was not statistically significant. The **Table IV** summarizes the results of the evaluation of two preservation modes of cowpea seeds in presence of the essential oil extracted from *T. vogelii* leaves persistent method.

Table IV : Rate of *C. maculatus* dead, of emergence, of number of cowpea attacked (A) and of weight loss (B) provoked by *T. vogelii* essential oil in persistent method

Doses (µL.mL^{-1})	**cowpea grain (without pod)**			
	Dead	**Emerged**	**A**	**B**
0	0.07±0.02(1.11)a	9.11±0.14(48.01)a	1.09±0.07(2.03)a	3.99±0.00(+13.33) a
5	0.08±0.88(6.04)a	8.88±0.21(69.01)a	1.21±0.00(2.43)a	3.99±0.10(+12.19) a
10	0.08±0.01(5.01)a	8.76±0.00(66.21)a	1.11±0.23(2.32)a	2.99±0.13(-9.21)a
15	0.06±0.88(3.82)a	8.07±0.25(65.01)a	1.02±0.02(1.99)a	2.84±0.01(-9.03)a
20	0.07±0.00(4.09)a	7.88±0.61(51.83)a	0.99±0.99(1.01)a	2.95±0.00(-1.62)a
Probability	0.0008ns	0.1111ns	0.0033ns	0.2218 ns
CV (%)	56.01	91.01	14.14	43.33
Doses (µL.mL^{-1})	**cowpea grain (in pod)**			
	Dead	**Emerged**	**A**	**B**
0	0.00±0.00(0.00)a	11.02±0.00(122.99)a	1.09±0.10(2.05)b	2.94±0.10(+8.91)a
5	0.00±0.00(0.00)a	10.30±0.33(103.22)a	1.01±0.60(2.01)b	2.88±0.22(-8.11)a
10	0.00±0.00(0.00)a	10.23±0.54(101.10)a	1.01±0.02(41.99)b	2.43±0.99(+7.09)a
15	0.05±0.01(03.11)a	09.93±1.31(99.24)a	0.98±0.00(1.87)b	2.65±0.12(+7.95)a
20	0.03±0.11(10.01)a	10.10±0.09(101.03)a	0.99±0.00(1.03)b	2.55±0.43(+7.69)a
Probability	0.0301ns	0.9999ns	0.2929ns	0.8884ns
CV (%)	71.22	14.03	67.11	65.43
ns = not significant at 5%; 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 V : Rate of *C. maculatus* dead, of emergence, of number of cowpea attacked (A) and of weight loss (B) provoked by *densiflora* essential oil in persistent method

Doses (µL.mL^{-1})	**cowpea grain (without pod)**			
	Dead	**Emerged**	**A**	**B**
0	0.03±0.00(0.00)a	10.09±0.11(11.00)b	1.17±0.07(1.98)a	2.11±0.14(+4.18)a
5	0.01±0.00(0.00)a	10.01±0.99(0.19)b	1.08±0.02(1.32)a	2.09±0.00(+4.01)a
10	0.05±0.00(0.00)a	10.01±0.02(20.13)b	1.03±0.00(1.10)a	1.99±0.00(-3.95)a
15	0.01±0.00(0.01)a	9.89±0.99(51.05)b	0.99±0.66(1.08)a	1.81±0.11(-3.03)a
20	0.04±0.01(1.66)a	9.68±0.00(49.01)b	0.90±0.09(0.89)a	1.09±0.21(+1.21)b
Probability	0.8301ns	0.4001ns	0.3291ns	0.3301**
CV (%)	10.03	57.30	22.22	30.57
Doses (µL.mL^{-1})	**cowpea grain (in pod)**			
	Dead	**Emerged**	**A**	**B**
0	0.00±0.00(0.00)a	9.36±0.01(9.20)a	1.25±0.78(1.58)a	2.00±0.00(+4.01)a
5	0.00±0.00(0.00)a	9.19±0.77(55.78)a	1.18±0.44(1.49)a	1.99±0.790(+3.97)a
10	0.00±0.00(0.00)a	9.13±0.00(63.78)a	0.93±0.09(0.88)a	2.66±0.02(-2.75)a
15	0.01±0.00(0.00)a	8.89±0.01(81.18)a	0.71±0.72(0.68)a	1.81±0.99(+1.78)a
20	0.00±0.00(0.00)a	9.00±0.10(89.01)a	0.71±0.09(0.55)a	1.76±0.0.98(+1.00)a
Probability	0.5901ns	0.6711ns	0.3701ns	0.7371ns
CV (%)	43.69	91.01	44.44	19.43
ns = not significant at 5%. 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)				

The emergence averages noted, the rates of seeds attacked and the weight loss were not statistically significant in both cases of conservation evaluated for cowpea (in pod or not). We noted through these results that *T. vogelii* essential oil had no action in persistent method on the control of the flow of emergence of young *C. maculatus*, its eggs and its larvae. The essential molecules of this volatile extract would probably have no long-term persistent effect which can act as insecticide for the benefit of the crop conservation in storage.

The emergence rates obtained at the tested concentrations were statistically identical to that of the control indicating an absence of effects being able to emanate from the molecules of *T. densiflora* essential oil on *C. maculatus* living on cowpea (in pod or not). The inactivity observed would result from antagonist effects which would inhibit the insecticidal, larvicidal and ovicidal properties of some compounds of the *T. densiflora* essential oil unlike that of *T. cenerea* of which the effectiveness has been proven on *C. maculatus* larvae in Brazil [21].

4. Conclusion

T. vogelii and *T. densiflora* are two aromatic plants whose essential oils analyzed by GC/MS were potentially rich in sesquiterpenoïc compounds. Caryophyllene oxide was the dominant compound of the essential oil extracted from *T. vogelii* leaves whereas that of *T. densiflora* contained mostly germacrene-B, α-muurolol and γ-eudesmol. This work allowed highlighting the ineffectiveness of insecticidal character, larvicidal and ovicidal of the two essential oils on *C. maculatus* evaluated through the study of their curative and persistent effects. These works could be thus pursued with other aromatic plants to identify and make available to farmers a practical use bluntly of effective essential oils in the protection of cowpea stocks.

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