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Fumigant and Repellent Effects of *Eucalyptus cinerea* and *Eucalyptus maidenii* Essential Oils on *Callosobruchus maculatus* F. 1775 (Coleoptera: Bruchidae) and *Sitophilus oryzae* L. 1763 (Coleoptera: Curculionidae)

[1]Hedjal-Chebheb Mariam,
[2]Medjdoub-Bensaad Ferroudja*,
[3]Toudert-Taleb Karima,
[4]Mediouni-Bendjemaa Djouda

Author's Affiliation:
[1,2,3]Laboratoire de production, sauvegarde des espèces menace et des récoltes. Influence des variations climatiques. Département de Biologie. Faculté des Sciences Biologiques et des Sciences Agronomiques. Université Mouloud Mammeri de Tizi-Ouzou 15000. Algérie.
[4]Laboratory of Biotechnology Applied to agriculture, INRAT, Rue Hedi Karray, University of Carthage, 2080 Ariana, Tunisia.

***Corresponding Author:**
Medjdoub-Bensaad Ferroudja,
Laboratoire de production, sauvegarde des espèces menace et des récoltes. Influence des variations climatiques. Département de Biologie. Faculté des Sciences Biologiques et des Sciences Agronomiques. Université Mouloud Mammeri de Tizi-Ouzou 15000. Algérie.
E-mail: medjdoubferroudja@yahoo.fr

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ABSTRACT
Eucalyptus maidenii and Eucalyptus cinerea essential oils of were extracted by the drive technique with water vapor and analyzed by means of gas chromatography coupled to mass spectrometry. The results reveal that the monoterpene compounds are the majority (57.69 and 51.28%) compared to sesquiterpenes (37.14 and 23.07%), and the 1.8- cineole is the most represented (70, 89 and 71.93%), respectively for E. cinerea. and E. maidenii. In fumigation tests, after 24 hours of exposure, with a dose of 12.5µl/l, E. cinerea and E. maidenii caused 100% adult mortality in Sitophylus oryzae. The same mortality rate was achieved at a dose of 25µl/l, with adults of Callosobruchus maculatus. The adults of S. oryzae are more sensitive to E. cinerea and E. maidenii with respectively, LD50 = 8.45 µl/l and 8.95 µl/l, LD95 = 10.45µl/l and 11.62 µl/l, compared to C. maculatus, with LD50 = 11.75 µl/l and 12.35 µl/l, and LD95 = 26.90 µl/l and 19.07µl/l for, respectively, E. cinerea and E. maidenii essential oils.

KEYWORDS: *Essential oils, CGMS, Callosobruchus maculatus, Sitophilus oryzae, Inhalation, Repellency, LD50, LD95.*

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1. INTRODUCTION

The protection of cereal and vegetable crops is of vital importance in terms of food for African populations. Insect pests cause significant damage in grain storage warehouses especially in African countries, up to 800g/kg (Ouedraogo and *al.*, 1996). Different methods can be used to fight against these pests, but chemical control still remains frequent. Pesticide use is the most widespread method to reduce the damage caused by insect pests, despite all the drawbacks. The use of biopesticides may represent a promising alternative.

The drawbacks associated with the use of chemicals in the warehouse have encouraged many authors to search for an alternative. They have been able to identify the toxic impact of several natural substances (powders, vegetable oils and essential oils), by contact, fumigation and repellency on many insect pests of stored products. This is the case, for example, of Keita (2000), Tapondjou and *al.* (2003), Kellouche and *al.* (2004), Kellouche and *al.* (2010), Hedjal-Chebheb and *al.* (2013) and Toudert-Toudert and *al.* (2014) on *Callosobrochus maculatus*; Huang and *al.* (2002) on *Sitophilus zeamais* and *Tribolium castaneum* (Herbest: 1797); of Tapondjou and *al.* (2005) on *S. zeamais* and *T. castaneum*; Mohamed and Abdelgaleil (2008) on *Sitophilus oryzae* L. and *T. castaneum*; of Bachrouh and *al.* (2010) on *T. castaneum*, of Mediouni-Bendjemaa and *al.* (2011) on *T. castaneum* and *R. dominica*; and Hamdi-Haouel and *al.* (2015) on adults of three populations of insects of Algerian and Tunisian origin (*R. dominica, T. castaneum and C. maculatus*).

It is in this framework that our work is conducted; the aim is to study the toxicity of two essential oils, that belong to the Myrtaceae family (*Eucalyptus cinerea* and *Eucalyptus maidenii*), against two insect pests of stored products, *S. oryzae* and *C. maculatus*.

2. Material and methods

2.1 Rootstock

E. maidenii and *E. cinerea* come from Souinat arboretum, located some ten kilometers from Ain Drahem (Northern Tunisia), situated in the humid bioclimatic stage with mild winters. The samples collected are placed in a dry place, away from lightand heat, and spread over paper to dry for a week.

2.2 Extraction of essential oils

The extraction was performed in the INRGREF laboratory of ecology and pastoral forestry improvement (Ariana, Tunis). The experimental device consists of a still and a condenser. The vapors leaving the still go to a condenser filled with water and are collected in a stainless metal tube.

The essential oils are then separated from the mixture (essential oil and water) and collected in a separator funnel. Those oils are stored at a temperature below 20°C and away from light.

The samples are first deposited on a sieve within the still containing water heated to 100°C. The essential oils are driven by the water vapor to a condenser where they condense in a coil.

The mixture is collected in a separatory funnel and separated into two immiscible phases. In the lower part is water (gas phase) and the upper part comprises the essential oil (organic phase).

2.3. Essential oils analysis

E. maidenii and *E. cinerea* essential oils were analyzed by gas chromatography coupled to mass spectrometry conducted at the National Institute of Research and Physicochemical analyses (INRAP), Technopole, Sidi Thabet, located at 30 km from Tunis.

2.4. Chromatography conditions

The GC/MS de vice is an Agilent and the injection system is a splitless split. The column length is 30mwith a diameter of 0.25 mm. The column, has a thickness of 0.25 microns.

The initial temperature of 40°C is maintained for one minute. It increases at 2°C/min up to 240°C. The latter temperature is maintained for 20 minutes. The temperature in the injector and the interface is 250°C, and that of the source is 230°C. The chromatogram of total ions is recorded using an electron impact source, and the ion kinetic energy is 70 eV.

The results of essential oils analyses are presented as chromatograms and a NIST

Database (National Institute of Standard and Technology) report.

The chromatogram of each essential oil has several peaks. Each peak is represented by a retention time which indicates the nature of the compound of the essential oil and as a percentage of the peak area, which is the percentage of the compound of oil compared to other compounds. The NIST Database report is a table that gives the characteristics of each peak in the chromatogram (essential oil) according to the method C/msdchem/1 Method/HP1-HE.SAM-0.1.M.

After the identification of the various constituents of the essential oils, the terpene compounds was classified on the basis of the number of units in C10 they contain, in relation to the total number of compounds of each essential oil (monoterpenes: C10H16; sesquiterpenes: C15H24 and diterpenes: C20H32) (Guignard, 2004).

2.5. Mass breeding of cowpea beetle

The insects used during our tests come from mass breeding realized in a dark oven in which prevailing temperature conditions are 30 ± 1°C with a relative humidity of 70 ± 5%. The *C. maculates* individuals, emerging from seeds of *V. unguiculata,* are introduced into glass jars (1l) containing healthy cowpea seeds. The adult weevils used are younger than 24 hours. The cowpea seeds used as food for weevils come from the local market.

2.6. Organic insecticides tests

2.6.1. Inhalation tests

The test focuses in assessing the insecticidal effect of essential oils by fumigation on *C. maculatus* adults. In glass jars, one liter of volume, a pure essential oil dose is deposited on a piece of Whatman No. 1 filter paper, suspended with a thread in the inner face of their lids. The doses tested for all the two oils are: 6.5, 12.5, 25 and 50 µl/l of air. Meanwhile, a control is prepared (without essential oil). Ten pairs of *C. maculatus*, aged 0 to 24 hours, are rapidly introduced into each jar, which is then sealed. A count of dead individuals is then performed after a variable exposure time: 24, 48, 72 and 96 h.

2.6.2. Repellency test

Filter paper discs of 11cm diameter are cut into two equal parts. One half-disc is treated with a dose of essential oil diluted in 1ml acetone. The second half-disc is treated only with the solvent (1ml acetone). After complete evaporation of the solvent in the open air for 15 minutes, the filter paper half-discs are put together with an adhesive and then placed at the bottom of Petri dishes. In the middle of these half-discs, we released 10 couples of adult weevils aged under 24h.

The doses tested were: 6.5, 12.5, 25 and 50µl, and four repetitions are performed for each dose. After one hour, a count of weevils present on both parts is performed. The same procedure is applied for both *S. oryzae* and *C. maculatus*.

The percentage of repellent essential oils against adult insects is calculated using the formula suggested by McDonald and Guy (1970):

PR (%) = [(NH NAc) / (Nac + NH)] × 100.

Nac=number of individuals present on the part treated with acetone only. NH = number of individuals present in the area treated with the essential oil diluted in acetone.

2.7. Statistical analysis

Considering the normal nature of the results obtained, the ANOVA test was used, on the basis of several classification criteria. When the treatment effect is significant, the analysis is completed with the Newman and Keuls test at 5% (Software STATITCF; Dagnelie 1998). LD50 and LD95 are calculated with the probit software (Finney, 1971).

3. Results

3.1. Analysis of essential oils

The results of analyses of the essential oils of both Myrtaceae show that the rate of monoterpenes is higher than that of sesquiterpenes. It is on average 57.69% for *E. cinerea* and 51.28% for *E. maidenii* (Table 1).

Table 1: Composition (%) in terpene hydrocarbon of the two essential of *E. cinerea* and *E. maidenii* of Tunisian origin

	E. cinerea	***E. maidenii***
Monoterpene compounds (%)	57.69	51.28
Sesquiterpene compounds (%)	37.14	23.07
Identified compounds (%)	99.48	98.28

The majority compounds of *E. maidenii* and *E. cinerea* essential oils are, respectively, eucalytpol (71.93% and 70.89%), α-pinene (14.01% and 7%) and 4-Carene (12.68 and 0.19). Some compounds are only present in the essential oil of one species: β pinene, camphor and D α terpinene in *E. maidenii*; terpinolene, nerolidol and spathulenol in *E. cinerea* (Table 2a and tab. 2b).

Table 2a: Rate of different monoterpenes compounds in *E.cinerea* and *E.maidenii* essential oils

Monoterpene Hydrocarbons (%)	***E. maidenii***	***E. cinerea***
α Pinène	14.01	7
β Pinène	0.34	-
Camphène	0.19	0.10
4 Carène	0.19	12.68
β Myrcène	0.17	-
Terpinolène	-	0.11
O. Cymène	0.13	-
α Terpinène	0.65	-
Eucalyptol	71.93	70.89
Camphor D	0.25	
Fenchol	0.12	0.13
Terpinène 4 ol	0.24	0.66
Verbenol	0.12	-
Terpinèol	0.17	3.54
D carvone	0.11	-
Carvacrol	0.12	-
Bornéol	0.27	0.40
α pinène époxide	0.62	0.14
Isopinocarvéol	1.75	0.25
Trans carvéol	0.13	0.11
β Citral	0.38	-

Table 2b: Rate of different sesquiterpene compounds in *E.maidenii* and *E. cinerea* essential oils.

Sesquiterpene hydrocarbons	***E. maidenii***	***E. cinerea***
Caryophyllène	0.62	0.52
Aromadendrène	2.15	0.18
α Salinene	0.12	-
β Calarène	0.10	-
Gurjunene		0.11
Globulol	1.99	1.42
Spathulénol		0.36
Epiglobulol	0.34	-
Eudesmol	0.17	-
Selinenol	0.90	-
Nerolidol	-	0.37
Identified Compounds(%)	97.81	97.54

3.2. Fumigation test

3.2.1. Effect of *E. cinerea* essential oil on *C. maculatus* and *S. oryzae*

The results of the analysis of variance have shown a highly significant effect for the insect factor (F = 137.06, P = 0.000; DDL = 1), for the time factor (F = 36.22, P = 0.000; DDL = 3), for the dose factor (F = 1795.92, P = 0.000, df = 4), and for the interaction of the three factors (F = 11.47; P = 0.000, df = 12). *C. maculatus* seems more resistant to treatment with the lowest dose (6.5µl / l), after 72 h of exposure to the essential oil of *E. cinerea*. This is confirmed in the treated groups with the dose 12.5 µl/ l: the mortality rate in *S. oryzae* is 100%, while it is only 35% in *C. maculatus* after 24 h exposure (Table 3). The essential oil dose required to achieve 100% mortality after 24 hours of exposure is 12.5 µl / l in *S. oryzae* and 25 µl/ l in *C. maculatus*.

Table 3: Mortality rate (average ± standard deviation) of *C. maculatus* and *S. oryzae*) adults treated with essential oil of *E. cinerea* at different exposure times (average flowed by a different letter vary very singnificantly at the 5% threshold for each insect species.

Insects pest	**Time (h) Dose µl/1**	**24**	**48**	**72**	**96**
C. maculatus	0	0.00 ± 0.00 (f)	0.00 ± 0.00 (f)	0.0 ± 0.00 (f)	0.00 ± 0.00 (f)
	6.5	0.00 ± 0.00 (f)	0.00± 0.00 (f)	0.00 ± 0.00 (f)	35 ± 5.77 (e)
	12.5	35 ± 19.15 (e)	100 ± 0.00 (a)	72.5 ± 5 (b)	100 ± 0.00 (a)
	25	100 ± 0.00 (a)	100 ± 0.00 (a)	100 ± 100 (a)	100 ± 0.00 (a)
	50	100 ± 0.00 (a)	100 ± 0.00 (a)	100± 0.00 (a)	100 ± 0.00 (a)
S. oryzae	0	0.00 ± 0.00 (f)	0.00 ± 0.00 (f)	0.00± 0.00 (f)	0.00 ± 0.00 (f)
	6.5	0.00 ± 0.00 (f)	0.00 ± 0.00 (f)	35 ± 5.77 (e)	43.50 ± 2.50 (d)
	12.5	100 ± 0.00 (a)	100 ± 0.00 (a)	100 ± 0.00 (a)	100 ± 0.00 (a)
	25	100 ± 0.00 (a)	100 ± 0.00 (a)	100 ± 0.00 (a)	100 ± 0.00 (a)
	50	100 ± 100 (a)	100 ± 0.00 (a)	100 ± 0.00 (a)	100 ± 0.00 (a)

3.2.2. Effect of *E. maidenii* essential oil on *C. maculatus* and *S. oryzae*

The results of the analysis of variance have shown a highly significant effect for the time factor (F = 331.53, F = 0.000; DDL = 3), the dose factor (F = 8059.23, P = 0.000; DDL = 3), the insect factor (F = 193.4, P = 0.00, df = 1) and for the interaction of three factors (F = 61.06; P = 0.000; df = 1). The adults of *C. maculatus* and *S. oryzae* exposed to the essential oil of *E. maidenii* suffer increased mortality rates as and when the dose and duration of exposure increase (Table 4).The lowest dose of essential oil (6.5µl / l) causes about 50% mortality after 96 hours of exposure in both pest species. It takes 48 hours of exposure to obtain 100% mortality at a dose of 12.5µl / l in *S. oryzae*, whereas for *C. maculatus*, it takes 72 hours.The same mortality rate (100%) is obtained in both insects with a dose of 25 µl / l and after 24 hours of exposure (Table 4).

Table 4: Mortality rate (average ± standard deviation) of *C. maculatus* and *S. oryzae*) adults treated with essential oil of *E. maidenii* different exposure times (average flowed by a different letter vary very singnificantly at the 5% threshold for each insect species.

Insects pest	**Time (h) Dose µl/1**	**24**	**48**	**72**	**96**
C. maculatus	0	0.00 ± 0.00 (f)	0.00 ± 0.00 (f)	0.00 ± 0.00 (f)	0.00 ± 0.00(f)
	6.5	0.00 ± 0.00 (f)	0.00 ± 0.00 (f)	20 ± 0.00 (e)	55 ± 5.77(b)
	12.5	28.75 ± 6.29 (d)	50 ± 11.55 (b)	100 ± 0.00 (a)	100 ± 100 (a)
	25	100 ± 0.00 (a)	100 ± 0.00 (a)	100 ± 0.00 (a)	100 ±0.00 (a)
	50	100 ± 0.00 (a)	100 ± 0.00 (a)	100 ± 0.00 (a)	100 ± 0.00 (a)
S. oryzae	0	0.00 ± 0.00 (f)	0.00 ± 0.00 (f)	0.00 ± 0.00 (f)	0.00 ± 0.00 (f)
	6.5	0.00 ± 0.00 (f)	0.00 ± 0.00 (f)	35 ± 5.77 (a)	52.50 ± 9.75 (b)
	12.5	97.50 ± 0.00 (a)	100 ± 0.00 (a)	100 ± 0.00 (a)	100 ± 0.00 (a)
	25	100 ± 0.00 (a)	100 ± 0.00 (a)	100 ± 0.00 (a)	100 ± 0.00 (a)
	50	100 ± 0.00 (a)	100 ± 0.00 (a)	100 ± 0.00 (a)	100 ± 0.00 (a)

3.2.3. Toxicity of *E. cinerea* and *E. maidenii* essential oils against *C. maculatus* and *S. oryzae* adults

The calculation of lethal doses (LD50 and DL95) reveals a comparable toxicity of the two essential oils tested. *S. oryzae* adults are more susceptible to *C. maculatus*. In fact, the LD50s are successively of 8.45 to 8.943 µl/l of air in *S. oryzae*, and 11.75 to 12.35 µl/l of air in *C. maculatus*. The same is true for the LD95 values, which are higher with the cowpea beetle (Table 5).

Table 5: LC50 and LC95 values of *E. cinerea* and *E. maidenii* essential oils against *S. oryzae* and *C. maculates*

	S. oryzae		***C. maculates***	
	E. cinerea	***E. maideni***	***E. cinerea***	***E. maiden***
$CL_{50}^{a,b}$ (µl/l)	8.456 (8.196-8.831)	8.943 (8.560-9.395)	11.755 (9.673-16.294)	12.356 (7.755-28.381)
$CL_{95}^{a,b}$ (µl/l)	10.476 (9.782-11.829)	11.628 (10.811-13.030)	19.907 (14.955-55.031)	26.906 (16.624-1120.776)
Slope ± SEM	17.681± 2.656	14.42 ± 1.84	7.18 ± 1.36	4.867 ± 1.232
Degree of freedom	8	8	8	8
χ^2	4.086	3.071	8.034	18.183

[a] Units LD50 and LD95 = µl/ air, applied for 24 h at 25 °C.
[b] 95% lower and upper confidence limits are shown in parenthesis.

3.3. Repellent test

The results of the analysis of variance tests for the repellency parameter show that there is a very highly significant difference in the dose factor (F = 43.03, P = 0.000; DDL = 3), a highly significant difference for the oil factor (F = 11.88, P = 0.0017; DDL = 1) and a non-significant difference for the insect factor (F = 1.36, P = 0.2502; DDL = 1).

According to the results obtained, *E. cinerea* and *E. maidenii* essential oils are considered moderately repulsive against *S. oryzae* and *C. maculatus*. The repellency rate varies from 45 to 60%, depending on the essential oil and the pest (Table 6).

Table 6: Reppellency rate (%) of *E. cinerea* and *E. maidenii* essential oils, against *C.maculatus* and *S. oryzae*

	Doses	***C. maculatus***	**Average**	***S. oryzae***	**Average**
E. cinerea	6.5µl	26.67±11.55	46.66 % Averagly repelent	20±0.00	45% Averagly repelent
	12,5 µl	40±0.00		26.67±11.55	
	25 µl	50 ±0.00		53.33±11.55	
	50 µl	70±0.00		80±0.00	
E. maiideni	6,5 µl	40 ± 0.00	51.58 % Averagly repelent	46.67±23.09	60% Averagly repelent
	12,5 µl	46.33±1.15		46.76±11.5	
	25 µl	50±0.00		60±0.00	
	50 µl	70±0.00		86,67±23.09	

4. DISCUSSION

In both essential oils, monoterpenes are predominant in comparison with sesquiterpenes. Their average rate varies between 51.28% and 57.69%. Bruneton (2005) noted that monoterpenes account for more than 90% of essential oils. Our results are similar to those of El Aissi (2011), who noted that in several species of the genus *Eucalyptus*, the rate of monoterpenes is higher than that of sesquiterpenes in *E. cinerea* (90.60% and 2.3%) and *E. maidenii* (86.5% and 12%). The major compound in both essential oils is the 1.8- Cineole for *E. cinerea* (70.89%) and *E. maidenii* (71.93%) species. According to Toudert-Taleb and *al.* (2014), eucalyptol predominates in *E. globulus* (47.05%) and *E. radiata* (66.34%). For Haouel and *al.* (2010), this rate is 19.87% in *E. rudis* and 20.62% in *E. camaldulensis*. El Aissi (2011) confirms this result with *E. occidentalis* (18.8%), *E. largiflorens* (63.6%), *E. leucoxylon* (59. 2%), *E. biscota* (68%), *E. gracilis* (68%), *E. torquata* (12%) and *E. salmonaphiloria* (37%). Thus we note some variability in eucalyptol composition in the same family of Myrtaceae. This can be due to several factors: the climate, soil and tillage practices (Regnault Roger and *al.* (2008). The richness in eucalyptol of the *Eucalyptus* genus has been confirmed by several authors (Guignard, 2004; Bruneton, 2005; Dellil, 2010).

In inhalation tests, we have found that the two essential oils of *E. cinerea* and *E. maidenii* caused 100% mortality in *S. oryzae* and *C. maculatus* adults, at a dose of 12.5 µl/ l, for 24 and 72 h exposure, respectively. Several authors have also noted a difference in the mortality of pests depending on the duration of exposure to essential oils. Thus, Kim and *al.* (2003) obtained a 90% mortality of *S. oryzae* adults treated with the essential oil of *Brassica juncea*, *Cinnamonum cassia* and *Cocholeria Arocaria*, with a dose of 3.5mg/cm^2, after one day exposure; whereas with the other essential oils *Acarus calamus*, *Acarus gramineus* and *Agastache rugosa*, the mortality rate is 100% after 3 days of exposure. It appears that the mode of action of essential oils against insects is attributed largely to the penetration of the terpene compounds in the respiratory system.

S. oryzae adults are more sensitive to essential oils of *E. cinerea* and *E. maidenii* (LD50 = 8.45µl/1 and 8.94µl/1 of air) compared to *C. maculatus* adults (LD50: 11.75 and 12.35µl/l). Some authors have also demonstrated a sensitivity difference of several insect pests in stored grains to certain natural substances. Mohamed and Abdelgaleil (2008) have noted that *S. oryzae* is more sensitive to treatment with the essential oil of *Mentha microphylla* (LC50 = 0.21µl/l) compared to *Lantana camara* (LC50 = 29.47µl/l) and *Eucalyptus camaldulensis* (LC50 = 50 .µl/l). Similarly, Kim and *al.* (2003) have shown that the toxicity of essential oils varies with the insect and the chemical composition of the oils.

Furthermore, several studies have shown the toxic effect of *Eucalyptus* against insect pests of stored products. Toudert-Taleb and *al.* (2014) have reported the toxicity of *E. globulus* and *E. radiata* against *C. maculatus* adults, with a dose of 8µl/l, after 48 hours of exposure. Similarly, Kellouche and *al.* (2010) have noted the same effect with the essential oil of *E. globulus* and *E. citiodora* with a dose of 20 µl/l, after 24 hours of exposure on the same pest. Moreover, Hamdi-Haouel and *al.* (2015) have also shown the insecticidal effect of *E. lehmanii* and *E. astingens* on *C. maculatus*, *R. dominica* and *T. castaneum*. As regards the insecticidal activity of the essential oil components, the work conducted by Agarwall and *al.* (2001a and b) highlighted the high toxicity of 1-8 Cineole that causes 100% mortality in three beetles that are pests of stored products (*C. maculatus*, *R. dominica* and *S. oryzae*), with a dose of 1µl/l. In addition, Regnault Roger (1997) has highlighted the toxic effect of monoterpenes by fumigation on the bean weevil, *Acanthoscelides obtectus*. Kim and *al.* (2003), who have studied the fumigation of essential oils on *S. oryzae* and *C. chinensis*, obtained results which show that toxicity depends on the insect species, the plant and the time of exposure to the essential oil. We believe that the toxicity of these essential oils can be linked to the action of their major compound, namely eucalyptol. Mill and *al.* (2010), cited by Regnault Roger and *al.* (2008), have noted that the essential oils monoterpenes are neurotoxic elements that act according to their chemical nature. Whatever the essential oils tested in the fumigation tests, terpene compounds act on the motor activity of insects. It is strongat the beginning, and then it slows down gradually till death. Peterson and Peterson and *al.* (2003) have reported that monoterpene compounds, eucalyptol, fenche, and pulgenone, at a dose of 50 mg/ml of air, can cause mortality in *T. castaneum*, *S. oryzae* and *Oryzaephilus surinamensis* .

In the repellency tests, *E. cinerea* and *E. maidenii* have proven to be moderately repellent at a dose of 50 µl/l. The repellent effect of these essential oils is related to the presence of monoterpene and sesquiterpene compounds. For Nerio and *al.* (2010), the compounds that have repellent activity are α pinene and limonene. The same authors, Nerio and *al.* (2009), have highlighted a moderate repellent effect on *C. maculatus* of *E. globulus*. For Toudert-Taleb and *al.* (2014), the essential oil of *E. globulus* has been shown to be highly repulsive at a dose of 12.5µl/l against *C. maculatus*.

The *E. saligna* essential oil has proven highly repellent to *C. maculatus* at a dose of 0.46 µl/cm^2 (Tapondjou and *al.*, 2005).

Enan (2001) has estabished the link between the application of eugenol, α terpineol and cinnamic alcohol and blocking of the receptor sites of octopamine (a regulating effect on the heart beat, movement, breakdown, flight and metabolism of invertebrates). This author has reported that the effect may vary from one terpene to another and that the essential oil may act as an antagonist of neurotransmitters. Coats and *al.* (1991) have reported that monoterpenes are neurotoxic, as they inhibit the receptor sites of acetylcholinesterase. Regnault Roger and *al.* (2008) have noted that, regardless of the essential oils tested in fumigation tests, terpene compounds act on the motor activity of insects.

5. CONCLUSION

1.8- Cineole is the main component (70-72%) in *E. cinerea*. and *E. maidenii* essential oils. The high toxicity by fumigation of these natural substances, against the two main insect pests of stored grains, was highlighted. In fumigation tests, after 24 hours of exposure with a dose of 12.5µl/l, *E. cinerea* and *E. maidenii* caused 100% adult mortality in *S. oryzae*. The same mortality rate was achieved at a dose of 25µl/l, with adults of *C. maculatus*. The adults of *S. oryzae* are more sensitive to *E. cinerea* and *E. maidenii,* with respectively, LD50 = 8.45 µl/l and 8.95 µl/l, compared to *C. maculatus*, with LD50 = 11.75 µl/l and 12.35 µl/l.

In order to determine more precisely the effect of these two essential oils, it would be interesting to study their synergistic effect on these two major insect pests, and on other species that are dependent on stored seeds.

It would also be useful to complement this study with other toxicity tests on other insects dependent on cereal grains (*R. dominica* and *T. castaneum*) and legumes (*A. obtectus* and *C. chinensis*). The assessment of the toxicity of these natural substances with topical applications would also be of interest.

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