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Biodegradation of Insecticidal Compounds of *Clausena anisata* and *Plectranthus glandulosus* Essential Oils Applied as Protectant on Stored Grains

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Abstract: Essential oils of some aromatic plants are suggested in Northern Cameroon as alternatives to hazardous pesticides having harmful effects on the consumer and the environment. The active compounds of these essential oils are very volatile, easily biodegradable. To be effective, treatments should be made with short interval and regular time. This mode of use generates the accumulation of constituents of these essential oils on the treated food and could limit food security and safety. The present study aimed at evaluating the variation of the constituent's quality of *Clausena anisata* and *Plectranthus glandulosus* essential oils and their levels on food products according to time. In this way, samples of corn grains and flour were treated with these essential oils and stored during 150 days. During this storage, the persistent compounds present in these samples were extracted by hydrodistillation and analyzed by GC/FID. The obtained results show that essential oils concentration decreases on food products according to the duration of storage with half-life times (IT50) of 24.16 and 34.61 days for *C. anisata* and 25 and 38.75 days for *P. glandulosus*, respectively on grains and flour. At 150 days after the treatment, there is no more that 6 constituents of *C. anaisata* and 3 of *P. glandulosus* on the grains and 10 and 7 constituents on the flour, respectively for these two essential oils. The rates of these persistent constituents are >62.5 times lower than the toxic concentration observed from the day of treatment. At these used doses, these constituents are not toxic to consumers.

Key words: Persistent compounds, essential oils, safety food, toxicity, stored products, Cameroon

INTRODUCTION

Attacks of insects on food products are so significant that the only effective strategy of protection of these resources is the treatment with chemical pesticides. The used products are sometimes dangerous with significant negative impacts on consumers and the environment. This is noted at the level of the quality and the quantity of residues of these pesticides. To promote essential oils which are strongly biodegradable implies to privilege a track that leaves few residues on the food products.

Sitophilus zeamais and *Tribolium castaneum* are important pests of stored products in Cameroon. It is also insects in which the risk of development of resistance to insecticides is high (Bughio and Wilkins, 2004). *T. castaneum* attacks the flour for which any treatment with chemical weapon led to a direct intoxication of the consumer.

It becomes therefore, useful to build up alternative methods of controlling pest by methods that are user-friendly as the use of agents with high efficacy on the pest and low persistence in the food. There are needs to develop and popularize such control techniques that are clean and unharmful to the user as the natural essential oils. These natural products as essential oils are often highly specific and biodegradable of low persistence. In the early 1970's, an alternative was the use of natural products as pesticides to control pests during storage. Ethno botany has therefore, played a very important role in the protection of crops against pests in Africa and Asia (Kouninki *et al*., 2007); plants were used at the time in granaries by the farmers naturally to protect their product. Most of the essential oils or vegetable oils used in crop protection are extracted from plants formerly known to have insecticidal effects on the population.

In this way, *C. anisata* and *P. glandulosus* with remarkable insecticidal and preserving activities are very volatile, easily biodegradable, consequently treatments should be made with the short interval and regular time to be effective. This mode of use could make unsuitable the treated food by the accumulation of constituents of these essential oils and involve on the one hand, a problem of

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food surety and safety and on the other hand the development of the resistance of the targeted insects. The compounds present on the food products after their anti-insect effectiveness can be regarded as persistent. The objective of this study was to seek and quantify these persistent bioactifs compounds of *C. anisata* and *P. glandulosus* essential oils in order to evaluate their toxicity.

MATERIALS AND METHODS

Hydrodistillation and conditions of treatments: Essential oils were obtained by hydrodistillation of the sheets of *C. anisata* and *P. glandulosus* as described by Goudoum *et al.* (2009). Grains used were CMS 8504 variety. The flour of these grains was obtained using a crusher Polymix (Px-mfc Model, Germany) with mesh of 1 mm. These grains and flour were treated using the CL_{80} of essential oils as described by Goudoum *et al.* (2010).

Treatment of corn grains and flour: About 200 g of corn grains or flour are introduced in 1000 mL bottle and treated with 500 μL of LC_{80} of each oil. The corn grains were treated as described by Goudoum *et al.* (2010) and corn flour according to Keita *et al.* (2001)'s method. Twice food products are storage during 150 days. All products are receipt three consecutively treatments of each oil after 10 days of interval.

Extraction of the essential oil contained in the flour and corn grains: The Persistent compounds of essential oil contained in the flour and corn grains treated with *C. anisata* and *P. glandulosus* is extracted by hydrodistillation during 2 h with a Clevenger apparatus as described by Goudoum *et al.* (2010). The tridecane was selected like internal standard for these following reasons: it is eluted in an exploitable zone of the chromatogram and it does not interfere with essential oil constituents; its index of Kovats 1300 is a good compromise for the major of components. Four repetitions were carried out. The untreated corn grains and flour are constituted the control.

Chromatographic analysis: The chromatographic analysis was made according to conditions described by Goudoum *et al.* (2009) in a GC/FID (Chromatograph SHIMADZU HP-5989).

Identification of molecules: The identification of the pure compounds was carried out by comparison between times of retention and the spectral data of the library (Davies, 1990; Kouroussou *et al.*, 1998).

Calculation of the concentration of persistent compounds of essential oils present in food: The calculation of the concentration was carried out by comparison with that of the internal standard. As regards the expression of the residual quantities of essential oil, the relarguage was given by proportioning the quantity of persistent oil. This extracted oil was analyzed by GC/FID and made it possible to express by the various modified formulas of Noudjou as described by Goudoum *et al.* (2010).

RESULTS AND DISCUSSION

Chemical composition of essential oils of *C. anisata* and *P. glandulosus*: The chemical analysis of *C. anisata* and *P. glandulosus* essential oils is shown in Table 1. *C. anisata* essential oil studied contains 18 compounds. These 18 compounds represent 95.12% (Table 1). The sabinene, trans linalool oxide, estragole, E-caryophyllene, β-copaene, α-humulene, germacrene D and E-nerolidol are the major compounds constituting 71.73% of the crude essential oil obtained by distillation with Clevenger apparatus. *P. glandulosus* essential oil contains 15 compounds (Table 1).

Table 1: Chemical composition obtained by GC/FID of *Clausena anisata* and *Plectranthus glandulosus* essential oils from Cameroon

		Essential oils (Crude oil)	
KI	Composition	*C. anisata*	*P. glandulosus*
851	1-hexanol	-	1.23
943	α-pinene	-	1.06
977	Sabinene	4.91	-
991	β-myrcene	-	5.13
1008	d-3-carene	-	1.10
1027	Limonene	2.70	-
1076	Trans-linaloo oxide	4.25	-
1089	Fenchone	-	29.81
1090	α-terpinolene	2.94	28.29
1091	Cis linalool oxide	1.08	-
1100	linalool	1.21	-
1127	cis-p-menth-2-en-1-ol	1.73	-
1142	Camphor	-	1.34
1146	Terpinene-4-ol	-	2.51
1179	P-cymene-8-ol	-	2.80
1193	Estragole	23.68	-
1201	Methyl salicylate	2.12	-
1234	Z-ocimenone	2.11	-
1243	E-ocimenone	2.08	-
1247	Cis-piperitone oxide	-	2.82
1292	Thymol	6.07	-
1315	Piperitenone	-	1.23
1348	Δ-elemene	2.07	-
1353	Piperitenone oxide	-	11.08
1389	α-copaene	1.11	-
1399	Isopulegone-4-methyl	-	1.11
1438	E-caryophyllene	4.68	-
1445	β-copaene	4.57	-
1473	α-humulene	9.78	-
1499	germacrene D	10.61	1.61
1571	E-nerolidol	10.12	-
	Total	95.12	93.82

Table 2: The reduction of the speed of concentrations and half life of *Clausena anisata* and *Plectranthus glandulosus* essential oils on the treated foods

		Reduction of concentration speeds (%/day)					
Essential oils	Foods	10	50	100	150	R	$T_{1/2}$ (days)
C. anisata	Grains	4.41[a]	0.70[b]	0.07[c]	0.04[c]	0.94	24.16
	Flour	3.4[a]	0.66[b]	0.14[c]	0.05[c]	0.97	34.61
P. glandulosus	Grains	5.17[a]	0.56[b]	0.10[c]	0.05[c]	0.93	25
	Flour	3.8[a]	0.47[b]	0.11[c]	0.07[c]	0.94	38.75

$T1_{1/2}$ = Essential oils half-life time; R = Regression between durations of observations; Mean values followed by the same letter in the same column do not differ significantly at $p<0.05$ (Duncan's test)

Where the β-myrcene, limonene, the fenchone, α-terpinolene and piperitenone oxide represent 74.31% of the crude oil compounds.

Reduction of the concentration of essential oils in flour and on corn grains: According to the manipulation conditions, studied essential oils lose gradually with time and the food product to which they are applied, their compounds. This reduction of concentrations follows kinetics with an equation of $Y = ax^2 + bx + C$ with a regression R = 0.9 between durations (Fig. 1). From Fig. 1, it results 3 phases, each one separate by an inflection point which are:

- First which is that ranging between day 0 and 10. The 10th day corresponds to the first point of inflection where nearly 50% of essential oils concentrations are lost on grains and 35 % on the flour. The reduction of the speed of concentrations (Table 2) are highest at this phase with 4.41 and 5.17% day^{-1} on grains and 3.40 and 3.80% day^{-1} on flour, respectively for *C. anisata* and *P. glandulosus*
- Second, ranging between 10 and 50th day is the one which the final concentration of the first phase decrease by 35% for *C. anisata* and 25% for *P. glandulosus*. Speeds associated to this phase are 0.66-0.69 for *C. anisata* and 0.50 and 0.56% day^{-1} for *P. glandulosus* respectively on corn grains and flour
- The last, phase is the one which goes from 50-150th day with any inflection point. Speeds between 50 and 100th day are 0.07 and 0.14% day^{-1} for *C. anisata*, respectively on grains and flour and 0.11 for *P. glandulosus* for two food products. These various speeds are reduced by half between 100-150th of exposure for two food products

Half-lives time associated with the reduction of concentrations of these essential oils are 24.16 days on grains and 34.61 days on the flour for *C. anisata* and 25 days on grains and 38.75 days on flour for *P. glandulosus* (Table 2).

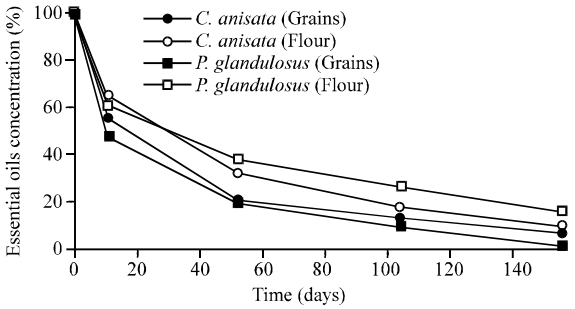

Fig. 1: The reduction of concentrations of *Clausena anisata* and *Plectranthus glandulosus* essential oils according to the storage period

Reduction of the content of essential oils compounds: It emerges from the results of Table 3 and 4 that the persistent compounds depend on the used essential oil and form of the food on which these oils are applied.

On corn grains: From 18 and 15 composed of departure, respectively for *C. anisata* and *P. glandulosus* essential oils that only 9 persisted up at 50 days and 6 at 150 days after treatment of grains by *C. anisata* and 10 at 50 days, 3 at 150 days by *P. glandulosus* (Table 3 and 4).

For the *C. anisata* essential oil, the sabinene rate on grains exposed to 28±2.2°C and 65±5.7%, falls from 798.37-245.53 µg/200 g at the 10th day and 16.16 µg/200 g after 100 days. The estragole rate falls slowly from 3850.41-793.50 µg/200 g at the 150th day. In the same way, E-nerolidol shows a progression similar to that of the estragole. Some minor compounds such as linalool, methyl salicylate, E-ocimenone and the thymol persist on grains after 150 days with 53.66, 82.93, 9.76 and 175.61 µg/200 g, respectively rate (Table 3).

As regards to the *P. glandulosus* essential oil, major compounds least persistent α-terpinolene and α-myrcene disappear, respectively beyond 10 and 50 days from exposure. The most persistent are:

- Piperitone oxide which smoothed out around 100th day, then contents go from 1801.63 at the treated day to 29.27 µg/200 g after 100th day
- Fenchone, one of most persistent presents at 1151.22 rate at 150th day

Table 3: The content of the various persistent compounds of *Clausena anisata* on corn grains

	Time (days)				
Compounds (µg/200 g)	0	10	50	100	150
Sabinene	798.37±22.12	245.53±3.02	19.51±0.41	16.26±0.07	0.00
Trans-linalool oxide	691.06±17.07	549.59±5.72	0.00	0.00	0.00
α-terpinolene	478.05±11.61	287.80±3.12	0.00	0.00	0.00
Cis linalool oxide	175.61±5.78	14.63±0.76	0.00	0.00	0.00
Linalool	196.75±2.82	190.24±1.82	165.85±2.03	143.09±1.03	53.66±0.52
Cis-p-menth-2-en-1-ol	281.30±4.67	0.00	0.00	0.00	0.00
Estragole	3850.41±88.34	3217.89±25.92	1969.11±12.05	1317.07±12.42	793.50±5.33
Methyl salicylate	344.72±4.23	227.64±2.73	198.37±1.12	125.20±1.32	82.93±0.67
Z-ocimenone	343.09±2.05	331.71±4.77	196.75±1.06	0.00	0.0
E-ocimenone	338.21±3.89	78.05±0.72	29.27±0.11	22.76±0.52	9.76±0.06
Thymol	986.99±24.76	391.87±3.12	292.68±1.42	191.87±2.51	175.61±1.73
δ-elemene	336.59±5.62	78.05±0.57	27.64±0.12	0.00	0.00
α-copaene	180.49±2.82	65.04±0.45	0.00	0.00	0.00
E-caryophyllene	760.98±6.45	279.67±2.45	0.00	0.00	0.00
β-copaene	743.09±7.07	237.40±4.32	0.00	0.00	0.00
α-humulene	1590.24±34.23	1269.92±17.72	0.00	0.00	0.00
Germacrene D	1725.20±24.56	517.07±7.11	0.00	0.00	0.00
E-nerolidol	1645.53±17.23	986.99±5.21	920.33±6.45	891.06±4.92	604.88±4.78

Table 4: The content of the various persistent compounds of *Plectranthus glandulosus* on corn grains

	Time (days)				
Compounds (µg/200 g)	0	10	50	100	150
1-hexanol	200.00±11.56	0.00	0.00	0.00	0.00
α-pinene	172.36±4.72	134.96±5.04	6.50±1.71	1.63±0.56	0.00
β-myrcene	834.15±7.63	4.88±0.76	1.63±0.88	0.00	0.00
δ-3-carene	178,86±4.76	58.54±2.85	48.78±2.34	21.14±1.23	1.63±0.08
Limonene	439.02±3.05	65.04±3.11	52.03±2.62	45.53±1.78	29.27±1.03
Fenchone	4847.15±83.55	4313.82±56.71	2484.55±26.71	2208.13±27.34	1151.22±22.56
α-terpinolene	4600.00±58.72	1770.73±45.44	0.00	0.00	0.00
Camphor	217.89±4.67	0.00	0.00	0.00	0.00
Terpinene-4-ol	408.13±4.98	175.61±3.61	0.00	0.00	0.00
ρ-cymene-8-ol	455.28±3.57	76.42±3.67	34.15±1.67	4.88±1.05	0.00
Cis-piperitone oxide	458.54±7.21	99.19±3.92	81.30±2.82	14.63±0.82	0.00
Piperitenone	200.00±4.81	68.29±2.12	0.00	0.00	0.00
Piperitenone oxide	1801.63±4.32	1016.26±5.63	983.74±4.93	29.27±1.52	0.00
Isopulegone-4-methyl	180.49±6.78	165.85±8.67	151.22±7.62	50.41±5.37	0.00
Germacrene D	261.79±5.12	8.13±0.56	8.13±1.52	4.88±0.45	0.00

Close to these *P. glandulosus* major compounds, some one known as minor take a form comparable like fenchone which persistence going until 150th day. There are δ-3-careens and limonene which respectively have 02.78 and 29.27 µg/200 g rate at 150th day (Table 4).

In corn flour: Just like on corn grains, 13 compounds of *C. anisata* and 11 of *P. glandulosus* persist in corn flour after 50 days of treatment. Evolution of their content in food is shown in Table 5 and 6. Some compounds of *C. anisata* essential oil completely lose their activity after 1st 10 days. There are α-terpinolene, cis-linalool oxide, E-caryophyllene, β-copaene and α-humilene whith respective rate of 312.20, 139.84, 261.79, 217.89 and 1339.84 µg/200 g. Close to these, compounds are located α-copaene (19.51), germacrene D (16.26 µg/200 g) which disparate after 50th day and trans-linaloo oxide and linalool with a respective rate of 13.01 and 6.50 µg/200 g at 100th day on flour.

Most persistent compounds found on corn flour at 150th day are: sabinene (11.38 µg/200 g), linalool (99.19 µg/200 g), cis-ρ-menth-2-in-1-ol (35.77 µg/200 g), estragole (956.10 µg/200 g), methyl salycilate (131.71 µg/200 g), Z-ocimenone (66.67 µg/200 g), E-ocimenone (50.41 µg/200 g), thymol (200.00 µg/200 g), δ-elemene (17.89 µg/200 g) and E-nerolidol (660.16 µg/200 g) (Table 5).

The compound rates present on flour treated with *P. glandulosus* are shown at Table 6. It is deduced from Table 6 that major compounds such as fenchone, α-terpinolene and piperitenone oxide are present until 150th day with respective rate of 988.62; 891.06 and 409.76 µg/200 g. Near to these major compounds, there are minor compounds which persist until 150th day. It is limonene (19.51 µg/200 g), terpinene-4-ol (40.65 µg/200 g), isopulegone-4-methyl (53.66 µg/200 g) and germacrene D (14.63 µg/200 g).

The persistent rate compounds found beyond the duration of efficacity are not toxic for vertebrates. The following Table 7 shows the level of toxicity of some essential oils compounds common to those of *C. anisata* and *P. glandulosus*.

Table 5: The rate of the various compounds of *Clausena anisata* essential oil persistent in corn flour

	Time (days)				
Compounds (µg/200 g)	0	10	50	100	150
Sabinene	798.37±32.52	245.53±8.72	78.05±6.54	56.91±5.76	11.38±4.82
Trans-linaloo oxide	691.06±27.78	611.38±12.2	214.63±8.23	13.01±5.05	0.00
α-terpinolene	478.05±18.55	312.20±10.43	0.00	0.00	0.00
Cis linalool oxide	175.61±11.71	139.84±9.67	0.00	0.00	0.00
Linalool	196.75±9.67	191.87±7.88	182.11±6.03	152.85±10.56	99.19±8.66
Cis-ρ-menth-2-en-1-ol	281.30±10.02	55.28±8.03	182.11±7.67	108.94±7.33	35.77±6.23
Estragole	3850.41±56.71	3258.54±51.66	2691.06±31.71	1409.76±14.47	956.10±9.77
Methyl salicylate	344.72±8.55	300.81±9.12	234.15±11.06	196.75±8.48	131.71±8.21
Z-ocimenone	343.09±7.66	330.08±8.05	245.53±7.22	136.59±6.32	66.67±6.03
E-ocimenone	338.21±11.88	217.89±10.14	182.11±5.87	84.55±4.66	50.41±7.34
Thymol	986.99±12.91	497.56±9.91	336.59±7.33	273.17±9.04	200.00±11.01
δ-elemene	336.59±6.55	138.21±5.92	66.67±5.91	45.53±5.71	17.89±4.91
α-copaene	180.49±10.33	104.07±6.44	19.51±8.61	0.00	0.00
E-caryophyllene	760.98±22.34	261.79±8.42	0.00	0.00	0.00
β-copaene	743.09±17.44	217.89±10.68	0.00	0.00	0.00
α-humulene	1590.24±31.51	1339.84±26.25	0.00	0.00	0.00
Germacrene D	1725.20±44.11	881.30±21.56	16.26±4.73	0.00	0.00
E-nerolidol	1645.53±17.56	1370.73±10.12	1118.70±12.52	934.96±9.45	660.16±12.33

Table 6: The rate of the various compounds of *Plectranthus glandulosus* essential oil persistent in corn flour

	Time (days)				
Compounds (µg/200 g)	0	10	50	100	150
1-hexanol	200.00±7.88	91.06±3.24	0.00	0.00	0.00
α-pinene	172.36±5.76	141.46±6.33	0.00	0.00	0.00
β-myrcene	834.15±6.21	377.24±4.91	87.80±3.56	0.00	0.00
δ-3-carene	178.86±5.75	82.93±4.03	0.00	0.00	0.00
Limonene	439.02±4.34	151.22±7.78	297.56±4.32	110.57±5.45	19.51±2.72
Fenchone	4847.15±54.66	4479.67±53.91	2741.46±23.67	1878.05±17.56	988.62±7.07
α-terpinolene	4600.00±61.21	2385.37±35.72	1769.11±22.56	1432.52±13.31	891.06±8.76
Camphor	217.89±5.03	0.00	0.00	0.00	0.00
Terpinene-4-ol	408.13±8.23	216.26±4.76	164.23±2.94	66.67±4.02	40.65±5.44
ρ-cymene-8-ol	455.28±8.67	141.46±4.45	21.14±2.77	16.26±2.78	0.00
Cis-piperitone oxide	458.54±7.23	143.09±2.88	175.61±4.04	50.41±3.82	0.00
Piperitenone	200.00±6.28	43.90±3.07	0.00	0.00	0.00
Piperitenone oxide	1801.63±8.83	1440.65±11.63	1026.02±8.86	686.18±7.26	409.76±7.71
Isopulegone-4-methyl	180.49±6.72	159.35±4.63	97.56±5.01	66.67±3.67	53.66±3.05
Germacrene D	261.79±5.05	102.44±3.22	29.27±1.96	17.89±2.11	14.63±2.02

Table 7: The toxicity level of some essential oil compounds of aromatic plants established in the literature

			Studied essential oils (mg kg^{-1})	
Compounds	Toxicity (mg kg^{-1})	Sources	0 day	10 days
Camphor	900*		1.09	0.00
ρ-cymene	4750		2.27	0.71
Estragole	1820		19.25	16.30
Limonene	4600*		2.20	0.75
Linalool	2790		1.00	0.92
Methyl salicylate	887	Golob *et al.* (1999)	1.72	1.23
α-pinène	ND		0.86	0.71
Terpine-4-ol	4300		2.04	1.10
Thymol	980		5.00	2.50
Germacrene D	ND		8.62	4.41
α-humulene	50	Rogerio *et al.* (2009)	8.00	6.70
E-nerolidole	ND		8.23	6.85
Fenchone	6160	Lachenmeier *et al.* (2008)	24.23	23.40
Piperitenones	400	Madhava and Nilesh (1998)	9.01	7.20

*Values obtained by ipr and expressed by LDLo; LD50: Letal Dose which killed 50% of experimental population; LDLo: The smallest lethal amount published; ipr: intraperitoniale; ND: Non Determined

It is deduced from Table 7 that all compounds rate of studied essential oils are less than the lethal dose which kills 50% of the experimental population of rats by oral administration. After 50 days of storage, the odors which emerge from food products would be due to compounds present at this precise date. The major compounds of the *C. anisata* essential oil are different from that analyzed by Avlessi *et al.* (2004) in West Africa. The composition of *C. anisata* studied is very similar to that found by Ngamo *et al.* (2007) but different in concentrations. As regards the *P. glandulosus* oil, the composition is qualitatively similar to that found by Ngassoum *et al.* (2001). This difference of composition and concentration would be due to the various geographical, ecological and physiological conditions of growth of these plants. *C. anisata* and *P. glandulosus* essential oils gradually lose their activity with a time and according to the food product on which they were applied. The essential oils on corn grains are quickly salted out than on flour. This could be due to the specific surface of retention of essential oils molecules on flour, higher than grains and/or flour being powdery could encapsulate bioactives molecules because of relative humidity within its particles.

Salting out rates of α-pinene, terpinene-4-ol, sabinene and phellandre are similar to those described by Noudjou which show that these compounds have a very high volatile rate during 3 first days and persist until 21st day with a 57.1% relative humidity in *X. aetropica* powder enriched to the same oil. This researcher also shows that germacrene D is weakly released with a slow rate than 40% after 21 days. In the same way, Huignard showed that 21% of careens-2, 38% of piperitone and estragole and 11% of linalool were released in the atmosphere bottle and were fixed by niebe grains.

These compounds are secondary metabolites of the plant, developed by this latter to fight against phytophagous. They are polyphenols, terpenes, alkaloids or glucosides cyanogenic. Of course, many allelochimic molecules are indexed in the pharmacopeias and are known for their pharmacological and therapeutic activities. *In situ*, they would not develop toxicity for vertebrates and are besides, numbers of them, regularly consumed by the human. It is the case of *C. anisata* and *P. glandulosus*, respectively used out of infusion in treatments of some affections such as the yellow fever, malaria, rheumatism, tires and facilitates childbirth (Ngamo *et al*., 2007).

CONCLUSION

C. anisata and *P. glandulosus* essential oils concentration decreases on the food products with the storage duration. The halt-times of this essential oils are 24.16 and 34.61 days for *C. anisata* and 25 and 38.75 days for *P. glandulosus*, respectively on the grains and flour. After 150 days of storage, number of compounds present is 6 for *C. anisata* and 3 for *P. glandulosus* on the grains, 10 and 7 on the flour, respectively. The persistent compounds of studied essential oils at used concentration would not toxic for the consumer.

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