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Adsorption of essential oil components of *Xylopia aethiopica* (Annonaceae) by kaolin from Wak, Adamawa province (Cameroon)

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ABSTRACT

Essential oils of aromatic plants are nowadays cited as suitable tools for better protection of stored grains from insect pest attacks. These chemical insecticides are less attractive to producers because of their low persistence and difficulty to use as pure product. There is therefore a need to formulate them as an easy handling chemical with better persistence. The present work aimed to study the formulation through adsorption of active components of *Xylopia aethiopica* Dunal (Annonaceae) essential oil on kaolin. *X. aethiopica* essential oil was obtained by hydrodistillation in a Clevenger type apparatus unit. Essential oil obtained was analysed by a GC-FID and a GC-MS. The adsorption measurements were performed with collected two kaolin fractions (Fl <100 µm; F2 <50 µm) and two kaolin fractions after H_2O_2 treatment (F3 <50 µm; F4 <2 µm). The kaolin was characterized by XRD, XRF, FTIR; its specific surface area was determined by the B.E.T method. The amount of essential oil adsorbed was inversely proportional to the particles size. Treatment of kaolin with hydrogen peroxide increased the adsorption capacity of essential oil components. The components adsorbed in highest amounts were sabinene, β-pinene, α-pinene and β-phellandrene.

Keywords*:* Kaolin; Adsorption; Essential oil components; *Xylopia aethiopica.*

1. Introduction

Clays are widely used in pesticide formulation as adsorbents or particulate fillers. Many researchers studied the adsorption of pesticide on clay material (Lagaly, 2001) to limit pest damages. So, the popular method to prevent stored products from insect attacks is the use of synthetic insecticides. This use is increasing each year. The intensive and uncontrolled use of chemical insecticides has direct effect on consumers and on the environment (Ngamo Tinkeu, 2004). There is therefore a need to develop products which respect to the environment and present less dangerous effect on consumer. Essential oils of aromatic plants currently used are cited as a good tool to prevent insect attacks on stored grains (Ngamo Tinkeu et al., 2007). The most important difficulty to popularise this new tool is formulation of a chemical active insecticide with moderate persistence (Lajide et al., 1995; Keita et al., 2001). Works carried out on the insecticidal activity of essential oil towards stored grains pointed out that insecticide possess repellent and killing activities (Szafranski et al., 1991 ; Prates et al., 1998; Ngamo et al., 2001, Regnault-Roger et al., 2002). Because of the higher volatility of essentials oils, the duration of their activities is very short. The local material suitable for the formulation of the essential oils seemed to be a kaolinite-type clay (Keita et al., 2001). It is cited as good adsorbent (Ghosh and Krishna, 2001) and is currently used in medicine, in cosmetics and for other insecticide (Borras, 1999). There are no adsorption studies of terpenic compounds on this clay material.

The aim of the present study is to determine adsorption of essential oil components on kaolin.

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2. Materials and methods

2.1. Plant material (Xylopia aethiopica) and hydrodistillation procedure

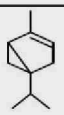
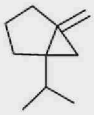
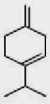
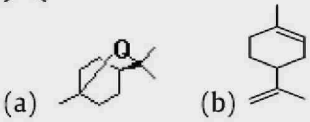
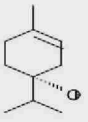
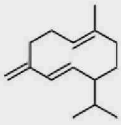
Dried fruits of X. *eathiopica* were purchased from Mbitom market (Adamaoua Province in Cameroon). They were harvested by local dealers. The essential oil was obtained by hydrodistillation of fruits for 4 h using a modified Clevenger-type apparatus. The essential oil obtained was kept at 4 °C in a sealed brown vial until its use.

2.1.1. Chromatographic analysis of essential oil

The essential oil was analysed by gas chromatography using a GC-14B with FID (Shimadzu Co.Japan) and integrator HP-3395B and a fused silica capillary column [Supelco 30 m x 0.25 mm ID, 0.25 µm film thickness coated with a 5% phenyl 95% dimethylpolysiloxane stationary phase (HP-5MS from Agilent)] and a split/splitless injector (splitless mode) at 230 °C. Detector temperature was 250 °C, injector port temperature 230 °C. The gases used were: nitrogen as carrier gas with a 1.5 ml/min flow rate, hydrogen gas at the detector was 30 ml/min and air at 250 ml/min. The pressure in the column was 2.9 kPa. Quantification was carried out by peak area calculations (GC/FID). The pure essential oil was qualified by GC/MS (Agilent 6890 GC coupled to an Agilent 5973 mass spectrometer) using the same GC/FID analytical conditions.

2.2. Preparation of the kaolin

Table 1. *GC-FID and GC-MS analysis: major components in Xylopia Aethiopica fruit essential oil.*

	Components	Structure	KI	%Area
1	α-thujene		936	2.22
2	α-pinene		946	11.10
3	Sabinene		987	23.90
4	β-pinene		992	27.90
5	α-terpinene		1036	2.22
6	β-phellandrene		1043	15.91
7	1,8-cineol (a) + limonene (b)		1045	3.25
8	Terpinen-4-ol		1192	5.13
9	Germacrene D		1508	3.54

The kaolin was collected from Wak locality (Adamaoua province of Cameroon) deposit. Stones and other heavy particles were removed from the samples, which were then crushed, ground by a Retsch Bioblock crushing BB1A type and sieved through a 230-mesh sieve. One part of the powder was simply passed through the 100 µm sieve (F1) and 50 µm (F2). A second part was kept dispersed in double distilled water for several hours before the mixture was sieved through a 50-mesh sieve. A small amount of 30% hydrogen peroxide was added to remove organic substances. The mixture was kept standing overnight and after decanting the clear solution, more water was added, stirred, and allowed to settle for 3-4 h. The clear solution was again decanted and the process was repeated several times to remove excess hydrogen peroxide. The separated kaolin was dried at 343 K (F3; diameter <50 µm purified clay). Finally, a last part of dispersed kaolin was fractionated by settling and decantation, using Stock's law to obtained the fraction <2 µm (F4); which was also dried at 343 K.

The kaolin was characterized by X-ray diffraction (XRD), X-ray fluorescence (XRF), Fourier transform IR spectroscopy (FTIR), and specific surface area determination by the BET method.

Table 2. *X-ray fluorescence analysis: oxide composition (%) of kaolin from Wak.*

Fractions	LOI	SiO_2	Al_2O_3	Fe_2O_3	CaO	MgO	Na_2O	K_2O	SO_3
F4	13.98	47.81	22.01	11.65	0.56	2.04	0.33	1.54	0.05
F3	10.18	54.22	20.15	9.22	0.53	1.70	0.46	3.43	0.05
F2	11.26	56.12	19.72	7.07	2.54	0.62	0.35	2.30	0.05
Fl	11.56	53.04	20.11	9.90	0.50	1.66	0.42	2.73	0.05

LOI: Loss on ignition.

2.2.1. Adsorption studies

Adsorption of essential oil by the different kaolin fractions was carried out in batch. Increasing amounts of clay fractions (0.1 ; 0.2; 0.3; 0.4; 0.5; 0.6;0.7;0.8; 0.9; 1.0 g) were dispersed in 2 ml oil solution (0.75 ml of oil in 10 ml of acetone); and equilibrated in an overhead shaker at room temperature (30 °C) for 3 h. In all cases, adsorption equilibrium was reached within 2 h. The particles were allowed to settle and separated by centrifugation. The oil concentration in the supernatant was determined by GC-FID.

2.2.2. Adsorption isotherms

Adsorption isotherms were determined using the peak area of each component of the essential oils. The adsorbed percentage was calculated as:

$$\%\text{adsorbed} = \frac{100(A_0 - A_x)}{A_0}$$

where A_0 is the peak area of each oil component in the initial solution; A_x the peak area of each component in the supernatant. The adsorbed amount per gram is:

$$\frac{X}{m} = \left[\frac{100(A_0 - A_x)}{A_0}\right] * \frac{(m_{eo})}{m_{ad}} * \%C$$

where x is the mass of each component adsorbed (mg), m is the mass of adsorbent (g), m_{eo} is the mass of essential oil (mg), m_{ad} is the mass of adsorbent in the solution (g), %C is the concentration (%) of each terpenic component in the crude essential oil. The equilibrium concentration was calculated as:

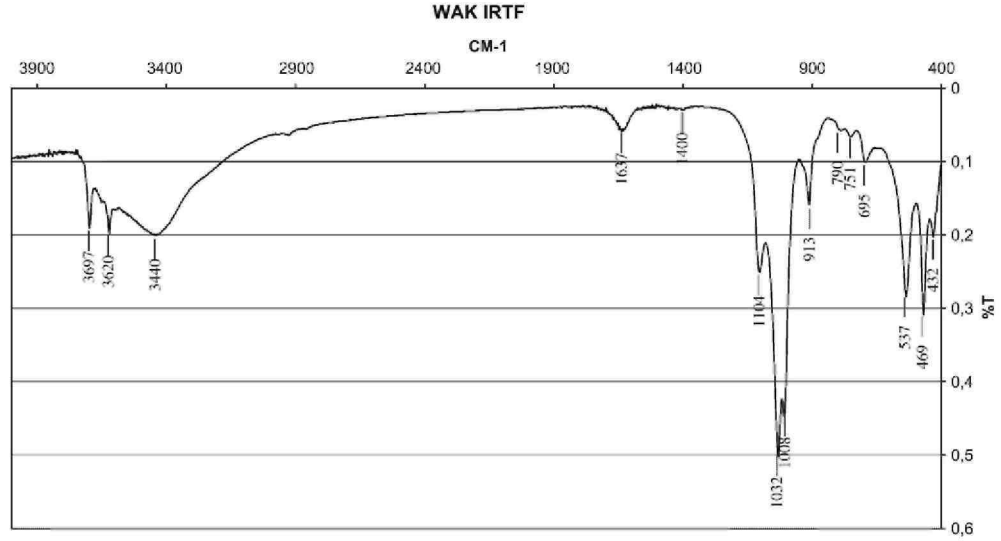
$$C_e = \frac{A_x}{A_0} * \frac{m_{eo}}{V_{eo}} * \%C$$ With V_{eo} the volume of solution (ml).

3. Results and discussion

Chromatographic analysis showed that X. *aethiopica* essential oil contains more than 50 components (Ngamo et al., 2001). Specific density of X. *aethiopica* oil was 0.87 mg/ml. For adsorption studies, 10 major compounds of X. *aethiopica* essential oil were selected (Table 1) : monoterpene hydrocarbons, MH, (α-thujene, α-pinene,

sabinene, β-pinene, α-terpinene, β-phellandrene, limonene), oxygenated monoter-penes OM (terpinen-4-ol, 1,8-cineole), sesquiterpene hydrocarbon ST, (Germacrene D). 1,8-cineole and limonene were not well separated by chromatography and will be considered together in the adsorption study.

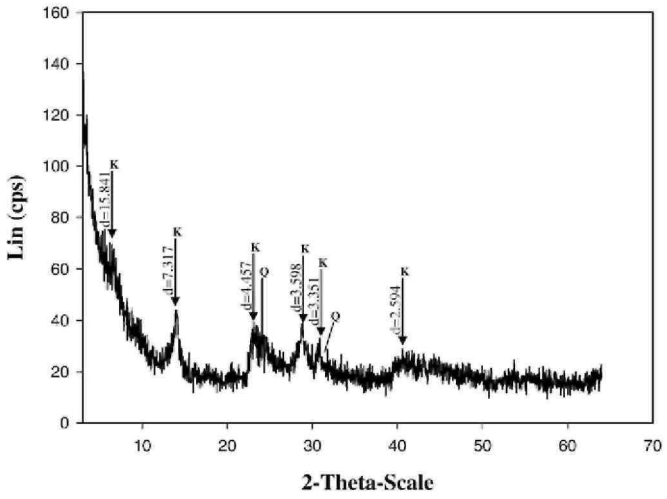
Fig. 1. *FTIR spectra of kaolin (F4).*



3.1. Characterization of the kaolin

The kaolin was rich in SiO_2 (~47.81%), in Fe_2O_3 (-11.65%); and in Al_2O_3 (~22.01%) and contained only small amounts of Ca^{2+}, Mg^{2+}, Na^{+} and K^{+} ions (Table 2). Loss on ignition (LOI) was 13.98%. The chemical composition indicated the presence of considerable amounts of silica-and iron-bearing impurities. The FTIR spectrum (Fig. 1) showed peaks at 3697, 3620, 3440, 1104, 1033, 913, 751 cm^{-1}, characteristics of kaolinite. A weak band at about 1412 cm^{-1} indicated presence of carbonate minerals. A shoulder at ~1104 cm^{-1} indicated quartz. The XRD pattern (Fig. 2) showed sharp reflections at d = 7.31; 4.45; 3.59; 3.35; 2.59 due to kaolinite and at d = 3.34, and 4.25 due to quartz. The specific surface area was 77 m^2/g.

Fig. 2. *DRX of kaolin (F4).*



3.2. Adsorption isotherms

Adsorption isotherms of *X. aethiopica* components are shown in Figs. 3, 4, 5 and 6. Adsorption decreased in the order sabinene >β-pinene>α-pinene>β-phellandrene for fractions F1, F2 F4; and in the order sabinene>β-pinene>β-phellandrene>α-pinene for F3 (group 1). Adsorption of the others majors components (α-thujene, α-terpinene, 1,8-cineole + limonene, terpinen-4-ol and germacrene D) changed with the particle size. The adsorption within group 2, decreased in the order terpinen-4-ol>germacrene D>1.8-cineole + limonene>α-thujene>α-terpinene for Fl; germacrene D>1.8-cineole + limonene> terpinen-4-ol>α-thujene>α-terpinene for F2; terpinen-4-ol>germacrene D>α-thujene>1.8-cineole + limonene>α-terpinene for F3 and terpinen-4-ol> 1.8-cineole + limonene>α-thujene>germacrene D>α-terpinene for F4. This adsorption amount of terpenics components between Fl, F2, F3 and F4 could be the result of many factors. The selectivity was affected by the abundance of each component in the crude essential oil; sabinene, β-pinene, β-phellandrene and α-pinene which are the most adsorbed were the most abundant compounds (23.90%, 27.90%, 15.91% and 11.10% respectively), while α-terpinene (2.22%) was the less adsorbed in all case. The selectivity of adsorption was affected by the polarity of terpenic components; germacrene D and terpinen-4-ol were adsorbed in larger amounts than some monoterpene hydrocarbons.

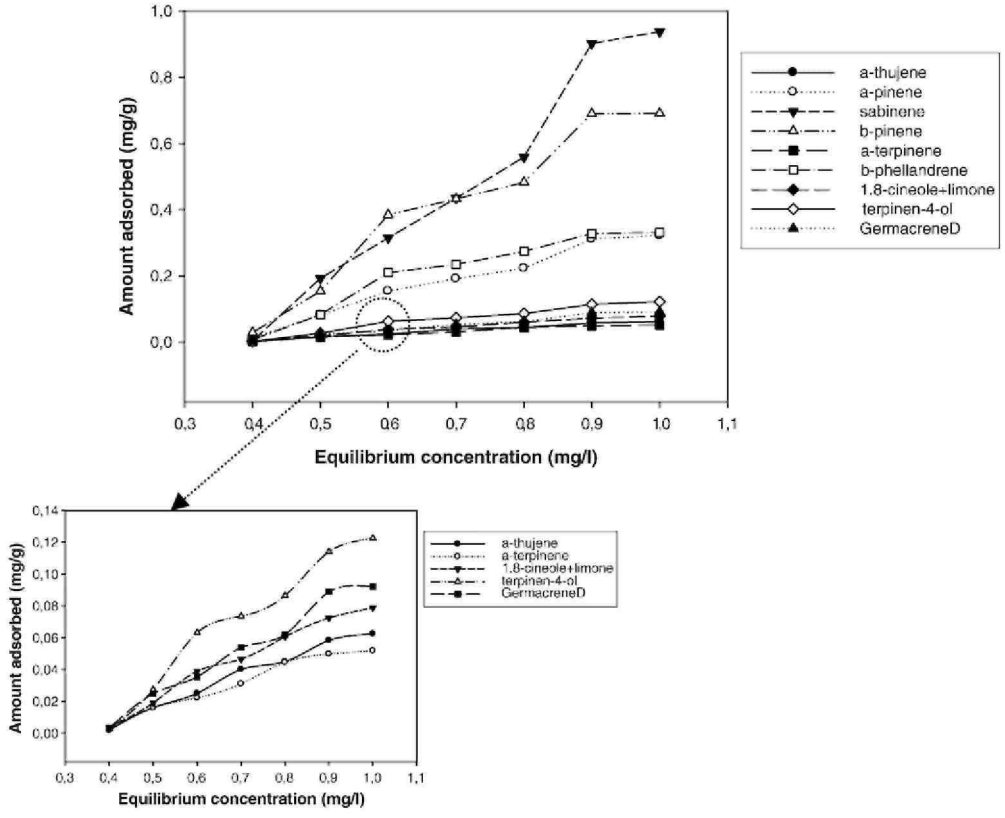
The isotherms are of L-type according to the classification of Giles et al. (1960). The amount of essential oil components adsorbed was inversely proportional to the adsorbent particle size (about 0.94 mg/g for F1; 1.13 mg/g for F2; 1.48 mg/g for F3 and 1.64 mg/g for F4). The changes can be explained by the fact that the content of kaolinite in the smaller fractions was higher for the same particle size fractions. The purified kaolin adsorbed higher amounts (F3>F2).

3.3. Fitting of adsorption isotherms

The adsorption isotherms could be well fitted by the Langmuir model (Table 3). Sabinene showed the highest amounts adsorbed and a high affinity for each kaolin fraction (Figs. 3-6).

The values of Langmuir *b* constants in Table 3 representing the affinity to the adsorbent were more significant for all terpenics components. The Langmuir value *a* indicated an effective adsorption of oil components (range from 1.26 to 40.30 dm^3/g). The R_L values for all particles size ($0<R_L<1$), revealed favourable adsorption of the oil compounds on the kaolin.

Fig. 3. *Adsorption isotherms of Xylopia aethiopica major components on the F1 fraction.*



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Considering some individual components, F4 presented the highest adsorption capacity for β-pinene (MO) with b = 1.79± 0.43 mg g^{-1} and a = 2.36 ± 0.58 $dm^3 g^{-1}$; b= 023 ± 0.08 mg g^{-1} andα = 22.07 ± 3.45 dm^3 g^{-1} for terpinene-4-ol and b = 0.11 ± 0.05 mg g^{-1} a =40.30 ±5.01 dm^3 g^{-1} for germacreneD.

Fig. 4. *Adsorption isotherms of Xylopia aethiopica major components on the F2 fraction.*

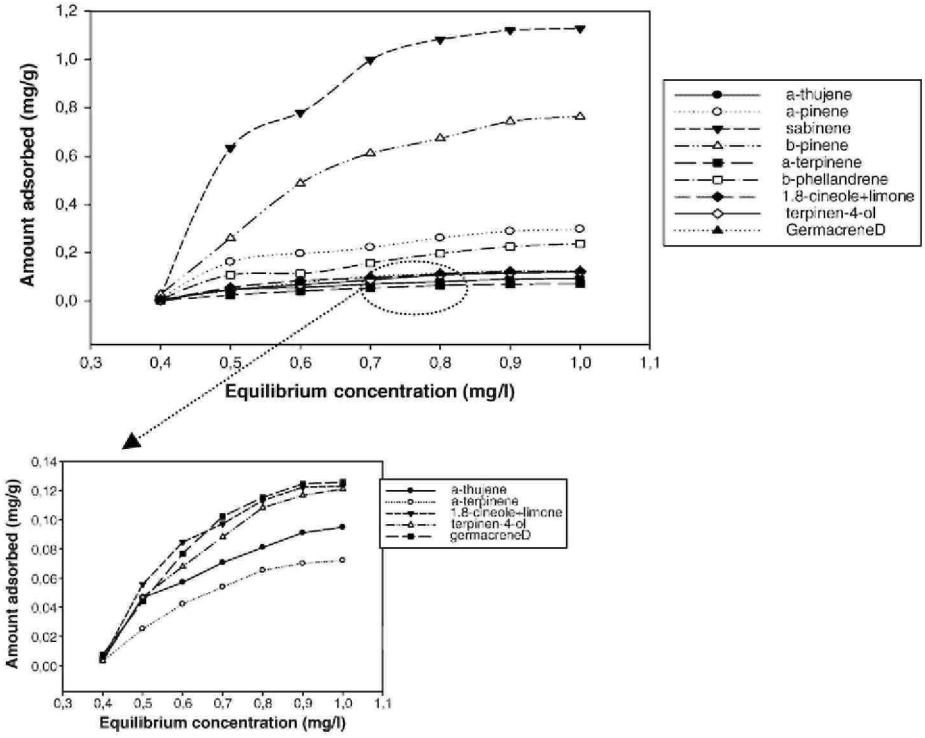
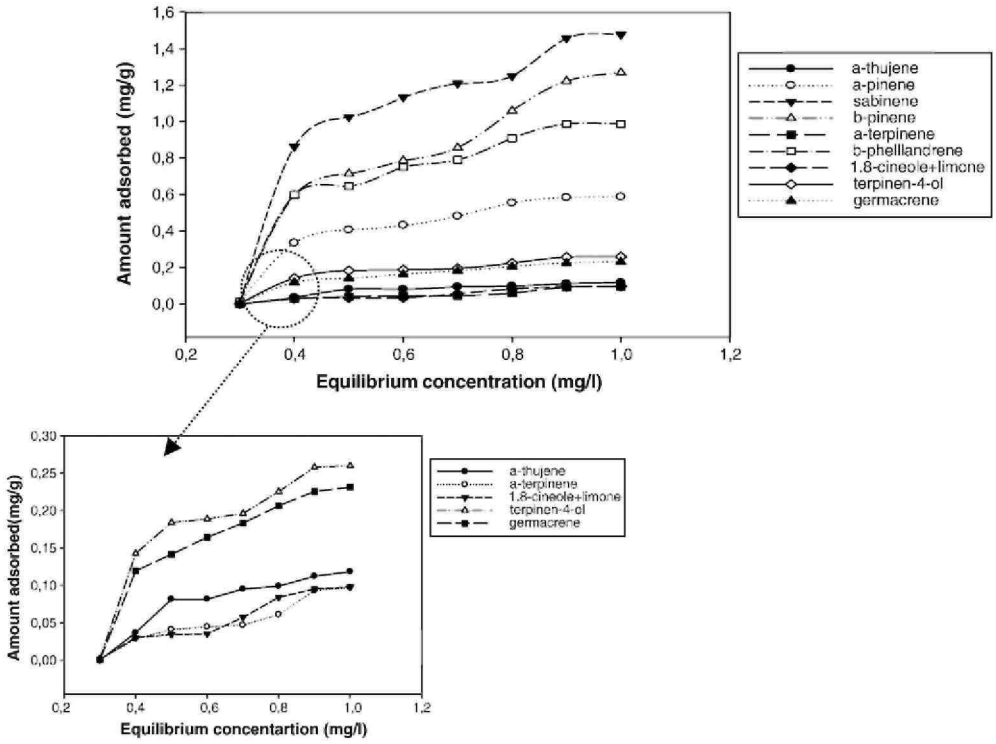


Fig. 5. *Adsorption isotherms of Xylopia aethiopica major components on the F3 fraction.*



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F3 fraction which was the most appropriated for an insecticidal formulation, presented a high adsorption capacity for sabinene with b = 1.70 ± 0.90 mgg^{-1} and a = 1.26 ± 0.91dm^{3}g^{-1} ; b = 0.21 ± 0.0l mgg^{-1} and a = 11.27 ± 1.51 dm^3 g^{-1} b = 0.11 ± 0.05 mg g^{-1} and a = 14.22 ± 2.05dm^3 g^{-1}.

In the crude *X. aethiopica* essential oil, β-pinene concentration is higher than sabinene (27.90 and 23.90% respectively). However, the adsorption capacity of these components was not statistically different: 1.35 ±0.14 dm^3 g^{-1} (β-pinene) and 1.26 ±0.07 dm^3 g^{-1} (sabinene) for F4; 1.33 ±0.09 dm^3 g^{-1} and 1.42 ±0.07 dm^3 g^{-1} (F3).

Fig. 6. *Adsorption isotherms of Xylopia aethiopica major components on the F4 fraction.*

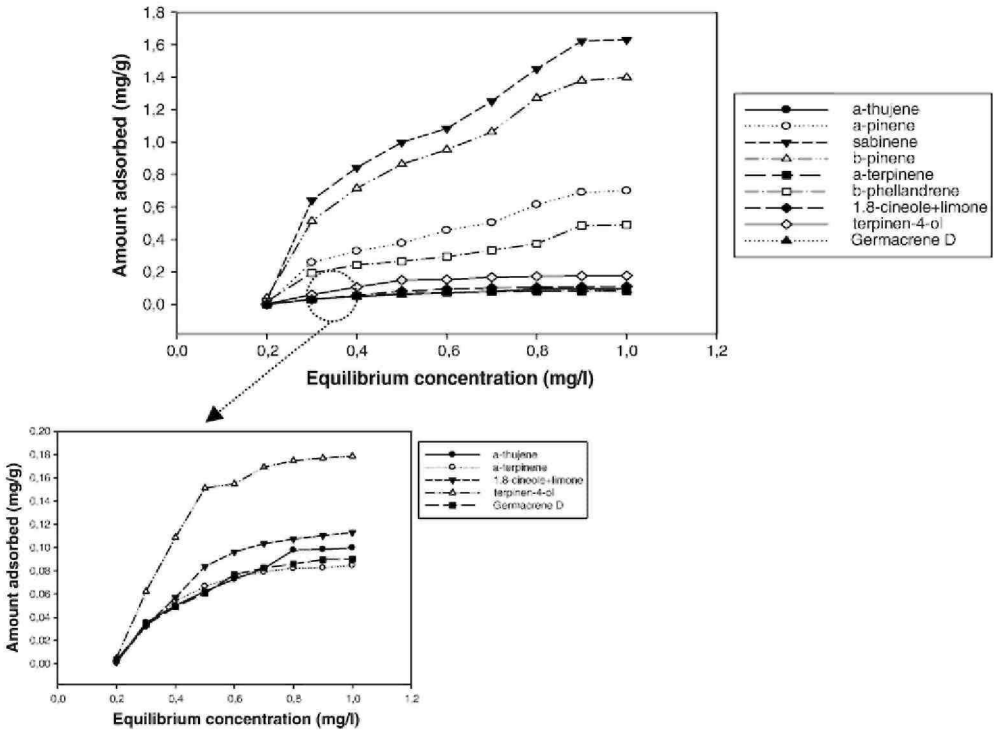

Table 3. *Langmuir adsorption constants of Xylopia aethiopica essential oil on clay.*

Fractions	**Fl**			**F2**			**F3**			**F4**		
Constants	a	b	R_L	a	b	R_L	a	b	R_L	a	b	R_L
Compounds	(dm^{3}g^{-1})	(mgg^{-1})		(dm^{3}g^{-1})	(mgg^{-1})		(dm^{3}g^{-1})	(mg g^{-1})		(dm^{3}g^{-1})	(mg g^{-1})	
α-thujene	21.80 ± 1.10	0.11 ± 0.07	0.42	22.44 ± 6.10	0.13 ± 0.07	0.31	20.10 ± 7.51	0.17 ± 0.04	0.29	3.05 ± 0.98	1.53 ± 0.06	0.77
α-pinene	4.23 ± 0.05	0.55 ± 0.10	0.42	5.10 ± 1.80	0.60 ± 0.13	0.40	4.50 ± 1.02	0.82 ± 0.07	0.27	4.40 ± 0.75	0.90 ± 0.04	0.25
Sabinene	1.26 ± 0.91	1.70 ± 0.90	0.45	1.33 ± 0.25	2.12 ± 1.24	0.40	1.84 ± 0.78	2.02 ± 082	0.27	2.02 ± 1.01	2.10 ± 0.31	0.23
β-pinene	2.01 ± 0.41	1.20 ± 0.04	0.42	2.46 ± 0.19	1.34 ± 0.79	0.34	1.81 ± 0.07	1.79 ± 0.14	0.30	2.36 ± 0.58	1.79 ± 0.43	0.23
α-terpinene	26.30 ± 3.14	0.09 ± 0.01	0.42	29.10 ± 4.89	0.11 ± 0.07	0.32	1.51 ± 0.19	2.70 ± 0.49	0.87	2.01 ± 0.04	2.09 ± 0.97	0.85
β-phellandrene	4.34 ± 0.21	0.56 ± 0.17	0.41	8.00 ± 2.71	0.41 ± 0.01	0.34	2.70 ± 0.34	1.37 ± 0.07	0.27	6.25 ± 0.98	0.62 ± 0.24	0.24
1,8-cineole + limonene	17.86 ± 1.70	0.13 ± 0.07	0.41	16.90 ± 3.74	0.18 ± 0.04	0.32	17.58 ± 3.12	0.15 ± 0.08	0.36	26.45 ± 2.31	0.15 ± 0.07	0.25
Terpinen-4-ol	11.27 ± 1.51	0.21 ± 0.01	0.42	12.94 ± 3.02	0.24 ± 0.10	0.39	10.05 ± 4.21	0.35 ± 0.01	0.27	22.07 ± 3.45	0.23 ± 0.08	0.20
Germacrene D	14.22 ± 2.05	0.16 ± 0.05	0.43	14.60 ± 5.21	0.21 ± 0.08	0.35	10.84 ± 3.48	0.32 ± 0.12	0.28	40.30 ± 5.01	0.11 ± 0.05	0.21

4. Conclusion

The terpenes sabinene, β-pinene, α-pinene and β-phellandrene were well adsorbed by the four kaolin fractions. The sesquiterpene germacrene D was adsorbed in higher amounts than some monoterpenes.

The adsorption capacity of the kaolin depended on the particle size fractions the finer fractions adsorbed higher amounts. Treatment of kaolinite with hydrogen peroxide improved adsorption capacity. The adsorption isotherm of the terpenic compound on Wak kaolin was fitted with the Langmuir model.

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References

Borras, J.B. (1999). Etude des andosols d'Olot. Cahier ORSTOM. Vol XIV. n°2, pages: 94-111.

Giles, C.H., MacEwan, T.H., Nakhwa, S.N., Smith, D., 1960. Studies in adsorption: part 11. A system of classification of solution adsorption isotherm, and its use in diagnosis of adsorption mechanisms and in measurement of specific surface areas of solids. Journal of the Chemical Society 3973-3993.

Ghosh, D., Krishna, G.B., 2001. Adsorption of methylene blue on kaolinite. Journal of Applied Clay Science 20, 295-300.

Keita, M.S., Vincent, C., Schmit, J.-P., Arnason, T.J., Bélanger, A., 2001. Efficacity 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 Products Research 37, 339-349.

Lagaly, G., 2001. Pesticide-clay interactions and formulations. Applied Clay Science 18, 205-209.

Lajide, L., Escoubas, P., Mitzutani, J., 1995. Termite antifeedant activity in *Xylopia aethiopica.* Phytochemistry 40 (4), 1105-1112.

Ngamo, T.L.S., Ngassoum, M.B., Jirovertz, L., Ousman, A., Nukenine, E., Moukala, O.E., 2001. Protection of Stored Maize against *Sitophilus zéamaïs* (Motsch) by use of essential oils of spices from Cameroon. Medlinden Faculteit Landbouww Universiteit Gent 66/2a, 473-478.

Ngamo Tinkeu, L.S., (2004). A la recherche d'une alternative aux Polluants Organiques Persistants utilisés pour la protection des végétaux. Bulletin d'informations phytosanitaires. N843 Avril-Juin 2004. 23 p.

Ngamo Tinkeu, L.S., Goudoum, A., Ngassoum, M.B., Mapongmetsem, Lognay, G., Malaisse, F., Hance, T., 2007. Chronic toxicity of essential oils of 3 local aromatic plants towards *Sitophilus zeamais* Motsch. (Coleoptera: Curculionidae). African Journal of Agricultural Research 2 (4), 164-167.

Prates, H.T., Santos, J.P., Waquil, J.M., Fabris, J.D., Oliverta, A.B., Foster, J.E., 1998. Insecticidal Activity of Monoterpen Against *Rhyzopertha dominica* (F) and *Tribolium castaneum* (Herbst). Journal of Stored Products Research 34 (4), 243-249.

Regnault-Roger, C., Philogene, B.J.R., Vincent, C., 2002. Biopesticides d'origine végétale. Lavoisier Tec & Doc, Paris, pp. 19-39.

Szafranski, F., Bloszyk, E., Drozdz, B., 1991. Activité biologique des extraits de quelques plantes des environs de Kisangani (Zaïre). Belgian Journal Of Botany 124 (1), 60-70.