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## Chemical Composition and Bioactivity of Four Plant Essential Oils from Nigeria against *Macrotermes subhyalinus* (Rambur)

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## Authors' contributions

This work was carried out in collaboration between all authors. All authors read and approved the final manuscript.

## Article Information

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## ABSTRACT

The present study was carried out to determine the anti-termitic activity of four plants essential oils (*Monondora myristica* seed, *Piper umbellatum* seed, *Pinus sylvestry* leaves and *Piper nigrum* seed) from Nigeria against *Macrotermes subhyalinus* (Rambus). The essential oils were extracted by steam distillation and graded into different concentrations (0.5, 1, 1.5, 2, 2.5 mL/L). The chemical composition of the oil was analyzed by GC-MS. The result of the contact and repellence test showed that, the activities of the essential oils against *Macrotermes subhyalinus* increases with dose and time. The results revealed that during contact test, all essential oils showed toxicity against *Macrotermes subhyalinus* but the effectiveness of the essential oils differs, the essential oil of *Monondora myristica* had the lowest contact activity for all doses administered,100% mortality of *M. subhyalinus* was observed with a dose of 0.5 mL/L of the essential oil after 10 minutes of exposure, while with the same dose, 100% mortality was observed with *P. nigrium* and *P. umbellatum* after 20 minutes and 5 minutes respectively. All the essential oils proved to be repellent against *Macrotermes subhyalinus* with the essential oil of *Monondora myristica* having the least repellency. From the GC-MS analysis, the major components of the essential oils of

Monondora myristica, Piper umbellatum, Pinus syvestry and Piper nigrum are trans-13octtadecenoic acid (25.18%), aromadendrene (13.74%),  $\alpha$ - terpineol (27.17%) and linalool (21.73%) respectively. Therefore, significant variation was observed in repellence, and contact activities between different doses and time of exposure (P<0.05). The result shows that the essential oils had termicite activity against *M. subhyalinus* and it was dose and time dependent.

Keywords: Essential oil; termite; repellence; mortality and components.

### **1. INTRODUCTION**

Termites are a large and diverse group of insects consisting of over 2600 species worldwide. Africa is by far the richest continent in termite diversity [1]. The species known to damage crops, trees, and rangeland belong to the family Termitidae. This family consists of four subfamilies: Macrotermitinae. Nasutitermitinae. Termitinae. and Apictotermitinae. It is estimated that, less than 20% of members of the family Termitidae are serious pests [2]. Over 90% of the termite damage in agriculture, forestry, and urban settings is attributed to members of the Macrotermitinae [2], which build the large mounds (hereafter called termitaria) that form the spectacular features of the African landscape [3,4].

It must be noted that, the pest activity is a part and parcel of the termite's beneficial role in various ecosystems [3,5]. *Macrotermitinae* collect up to 60% of the grass, woody material, and annual leaf fall to construct the fungus gardens in their nests [6].

The ongoing interest in sustainable agriculture and food security in Africa highlights the need for a more balanced approach to termite control and maintenance of their ecosystem services. To begin to address the mismatch between these two objectives, a holistic appraisal of the termite problem and opportunities for their sustainable management is needed. Sustainable termite management is defined as (1) control of the pest species without causing ecological damage and loss of the ecosystem services provided by termites, (2) conservation of the non-pest termite species, and (3) use of termites and associated resources without exhausting them [7].

Broad spectrum and organochloride insecticides have been largely relied upon for the control of termites [8]. The limitations associated with the application and efficacy of these chemicals such as the destruction of non-target insects, high cost of the chemical insecticides, accidental poisoning, accidental and environmental pollution emphazed the need for alternative methods [9]. Over the past decades, there has been an increase in effort to reduce the reliance on synthetic substances in the prevention of termite invasion due to its harmful effect on the environment and human. Botanical insecticides such as azadirachtin are often effective alternatives to organophosphates or other neurotoxins for pest control due to multiple modes of action, which includes toxicity, antifeedant and anti-oviposition effects [10,11]. Natural products containing secondary plant compounds such as terpenes, steroids, alkaloids, phenolics and cardiac glycosides [12], affect insect behavior and are toxic in some cases [13,14]. Identification of plant extracts that exhibit the above-described deleterious effects on pest insect physiology and behavior represents a potential alternative strategy for development of bio-rational controls that could replace synthetic neurotoxins.

In Africa the seeds of *Monondora myristica*, *Piper. umbellatum* and *Piper. nigrum* are usually used as spices and in some cases, they are used medicinally in the treatment of some ailments, *Pinus sylvestry* essential oil are used by farmers for medicinal purposes.

The present study was carried out to determine the chemical compositions and anti-termitic activities of essential oil of *M. myristica, P. umbellatum, P. syvestry* and *P. nigrum* against *M. subhyalinus* (Rambus) in relation to dose and exposure time.

### 2. MATERIALS AND METHODS

#### 2.1 Plant Material

African Nutmeg (*M. myristica*) seeds and *P. umbellatum* seeds were obtained from Orisumbare Market in Osogbo, Osun state, Nigeria. Pine needle (*P. syvestry*) were obtained from its tree at Awo Hall, Obafemi Awolowo University Ile-ife, Osun State, Nigeria. Black pepper seed (*P. nigrum*) was bought from Agbalata Market Badagary, Lagos State Nigeria.

The seeds and plant were identified at Federal Research Institute of Nigeria (FRIN), Ibadan, Nigeria. The seeds were air-dried and ground to powder.

### **2.2 Termites Collection**

Secondary nests of *M. subhyalinus* (Rambus) were collected from termite moulds at Osun State University, Osogbo Campus and placed in black garbage bags. The nests were immediately transported to the laboratory and placed inside 100 liter plastic containers with lids and kept in a room at 25°C for three days. Distilled water was sprayed on the sides of the container to keep the relative humidity above 80%.

#### 2.3 Essential oil Distillation

The ground powder (10 g) of seeds and pine needles were subjected to hydro distillation using a modified Clevenger-type apparatus for 6 hours, anhydrous sodium sulphate was used to remove water after extraction. Essential oils were stored in airtight containers in a refrigerator at  $4^{\circ}$ C.

#### 2.4 Contact Effect

The Contact effect of the essential oils against M. subhyalinus was evaluated on filter paper disc by treating a whatman No. 1 filter paper with the essential oils diluted in 100% acetone. A micropipette was used to suck out 1 µL, 2 µL, 3 µL, 4 µL and 5 µL of the essential oil and was diluted with 2 mL of acetone to form concentrations of 0.5 mL/L, 1 mL/L, 1.5 mL/L, 2 mL/L and 2.5 mL/L respectively. They were each poured and allowed to flow regularly on a disc of filter paper placed in a petri dish. The solvent was allowed to dry for 60 seconds after which 10 M. subhyalinus were introduced into the petri dish and then closed. Percentage mortality of M. subhyalinus was observed every minute. Each experiment was conducted in triplicate. Control experiment was done with the same setup using only acetone.

### 2.5 Repellent Effect

The repellent effects of the essential oils against *M. subhyalinus* were evaluated using the area preference method. Tested areas consisting of Whatman No. 1 filter paper cut in half. 1  $\mu$ L, 2  $\mu$ L, 3  $\mu$ L, 4  $\mu$ L and 5  $\mu$ L of the essential oils were diluted with 2 mL of acetone to form concentrations of 0.5 mL/L, 1 mL/L, 1.5 mL/L,

2 mL/L and 2.5 mL/L respectively. Full discs were subsquently remade by attaching treated halves to untreated halves with clear adhesive tape. 10 *M. subhyalinus* were released at the center of the filter paper disc and the Petri dishes were subsequently covered and oberservations were made. Each experiment was conducted in triplicate. Percentage repellency (PR) values were calculated as follows:

PR = [ ( Nc - Nt ) / Nc ]100%

Nc- Total number of insects introduced Nt- Number of insects in the treated

#### 2.6 GC-MS Analysis

Gas chromatography and mass spectrometry analysis was performed on an Agilent 6890 N instrument equipped with a flame ionization detector and HP-5MS (30 m × 0.25 mm × 0.25 µm) capillary column, while the essential oil components were identified on an Agilent Technologies 5973 N mass spectrometer. The GC settings were as follows: the initial oven temperature was held at 60°C for 1 min and ramped at 10℃ min<sup>-1</sup> to 180℃ for 1 min, and then ramped at 20℃ min-1 to 280℃ for 15 min. The injector temperature was maintained at 270°C. The samples (1 µL) were injected neat, with a split ratio of 1:10. The carrier gas was helium at flow rate of 1.0 mL min<sup>-1</sup>. Spectra were scanned from 20 to 550 m/z at 2 scans s<sup>-1</sup>. Most constituents were identified bv gas chromatography by comparison of their retention indices with those of the literature or with those of authentic compounds available in our laboratories. The retention indices were determined in relation to a homologous series of n-alkanes ( $C_8-C_{24}$ ) under the same operating conditions. Further identification was made by comparison of their mass spectra on both columns with those stored in NIST 05 and Wiley 275 libraries or with mass spectra from literature. Component relative percentages were calculated based on GC peak areas without using correction factors.

### 2.7 Statistical Analysis

The data were corrected using Abbott's formula [15] for the mortalities and the data were subjected to probit analyses using SPSS (2001) for Windows to estimate  $LD_{50}$  and  $EC_{50}$  values of the essential oils against *Macrotermes subhyalinus*. Percentage mortality values for different exposure times were subjected to

analysis of variance (one-way ANOVA) using the same statistical program (SPSS 2001) for probit analysis.

### 3. RESULTS AND DISCUSSION

The yield of the essential oils is *M. myristica* (1.2%), *P. umbellatum* (0.13%), *P. sylvestry* (0.21%) and *P. nigrum* (0.26%).

The comparative effectiveness of the contact activities of the four essential oils against *M. subhyalinus* at various doses and times of exposure are presented in Tables 1-4. On exposure of *M. subhyalinus* to the essential oil of

*P. umbellatum* (Table 2), no toxicity was observed in the first minute of the experiment for all doses, but toxicity was recorded in the first minute of the bioassay when *M. subhyalinus* were exposed to the essential oil of *P. syvestry* (Table 4). In the study, when *M. subhyalinus* were exposed to a dose of 0.5 ml/l of essential oils, the essential oils of *P. umbellatum* and *P. syvestry* were found to be more toxic in the contact treatment; both essential oils recorded 100% mortality of *M. subhyalinus* within five minutes. Essential oil of *M. myristica* had the lowest contact activity (Table 3), 100% mortality of *M. subhyalinus* was observed after 90 minutes of exposure time.

# Table 1. Percentage contact mortality of Macrotermes subhyalinus treated with Piper nigrum essential oil

Exposure		Dose (mL/L)						
time (min)	0.5	1	1.5	2	2.5	Control		
4	0.0±0.0 <sup>a</sup>	0.0±0.0 <sup>a</sup>	0.0±0.0 <sup>a</sup>	0.0±0.0 <sup>a</sup>	40.0±0.50 <sup>c</sup>	$0.0\pm0.0^{a}$		
8	$0.0\pm0.0^{a}$	20.0±0.39 <sup>b</sup>	60.0±0.30 <sup>d</sup>	80.0±0.36 <sup>e</sup>	100.0±0.0 <sup>f</sup>	$0.0\pm0.0^{a}$		
12	20.0±1.39 <sup>b</sup>	80.0±0.36 <sup>d</sup>	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>	$0.0 \pm 0.0^{a}$		
16	40.0±0.50 <sup>bc</sup>	100.0±0.0 <sup>e</sup>	100.0±0 0 <sup>e</sup>	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>	$0.0\pm0.0^{a}$		
20	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>	$0.0\pm0.0^{a}$		

The result shows the mean  $\pm$  SD of three replicates. Data within a row followed by the same letter are not significantly different at P < 0.05

## Table 2. Percentage contact mortality of Macrotermes subhyalinus treated with Piper umbellatum essential oil

Exposure	Dose (mL/L)						
time(min)	0.5	1	1.5	2	2.5	Control	
1	$0.0\pm0.0^{a}$	0.0±0.0 <sup>a</sup>	0.0±0.0 <sup>a</sup>	0.0±0.0 <sup>a</sup>	0.0±0.0 <sup>a</sup>	$0.0\pm0.0^{a}$	
2	40.0±0.50 <sup>b</sup>	40.0±0.50 <sup>b</sup>	60.0±0.30 <sup>c</sup>	60.0±0.30 <sup>c</sup>	60.0±0.30 <sup>c</sup>	$0.0\pm0.0^{a}$	
3	40.0±0.50 <sup>c</sup>	80.0±0.36 <sup>d</sup>	80.0±0.36 <sup>d</sup>	80.0±0.36 <sup>d</sup>	100.0±0.0 <sup>e</sup>	$0.0\pm0.0^{a}$	
4	80.0±0.36 <sup>d</sup>	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>	$0.0\pm0.0^{a}$	
5	100.0±0.0 <sup>e</sup>	$0.0\pm0.0^{a}$					

The result shows the mean  $\pm$  SD of three replicates. Data within a row followed by the same letter are not significantly different at P < 0.05

#### Table 3. Percentage contact mortality of Macrotermes subhyalinus treated with Monodora myristica essential oil

Exposure time (min)			Dose	e (mL/L)		
	0.5	1	1.5	2	2.5	Control
10	0.0±0.00 <sup>a</sup>	0.0±0.00 <sup>a</sup>	$0.0\pm0.00^{a}$	$0.0\pm0.00^{a}$	0.0±0.00 <sup>a</sup>	0.0±0.00 <sup>a</sup>
20	$0.0\pm0.00^{a}$	0.0±0.00 <sup>a</sup>	$0.0\pm0.00^{a}$	$0.0\pm0.00^{a}$	0.0±0.00 <sup>a</sup>	$0.0\pm0.00^{a}$
30	$0.0\pm0.00^{a}$	0.0±0.00 <sup>a</sup>	$0.0\pm0.00^{a}$	20±1.40 <sup>b</sup>	60±2.30 <sup>c</sup>	$0.0\pm0.00^{a}$
40	$0.0\pm0.00^{a}$	0.0±0.00 <sup>a</sup>	$0.0\pm0.00^{a}$	40±2.40 <sup>b</sup>	100±0.00 <sup>e</sup>	$0.0\pm0.00^{a}$
50	20±1.30 <sup>b</sup>	40±2.30 <sup>bc</sup>	60±1.30 <sup>c</sup>	60±3.30 <sup>c</sup>	100±0.00 <sup>e</sup>	0.0±0.00 <sup>a</sup>
60	40±2.33 <sup>c</sup>	80±1.60 <sup>d</sup>	80±0.00 <sup>d</sup>	100±0.00 <sup>e</sup>	100±0.00 <sup>e</sup>	$0.0\pm0.00^{a}$
70	60±1.33 <sup>°</sup>	100±0.00 <sup>e</sup>	100±0.00 <sup>e</sup>	100±0.00 <sup>e</sup>	100±0.00 <sup>e</sup>	$0.0\pm0.00^{a}$
80	80±0.00 <sup>d</sup>	100±0.00 <sup>e</sup>	100±0.00 <sup>e</sup>	100±0.00 <sup>e</sup>	100±0.00 <sup>e</sup>	0.0±0.00 <sup>a</sup>
90	100±0.00 <sup>e</sup>	$0.0\pm0.00^{a}$				

The result shows the mean  $\pm$  SD of three replicates. Data within a row followed by the same letter are not significantly different at P < 0.05

The results of the contact test revealed an increase in the mortality of *M. subhyalinus* with increase in the dosage of the essential oils and period of exposure. *M. subhyalinus* were significantly susceptible to the toxicity of the essential oils at different dose and exposure time at (P<0.05) compared to the control.

The essential oils from Ρ. nigrium, P. umbellatum, M. myristica and P. sylvestry exhibited potent repellent activity against adults M. subhyalinus (Tables 5, 6, 7 and 8). The repellency values of the essential oils significantly increased with concentration and exposure time when compared to the control. From Table VI and VIII, the essential oils of P. umbellatum and P. sylvestry showed the strongest repellent activity against M. subhyalinus adults. The use of the lowest dose of 0.5ml/l of the essential oils of P. umbellatum and P. syvestry recorded 100% repellency of M. subhyalinus after 5 minutes of exposure time. The essential of *M. myristica* recorded the lowest repellent toxicity against M. subhyalinus (Table 7). Repellent activity of the essential oils against M. subhyalinus adults gradually increased in a dose and time dependent manner at (P < 0.05).

Table 9 shows the major constituent of *P. umbellatum, P. syvestry, P. nigrium* and *M. myristica.* From the GC-MS analysis of the essential oils, it is shown that Aromadendrene

(13.74%), Caryophyllene (10.44%), y-Bisabolene (8.82%) and Linalool (8.55%) are the components with the highest values found in P. umbellatum. Trans-13-octadecanoic acid with a value of 25.18% Sabinol-cis (17.87%) and Linalool (9.11%) are the major components found in M. myristica, P. nigrium has Linalool as the main constituent (21.73%) and  $\alpha$  Terpineol is the highest constituent detected in P. sylvestry. The differences in the effectiveness of these essential oils can be attributed to the differences in the constituent of the essential oils. The termicidal activity of many plant essential oils might be attributed to monoterpenoids. Due to the high volatility, they might be of importance for controlling insects. Monoterpenoids were reported earlier as fumigants and contact toxicants on various insect pests [16].

There is a renewed interest amongst scientists to study the bioactivity of plant essential oils against phytophagous arthropod pests [17,18]. The high degree of damages caused by termites in Africa has made it important to look out for alternatives that are cheap and readily available for the control of termites. This is especially relevant in African local settings where poor status of the people do not allow them to have access to the synthetic chemical controls and where available, to be replaced with less toxic ones. Hence, the need to use local remedies to combat the menace of termites.

 Table 4. Percentage contact mortality of Macrotermes subhyalinus treated with

 Pinus sylvestris essential oil

Exposure	e Dose (mL/L)						
time (min)	0.5	1	1.5	2	2.5	Control	
1	20±1.30 <sup>b</sup>	40±3.10 <sup>bc</sup>	40±1.40 <sup>c</sup>	40±1.20 <sup>c</sup>	40±0.00 <sup>c</sup>	0.0±0.00 <sup>a</sup>	
2	20±2.40 <sup>b</sup>	40±1.30 <sup>b</sup>	40±1.20 <sup>b</sup>	60±2.30 <sup>c</sup>	60±0.00 <sup>c</sup>	0.0±0.00 <sup>a</sup>	
3	60±1.69 <sup>c</sup>	60±1.20 <sup>c</sup>	60±2.30 <sup>c</sup>	80±0.00 <sup>d</sup>	80±0.00 <sup>d</sup>	0.0±0.00 <sup>a</sup>	
4	80±1.40 <sup>d</sup>	80±3.00 <sup>d</sup>	80±1.70 <sup>d</sup>	80±0.00 <sup>d</sup>	100±0.00 <sup>e</sup>	0.0±0.00 <sup>a</sup>	
5	100±0.00 <sup>e</sup>	$0.0\pm0.00^{a}$					

The result shows the mean  $\pm$  SD of three replicates. Data within a row followed by the same letter are not significantly different at P < 0.05

Table 5. Percentage repellence activity of essential oil of Piper nigrium against
Macrotermes subhyalinus

Exposure			Dose (mL/L)		
time(min)	0.5	1	1.5	2	2.5
8	0.0±0.0 <sup>a</sup>	0.0±0.0 <sup>a</sup>	20.0±0.39 <sup>b</sup>	40.0±0.50 <sup>°</sup>	60.0±0.30 <sup>d</sup>
16	$0.0\pm0.0^{a}$	20.0±0.39 <sup>b</sup>	80.0±0.36 <sup>d</sup>	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>
24	20.0±0.39 <sup>a</sup>	60.0±0.30 <sup>c</sup>	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>
32	40.0±0.50 <sup>b</sup>	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>
40	100.0±0.0 <sup>a</sup>	100.0±0.0 <sup>ª</sup>	100.0±0.0 <sup>ª</sup>	100.0±0.0 <sup>ª</sup>	100.0±0.0 <sup>a</sup>

The result shows the mean  $\pm$  SD of three replicates. Data within a row followed by the same letter are not significantly different at P < 0.05

Exposure			Dose (mL/L)		
time(min)	0.5	1	1.5	2	2.5
1	0.0±0.0 <sup>a</sup>				
2	40.0±0.50 <sup>b</sup>	40.0±0.50 <sup>b</sup>	60.0±0.30 <sup>c</sup>	60.0±0.30 <sup>°</sup>	60.0±0.30 <sup>c</sup>
3	40.0±0.50 <sup>b</sup>	80.0±0.36 <sup>d</sup>	80.0±0.36 <sup>d</sup>	80.0±0.36 <sup>d</sup>	100.0±0.0 <sup>e</sup>
4	80.0±0.36 <sup>d</sup>	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>	100.0±0.0 <sup>e</sup>
5	100.0±0.0 <sup>e</sup>				

## Table 6. Percentage repellence activity of essential oil of Piper umbellatum against Macrotermes subhyalinus

The result shows the mean  $\pm$  SD of three replicates. Data within a row followed by the same letter are not significantly different at P < 0.05

# Table 7. Percentage repellence activity of essential oil of Monodora myristica against Macrotermes subhyalinus

Exposure	Dose (mL/L)						
time(min)	0.5	1	1.5	2	2.5		
10	$0.0\pm0.00^{a}$	$0.0\pm0.00^{a}$	0.0±0.00 <sup>a</sup>	0.0±0.00 <sup>a</sup>	$0.0\pm0.00^{a}$		
20	$0.0\pm0.00^{a}$	$0.0\pm0.00^{a}$	$0.0\pm0.00^{a}$	$0.0\pm0.00^{a}$	$0.0\pm0.00^{a}$		
30	20±0.00 <sup>b</sup>	40±0.00 <sup>b</sup>	$60\pm0.00^{\circ}$	$80 \pm 0.00^{d}$	100±0.00 <sup>e</sup>		
40	60±0.00 <sup>c</sup>	80±0.00 <sup>d</sup>	80±0.00 <sup>d</sup>	100±0.00 <sup>e</sup>	100±0.00 <sup>e</sup>		
50	100±0.00 <sup>e</sup>						

The result shows the mean  $\pm$  SD of three replicates. Data within a row followed by the same letter are not significantly different at P < 0.05

## Table 8. Percentage repellence activity of essential oil of Pinus sylvestris against Macrotermes subhyalinus

Exposure	Dose (mL/L)						
Time (min)	0.5	1	1.5	2	2.5		
1	20±0.00 <sup>b</sup>	20±0.00 <sup>b</sup>	20±0.00 <sup>b</sup>	40±0.00 <sup>b</sup>	40±0.00 <sup>b</sup>		
2	40±0.00 <sup>b</sup>	40±0.00 <sup>b</sup>	60±0.00 <sup>c</sup>	$60 \pm 0.00^{\circ}$	$60 \pm 0.00^{\circ}$		
3	40±0.00 <sup>b</sup>	$60 \pm 0.00^{\circ}$	$60\pm0.00^{\circ}$	$80 \pm 0.00^{d}$	$80 \pm 0.00^{d}$		
4	60±0.00 <sup>c</sup>	80±0.00 <sup>d</sup>	80±0.00 <sup>d</sup>	80±0.00 <sup>d</sup>	100±0.00 <sup>e</sup>		
5	100±0.00 <sup>e</sup>						

The result shows the mean  $\pm$  SD of three replicates. Data within a row followed by the same letter are not significantly different at P < 0.05

#### Table 9. Major constituents of four essential oils from Nigeria

Components		% In Sa	mple	
-	Piper umbellatum	African nutmeg	Piper nigrium	Pinus syvestry
Linalool	8.55	9.11	21.73	-
Caryophyllene	10.44	-	7.35	-
Aromadendrene	13.74	-	-	-
γ Bisabolene	8.82	-	8.75	-
Germacrene	6.56	-	-	-
Sabinol-cis	-	17.87	-	-
tr-13-octadecenoic acid	-	25.18	-	-
Palmitic acid	-	7.66	-	-
β Farnescene	-	-	6.12	-
Terpinen-4-ol	-	-	-	21.82
βTerpineol	-	-	-	14.07
Borneol	-	-	-	6.72
a Terpineol	-	-	-	27.17
Longifolene				5.45

Our study on the anti-termitic activity of four essential oils has shown that, the essential oils exhibited potent toxicity against *M. subhyalinus.* The activity of the essential oils against

*M. subhyalinus* was dose and time dependent at all levels of treatment. The essential oil of *P. sylvestry* was more toxic in the contact treatment against *M. subhyalinus* than the

essential oil of P. umbellatum as recorded by the lethal doses after 3 minutes of exposure, but the differences in the lethal dose was not significant (P<0.05). The LD<sub>50</sub> of P. syvestry and P. umbellatum are 0.363 ml/L and 0.596 ml/L respectively. The contact activity of the essential oils of P. syvestry and P. umbellatum against M. subhyalinus were significantly higher than the values obtained from the contact activity of the essential oils of P. nigrium and M. myristica. The result of the contact treatment of the essential oils of P. nigrium against M. subhyalinus with all doses administered showed the first sign of mortality (40%) after 4 minutes of exposure to a dose of 2.5 ml/L (highest in the study), while in the case of M. myristica, mortality was first observed (20%) 30 minutes after the M. subhyalinus had been exposed to a dose of 2.0 ml/L. Results reported in this study showed that all of the essential oils have toxicity on M. subhvalinus at all levels of treatment but varied with the exposure period and dose. This finding is similar with the observation of Chieng [19], who reported that fractions from the essential oil of leaves of Piper sarmentosum was tested against subterranean termite (Coptotermes sp.) and revealed that, mortality of termite increased with concentration and exposure time.. Carol and Vina [20] showed in their work that, the use of concentrated forms of 8 essential oils on R.Flavipes resulted in 100% mortality after 24 hours of exposure, while the diluent which is acetone shows no mortality.

Experiments were carried out to investigate whether the efficacy of the essential oils was purely contact in nature. From the repellency activity of the essential oils, it was established that the essential oils exhibited repellence activity against M. subhyalinus. P. syvestry recorded toxicity against M. subhyalinus with all dose in the first minute of the experiment, while P. umbellatum showed no toxicity, but 100% repellence was observed with all concentration within 5 minutes when both essential oils were exposed to M. subhyalinus. P. umbellatum showed higher toxicity against M. subhyalinus as revealed by the effective concentration obtained from probit analysis. EC<sub>50</sub> of *P. umbellatum* and P. sylvestry are 0.557 ml/L and 0.809 ml/L after 3 minutes, the lower value of EC<sub>50</sub> of P. umbellatum indicates its higher toxicity. M. subhyalinus was more tolerant to the essential oils of P. nigrium and M. myristica than those of P. umbellatum and P. sylvestry. This was noted by the longer exposure time required for M. subhyalinus to show sign of toxicity with doses employed.  $EC_{50}$  for *P. nigrium* and *M. myristica* after 30 minutes of exposure time are 0.830 ml/L and 1.22 ml/L respectively. This shows the effectiveness of the essential oil of *P. nigrium* as repellent against *M. subhyalinus* over *M. myristica*. The repellence activity could be as a result of the pungent smell from the volatile constituents of the essential oil.

The effectiveness of the different essential oils in the contact treatment as arranged in reducing order are P. syvestry, P. umbellatum, P. nigrium and *M.myristica*, while for the repellent activity arranged in the same order are P. umbellatum, P. syvestry, P. nigrium and M. myristica. M. myristica proved to be the least effective in both bioassays. The difference in toxicity of the oils may plausibly be due to the variation in the chemical composition of the oils which would have determined the bioactivity of the plant against the termite [16]. The results of this study have shown that the anti-termitic activity of the various essential oils was higher at lowest dose and longest exposure period than at higher dose and longest exposure time. Similar result was reported by Ayvaz et al. [21], who also stated the need to develop alternatives to the synthetic chemicals but emphasized on the need for the alternative product to be safe, effective and economical.

#### 4. CONCLUSION

This work have proven that the four essential oils are effective as anti-termitic substance, the study have also shown that at lower dose and higher exposure time the essential oil seems to be more effective, this knowledge if put in good use, will be a useful tool in producing anti-termitic substance which are economical. The possibility of leaving high residue which could be dangerous to mammals can be controlled with this technology. The study showed that these essential oils can play an important role in the protection of wood and other materials from termite invasion.

#### **COMPETING INTERESTS**

Authors have declared that no competing interests exist.

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