International Journal of Biochemistry Research & Review

22(3): 1-16, 2018; Article no.IJBCRR.42315
ISSN: 2231-086X, NLM ID: 101654445

Insecticidal Potentials and Chemical Composition of Essential Oils from the Leaves of *Phyllanthus amarus* and *Stachytarpheta cayennensis* in Nigeria

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Authors' contributions

This work was carried out in collaboration between both authors. Author OCO designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Author OCM managed the analyses of the study. Author OCM managed the literature searches. Both authors read and approved the final manuscript.

Article Information

DOI: 10.9734/IJBCRR/2018/42315
Editor(s):
(1) Dr. Muhammad Farhan Jahangir Chughtai, Assistant Professor, NUR International University, Lahore, Pakistan.
Reviewers:
(1) Prisila A. Mkenda, Sokoine University of Agriculture, Tanzania.
(2) Zeneida Teixeira Pinto, Brazil.
(3) Jack Williams, Mercyhurst University, USA.
Complete Peer review History: http://www.sciencedomain.org/review-history/25404

Original Research Article

Received 17th April 2018
Accepted 25th June 2018
Published 5th July 2018

ABSTRACT

This research evaluated the insecticidal efficacy of essential oils from the leaves of *Phyllanthus amarus* and *Stachytarpheta cayennensis* on *Periplaneta americana* (American cockroach), *Schistocerca americana* (American grasshopper) and *Anopheles gambiae* (African malaria mosquito). A gas chromatography-Mass spectrometry (GC-MS) analysis of essential oils was also carried out to determine the active components of the oil likely responsible for the observed Insecticidal properties. Insects were exposed to 0.5g of essential oils as well as the positive control (Raid) for 24 hours and mortality observed and recorded every 4 hours. Both essential oils caused 100% mortality in test insects at different time intervals. *P. amarus* and *S. cayennensis* caused 60% and 73% mortality in *A. gambiae* at 8hours, 60% and 90% mortality in *P. americana* at 12 hours and 40% and 90% mortality in *Schistocerca americana* at 16hours. In all cases, *S. americana* recorded the least mortality among the test insects as compared to the other insects. *S. cayennensis* oil proved to be more potent than *P. amarus* oil. The result of the GC-MS analysis carried out on essential oils from both plants revealed the presence of; Decanoic acid, ethylester (Ethyl decanoate)

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6.02%, Dodecanoic acid, ethylester (Ethyl dodecanoate) 11.26%, Tetradecanoic acid, ethylester (Ethyl tetradecanoate) 9.22%, Hexadecanoic acid, ethylester (Ethyl hexadecanoate) 10.16%, Phytol 28.52%, 9, 12, 15-Octadecatrienoic acid, rthylester (Ethyl linolenate) 11.34%, Stigmasta-7,25-dien 3-ol 7.95% and Decanoic acid, ethyl ester (Ethyl decanoate) 6.05%, Dodecanoic acid, ethyl ester (Ethyl dodecanoate) 10.89%, Tetradecanoic acid, ethyl ester (Ethyl tetradecanoate) 8.32%, Hexadecanoic acid ethyl ester (Ethyl palmitate) 5.01%, Phytol 8.27%, Linoleic acid, ethyl ester (Ethyl linolenate) 5.79%, 9,12,15-octadecatrienoic acid, ethyl ester (Ethyl linolenate) 5.79%, Vitamin E 8.29%, Stigmasterol 9.38% and beta-Sitosterol 16.67% for *S. cayannensis* and *P. amarus* respectively. The result from this study indicates that essential oils from these two plants contain potent insecticidal agents that are available, affordable, and effective in the control of insect pests as against the toxic synthetic chemicals currently in use.

Keywords: Insecticidal; potentials; chemical; composition; extracts; plants.

1. INTRODUCTION

Insecticides (natural and synthetic inclusive) are essential tools for preventing or minimizing insect damage to, and significantly increases the quality and quantity of crops, as well as for improving the quality of life for humans, domestic animals and livestock [1]. Insects are invertebrate animals that are part of the larger group of animals called arthropods. They are known to be the most successful and diverse animals on earth. Insects are the principal vectors of the pathogens causing many human, animal and plant diseases. Insect- transmitted pathogens, such as those causing malaria, dengue fever, yellow fever and leishmaniasis kill millions of people annually throughout the world. Malaria alone affects over 300 million people per year, thriving disproportionately in areas of poverty and lower economic growth [2]. Synthetic insecticides developed for dealing with insect pests have been associated with attendant hazards like low affordability, resistance, environmental pollution and other biohazards [3]. However, botanical insecticides have proven to be comparatively less toxic (or in most cases nontoxic at all) than the synthetic insecticides conventionally in use. This paved the way for the search of more botanical insecticides in the management and control of pests [4]. Again, plant extract mixtures may act synergistically [5], showing greater overall bio-activity as compared to their individual constituents [6]. Insect resistance and desensitization is also less likely to develop with mixtures of plant extracts [7,8] as pest resistance is one of the problems associated with intensive use of synthetic insecticides.

The application of plant products as alternative substitutes to the conventional chemical agents has been reported by many researchers like [9]. Over 120 plants and plant products have been shown to have insecticidal or deterrent activity against common insect pests of humans, higher animals and agricultural products [10] Many of the plants used to preserve crops, control or deter insect pests in household management of insect pests have been found to be safe and non-toxic to human upon consumption or inhalation [11]. However, only few of these plants have been scientifically evaluated and characterized successfully [12]. The traditional belief and practice that *Stachytarpheta cayennensis* and *Phyllanthus amarus* leaves when placed on a local lantern repel mosquitoes and other insects such as cockroaches led to the investigation of these two plants. The aim of this research is to extract essential bioactive primary metabolites of botanical origin from the leaves of *Stachytarpheta cayennensis* and *Phyllanthus amarus* that will be effective in pest control and at the same time safe to non target species and to the environment as a whole and may thus replace the environmentally harmful synrthetic chemicals currently in use and to find out the chemical constituents of these oils that may be responsible for their insecticidal action.

Phyllanthus amarus is a widely distributed small erect tropical annual herbal shrubs whose stem has green capsule, and grows up to 10-50 cm high and blows with flowers with 5 white sepals and apical acute anther. It is locally called *Iyin-olobe* by the Yorubas of South-West Nigeria [13] *Starchytarpheta cayennensis* is an erect, perennial, branching somewhat angular fibrous sub-shrub that is very resistant to traction. It usually has opposite, ovate leaves with a distinct petiole and serrated and indented edges, an acute or sub-acute apex, a slightly wrinkled appearance [13].

P. americana is the largest of the common peridomestic cockroaches belonging to the order *Blattodea* and family *Blattidae* [14]. It is native to

Africa and the Middle East and has an average length of 4 cm and about 7 mm tall. It is reddish brown in colour with a yellow margin on the protonum [2] *Schistocerca americana* is a specie of grasshopper belonging to the order *Orthoptera* and family *Acrididae* and commonly known as the American grasshopper [15]. The adult is yellow brown with brown spots on the wings. Two generations occur per year and the female lays up to 3 clutches of egg in a season [15]. *A. gambaie* are the most important vectors of malaria in sub Saharan Africa, particularly of *plasmodium falciarum* [16]. The larva has well developed head with mouth brushes used for feeding, a large thorax and a segmented abdomen [17].

2. EQUIPMENTS AND METHODS

The key equipments used include; Soxhlet extractor, B.BRAN Centrifuge-Manufactured by B.BRAN Scientific and Instrument Company England. Electric blender, AKAI TOKYO JAPAN, Model No: BDOO11DA-1033M, Serial No:10033M96.1198 made in PRC. Thermo Scientific Rotary evaporator Model R-300 USA, Gas chromatography-Mass spectrometry analyser (GCMS-QP2010 PLUS SHIMADZU, JAPAN).Weighing balance (Symmetry Colle-Parmer Instrument Co, USA.

2.1 Identification, Collection and Preparation of Plant Extracts

Fresh leaves of *Phyllanthus amarus* and *Starchytarpheta cayennensis* were obtained from University of Calabar botanical garden and were taken to the Herbarium room in Botany department, University of Calabar where they were identified by a botanist. After identification, the leaves were washed with clean tap water and air-dried for a period of 3 weeks. The dried plant materials were blended into powder using an electric blender. Two hundred (200) grams of each powdered sample was weighed for extraction. The oils were extracted by continuous extraction in soxhlet apparatus for 12 hours using 500 ml of n- hexane (50°C) as solvent according to the method described by the association of official analytical chemists [18] After evaporation using a water bath at 60°C, 11.3 g and 8.5 g of oils were recovered for *Phyllanthus amarus* and *Stachytarpheta cayennensis* respectively corresponding to a percentage oil yield of 5.65% and 4.25% for *Phyllanthus amarus* and *Stachytarpheta cayennensis* respectively. The extracted oils were stored in a dark amber container and kept in a dark, cool environment for further analysis.

2.2 Collection of Test Insects

A total of forty (40) live adult cockroaches and grasshoppers were obtained from an old building structure at Uwanse, and football field of the government secondary school Uwanse respectively, both in Calabar South LGA. Test insects were identified by an Entomologist at the Department of Zoology and Environmental Biology, Faculty of sciences, University of Calabar. All insects used were of adult stage (except for mosquito), this is because they exhibit the greatest destructive and infectious tendencies at this stage. Insects were healthy and very active as at the time of the experiment, no symptoms of any disease or weakness was observed. Their response to environmental factors and stimuli, movement and general behaviour indicated that they were physiologically sound at the time of the procedure. The adult cockroach was recognized by its reddish brown color and a pale brown band around the edge of the pronotum, they also have a pair of slender jointed cerci at the tip of the abdomen. *Schistocerca americana* was distinguished by its yellow brown colour with brown spots on its wing differentiating it from the nymph. All insects were very active and showed no signs of disease or disability as observed from their quick response to stimuli. The insects were kept in four (4) separate well perforated clean transparent plastic containers of 6 inches diameter and 12inches height each for a period of 24 hours at room temperature. Fresh food and green grasses were provided for them inside the containers throughout the experiment. For the mosquito larva, 4 separate medium sized plastic basins filled with natural rain water were kept at a lawn in front of the chemistry laboratory, University of Calabar, for 3 weeks. At the end of 3 weeks a good number of mosquito larvae were seen swimming in the water.

2.3 Grouping of Insects and Administration of Treatments

A total of 40 insects of each specie, were divided into four (4) groups (A, B, C and D) containing 10 insects each in a clean transparent container as described above. Experiment was done in replicate for each treatment on each insect and the average of three determinations was taken. The mosquitoes were divided into groups of 30 each. A piece of clean cotton lint was soaked into 0.5 g amount of each treatment, the soaked

cotton wool was placed inside the Group A (control) received no insecticidal treatment, while groups B and C were exposed to 0.5 g of essential oils obtained from *P. amarus* and *S. cayennensis* respectively. Group D was exposed to 0.5 g of the positive control (Raid). Mortality was observed at 4 hourly intervals for a period of 24 hours and recorded at 4 hours interval.

3. RESULTS

3.1 GasChromatography-Mass Spectrometry (GC-MS) Analysis of Extracted Compounds

GC-MS analysis of both plant extracts was performed using a Perkin-Elmer Gas Chromatography GC Clarus 500 system (Perkin-Elmer Scientific Co. Norwalk, CT06859, and USA), interfaced to a Mass Spectrometer MS equipped with an Elite- 1,5 fused silica capillary column composed of 100% Dimethyl Polysiloxane and an electron ionization system with ionizing energy of 70 eV. The carrier gas used was Helium (99.999%) at a constant flow rate of 0.5 ml/min. One microlitre (1 µ) sample injection volume (Split ratio of 10:1); the inlet/injection temperature was maintained at 250°C, Ion source temperature 300°C for 2 min, then an increase to 120°C, then programmed to increase to 280°C at a rate of 20°C for 5mins. Total run time was 77 min. The MS transfer line was maintained at a temperature of 300°C for 5 min. The Mass Spectra were taken at 70 Ev and a scan interval of 0.5 seconds with fragments from 45 to 415.00KD.

Identification of components: This was based on Interpretations using the database of National Institute Standard and Technology [19] mass spectral library having more than 62,000 patterns. The spectrum of the unknown component was compared with the spectrum of the component stored in the NIST library version, software, turbomas 5.2 [20].

Table 1. Effect of essential oils from *P. amarus (euphorbiaceae)* and *S. cayennensis (verbenaceae*) on percentage mortality of *Periplaneta Americana*

Groups	Time (Hours)								
n=3	0	4	8	12	16	20	24	TM	M%
A (control)	-	-	-	-	-	-		0/10	0%
B (0.5g *P.amarus*)	-	-	-	6	3	1		10/10	100
C (0.5g *S.caynenneses*)	-	-	7	2	1			10/10	100
D (0.5g Raid)	-	10						10/10	100

Table 2. Effect of essential oils from *P. amarus* and *S. cayennensis* on percentage mortality of *Schistocerca Americana*

Groups	Time (Hours)								
n= 3	0	4	8	12	16	20	24	TM	M%
A (control)	-	-	-	-	-	-	-	0	0
B (0.5 g *P.amarus*)	-	-	-		4	4	2	10/10	100
C (0.5 g *S.caynenneses*)	-	-	-	6	3	1		10/10	100
D (0.5 g Raid)	-	10						10/10	100

Table 3. Effect of essential oils from *P. amarus* and *S. cayennensis* on percentage mortality of *Anopheles gambiae*

Groups	Time (Hours)								
n = 3	0	4	8	12	16	20	24	TM	M%
A (control)	-	-	-	-	-	-	-	0	0
B (0.5 g *P.amarus*)	-	-	20	10				30	100
C (0.5 g *S.caynenneses*)	-	-	22	8				30	100
D (0.5 g Raid)	-	26	4					30	100

Key: TM= mortality, M%= percentage mortality

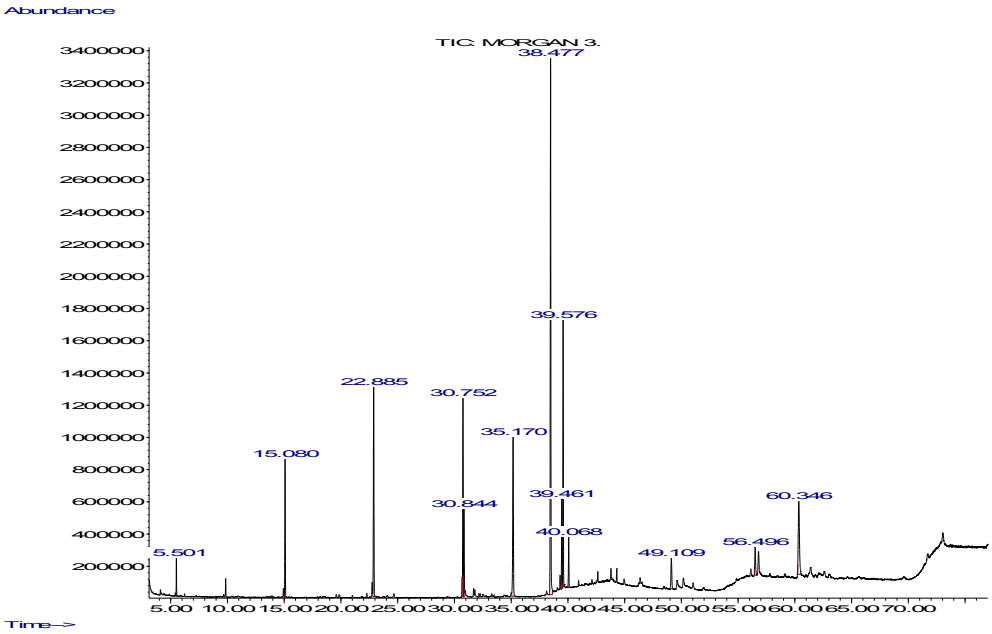


Fig. 1. Chromatogram for *S. cayennensis*

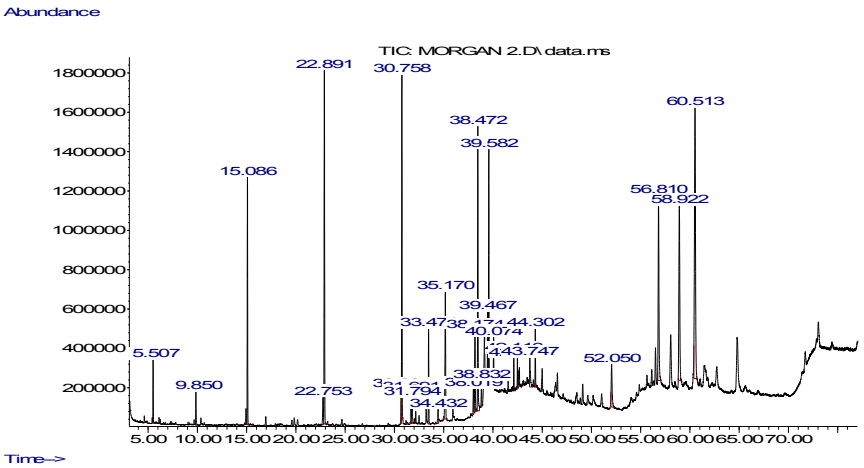
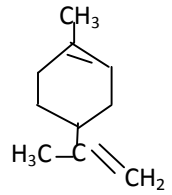
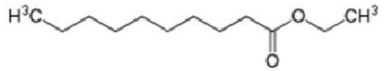
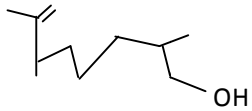
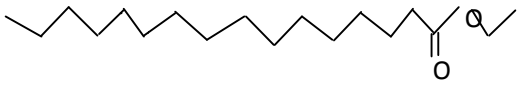


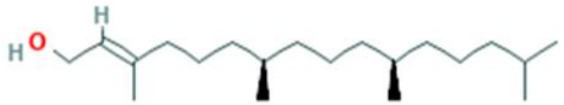
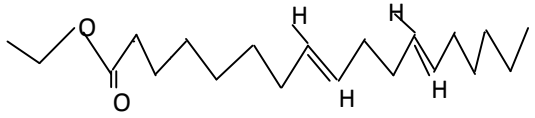
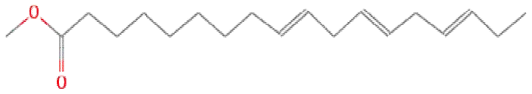
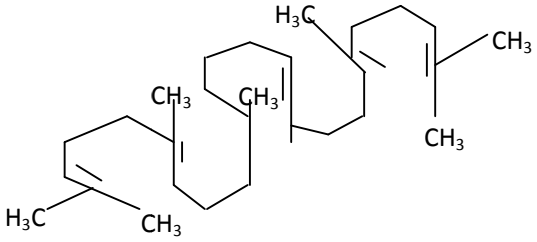
Fig 2. Chromatogram for *P. amarus* extract

4. DISCUSSION

In all cases, considerable differences in insect mortality were observed with different plant extracts and at different time intervals when the insects were exposed to the same amount (0.5 g) of each treatment. The percentage mortality caused by plant extracts on cockroach, grasshopper and mosquito larva are shown in table 1, 2 and 3 respectively. Percentage mortality varied from plant to plant as well as from one insect to another at different time intervals. The positive control (Raid) caused 100% mortality in *P.americana* in 4 hours, while *S. cayennensis* and *P. amarus* caused 100% mortalities in 16 hours and 20 hours respectively. For *S.americana,* Raid caused 100% mortality in 4 hours, while *S. cayennensis* and *P. amarus* caused 100% mortalities in 20 and 24hours respectively. In *A.gambaie* Raid cased 100% mortality in 8 hours, while *P. amarus* and *S. cayennensis* caused 100% morality after 12 hours. *S. cayennensis* seems to possess more insecticidal ability as it killed the insects in a shorter period of time as compared to *P. amarus*. Raid (positive control) was however faster.

Table 4. Chemical composition of n-hexane extract of *Stachytarpheta cayennensis* as revealed by GC-MS

Name of compound	Conc (M)	RT	Mol weight (K)	Molecular formulae	Structural formulae
D-Limonene	1.13	5.501	136.24	$C_{10}H_{16}$	
Decanoic acid, ethylester (Ethyl decanoate)	6.02	15.080	200.32	$C_{12}H_{24}O_2$	
Dodecanoic acid, ethylester (Ethyl dodecanoate)	11.26	22.885	228.371	$C_{14}H_{28}O_2$	
Tetradecanoic acid, ethylester (Ethyl tetradecanoate)	9.22	30.752	256.424	$C_{16}H_{32}O_2$	
Beta-santalol	3.99	30.844	220.356	$C_{15}H_{24}O$	
Hexadecanoic acid, ethylester (Ethyl hexadecanoate)	10.16	35.170	284.477	$C_{18}H_{36}O_2$	

Name of compound	Conc (M)	RT	Mol weight (K)	Molecular formulae	Structural formulae
Phytol	28.52	38.477	296.539	$C_{20}H_{40}O$	
9,12-octadecadienoic acid, ethylester, (Linoleaidic acid ethylester)	3.88	39.461	308.806	$C_{20}H_{36}O_2$	
9, 12, 15-Octadecatrienoic acid, rthylester (Ethyl linolenate)	11.34	39.576	306.483	$C_{20}H_{34}O_2$	
Octadecanoic acid, ethylester (ethyl octadecanoate)	2.23	40.068	312.530	$C_{20}H_{40}0_2$	
Squalene	2.36	49.109	410.73	$C_{30}H_50$	

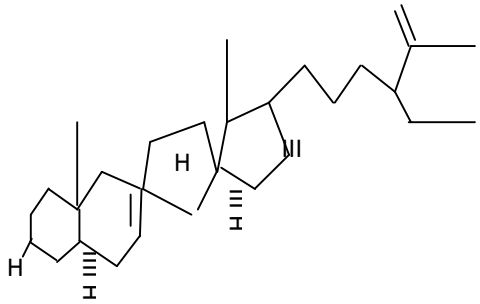
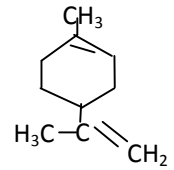
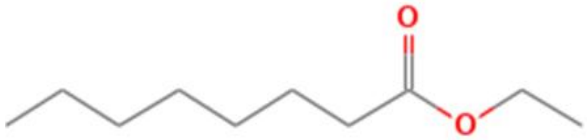
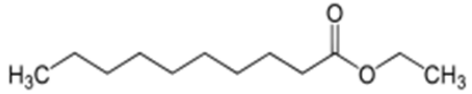
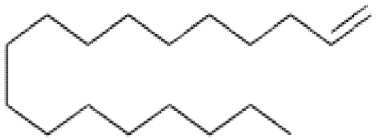
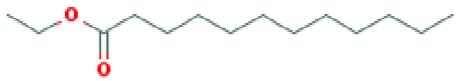
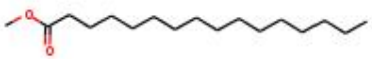
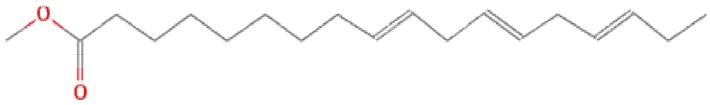
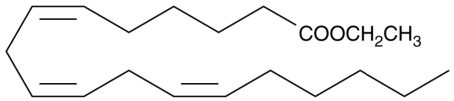
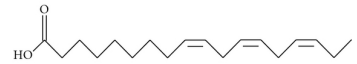
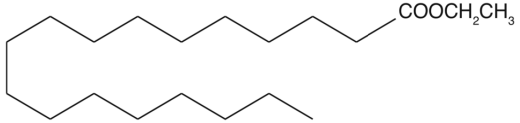
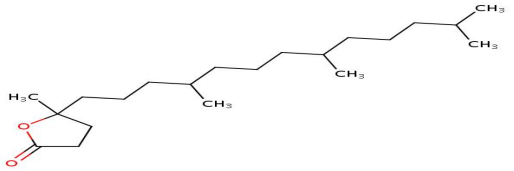
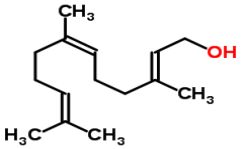
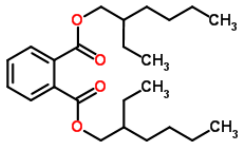
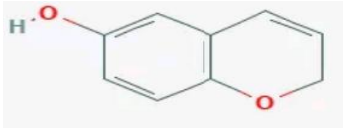
Name of compound	Conc (M)	RT	Mol weight (K)	Molecular formulae	Structural formulae
Vitamin E	1.98	56.496	430.71	$C_{29}H_{50}O_2$	
Stigmasta-7,25-dien-7.95 3-ol	7.95	60.346	412.702	$C_{29}H_{48}0$	

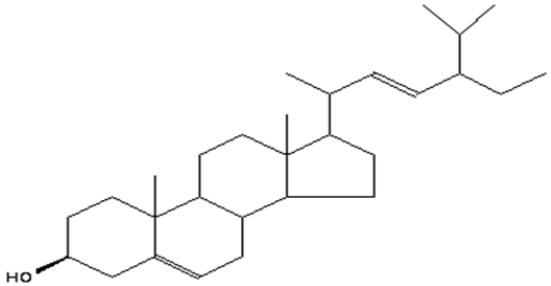
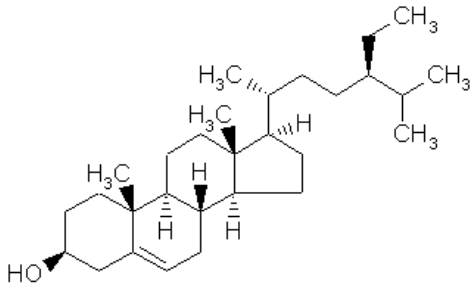
Table 5. Chemical composition of n-hexane extract of *Phyllanthus amarus* as revealed by GC-MS

Name of compound	Conc (M)	RT	Mol. weight (K)	Molecular formulae	Structural formulae
1) D-Limonene	1.10	5.507	136.24	$C_{10}H_{16}$	
2) Octanoic acid, ethyl ester (Ethyloctanoate)	0.78	9.850	172.268	$C_{10}H_{20}O_2$ $CH_3(CH_2)_6$ $CooCH_2$ CH_3	
3) Decanoic acid, ethyl ester (Ethyl decanoate)	6.05	15.086	200.32	$C_{12}H_{24}O_2$	
4) Cetene (5-octadecene)	0.98	22.753	252.486	$C_{18}H_{36}$	
5) Dodecanoic acid, ethyl ester (Ethyl dodecanoate)	10.89	22.891	228.371	$C_{14}H_{28}O_2$	

Name of compound	Conc (M)	RT	Mol. weight (K)	Molecular formulae	Structural formulae
6) l-octadecene	0.73	30.684	252.486	$C_{18}H_{36}$	
7) Tetradecanoic acid, ethyl ester (Ethyl tetradecanoate)	8.32	30.758	256.424	$C_{16}H_{32}O_2$	
8) 1,4-Eicosadiene	0.74	31.691	278.524	$C_{20}H_{38}$	
9) 2-Pentadecanone, 6,10,14- trimethyl	0.72	31.794	268.485	$C_{18}H_{36}O$	
10) Hexadecanoic acid, ethyl ester (methyl hexadecanoate)	2.46	33.471	270.457	$C_{17}H_{34}O_2$	
11) n- Hexadecanoic acid (palmitic acid)	0.57	34.432	236.43	$C_{16}H_{32}O_2$	

Name of compound	Conc (M)	RT	Mol. weight (K)	Molecular formulae	Structural formulae
12) Hexadecanoic acid ethyl ester (Ethyl palmitate)	5.01	35.170	284.477	$C_{18}H_{36}O_2$	
13) 9, 12- octadecatrienoic acid, (z,z) methyl ester (methyl linoleate)	0.77	38.019	294.472	$C_{19}H_{34}O_2$	
14) 9,12,15-octadecatrienoic acid, (methyl ester)	2.96	38.174	292.456	$C_{19}H_{32}0_2$	
15) Phytol	8.27	38.472	296.539	$C_{20}H_{40}0$	
16) Methyl stearate (methyl Octadecanoate)	0.65	38.832	298.511	$C_{19}H_{38}O_2$	
17) Linoleic acid, ethyl ester (Ethyl linolenate)	5.79	39.582	308.499	$C_{20}H_{38}0_2$	

Name of compound	Conc (M)	RT	Mol. weight (K)	Molecular formulae	Structural formulae
18) 9,12,15-octadecatrienoic acid, ethyl ester (Ethyl linolenate)	5.79	39.582	306.483	$C_{20}H_{34}O_2$	
19) Octadecanoic acid, ethylester (Ethyl octadecanoate)	1.20	40.074	312.530	$C_{20}H_{40}O_2$	
20) 4,8,12,16-tetramethylheptadecan-4-olide	0.77	42.116	324.549	$C_{21}H_{40}O_2$	
21) 2,6,10-dodecatrien-i-ol, 3,7,11-trimethyl (Farnesol)	0.52	42.488	222.366	$C_{15}H_{26}O$	
22) Eicosane	1.15	43.747	282.336	$C_{20}H_{42}$	
23) Bis (2-rthylhexyl) phthalate	1.33	44.302	390.55	$C_{24}H_{38}O_4$	
24) 2H-1-Benzopyran-9-01	2.25	52.050	148.161	$C_9H_8O_2$	

Name of compound	Conc (M)	RT	Mol. weight (K)	Molecular formulae	Structural formulae
25) Vitamin E	8.29	56.810	430.71	$C_{29}H_{50}O_2$	
26) Stigmasterol	9.38	58.922	412.702	$C_{29}H_{48}O$	
) beta-Sitosterol	16.67	60.513	414.718	$C_{29}H_{50}O$	

The high mortality rate recorded by the extracts could be as a result of the following predominant bioactive secondary metabolites present in the plants. *P. amarus* was found to contain; Decanoic acid, ethyl ester (Ethyl decanoate), Dodecanoic acid, ethyl ester (Ethyl dodecanoate), Tetradecanoic acid, ethyl ester (Ethyl tetradecanoate), Hexadecanoic acid ethyl ester (Ethyl palmitate), Phytol, Linoleic acid, ethyl ester (Ethyl linolenate), 9,12,15-octadecatrienoic acid ethyl ester (Ethyl linolenate), Vitamin E, Stigmasterol and beta-Sitosterol. For *S. cayennensis* the following compounds were present in good concentrations; Decanoic acid, ethylester (Ethyl decanoate), Dodecanoic acid, ethylester (Ethyl dodecanoate), Tetradecanoic acid, ethylester (Ethyl tetradecanoate), Hexadecanoic acid, ethylester (Ethyl hexadecanoate), Phytol, 9, 12, 15-Octadecatrienoic acid, ethylester (Ethyl linolenate), Stigmasta-7,25-dien- ß -ol.

Interestingly, with the exception of stigmasta-7,25-dien- ß -ol, all other compounds found in good concentrations in *S. cayennesis* were equally present in *P. amarus* in good concentration. *P. amarus* also contained; linoleic acid ethyl ester, vitamin E, stigmasterol and beta sitosterol in addition to the other compounds it shares with *S. cayennensis.*

Phytol; one of the compounds found in high concentration from the GC-MS result is an acyclic diterpene alcohol. Phytol and its metabolites (e.g phytanic acid) have been used as chemical deterrent agents against sumac flea beetles [21]. Recent reports have also revealed that phytol is the key bioactive compound in most botanical insecticides [22]. This could have contributed to the comparatively higher mortality exhibited by *S. cayennensis* in all cases compared to *P. amarus* as *S. cayennensis* contain a higher concentration of phytol (28.52%) than *P. amarus* (8.27%).

Khanna et al. [23] reported the insecticidal activity of ethanolic extract of aerial and root parts of *Phyllanthus amarus* against stored grain pest; *Tribolium castaneum*. Aqueous and ethanol leaf extracts of *P. amarus*, has also been reported to show insecticidal activity against *M. bellicosus* [21]. This confirms that *P. amarus* may actually posses some insecticidal properties as observed from our study. While there are no much information on the insecticidal properties of *Stachytarpheta cayennensis*, the plant is known to be responsible for the inhibition of acetyl cholinesterase [24] making it a potential neuromuscular pesticide. The plant is also used locally in the control of insect pests.

Some of the compounds found in reasonable concentrations and suspected to be responsible for the insecticidal activities observed in these plants include; Phytol which has been implicated as an insecticidal agent by some researchers in earlier studies [25], hexadecanoic acid ethyl ester and tetradecanoic acid ethyl ester which has been reported to posses some insecticidal activities [26]. Stigmast-7-en 3ß-ol; one of the predominant compound found in *S. cayennensis* has also been reported to possess some insecticidal activity [27]. These agree with our findings that these compounds may be largely responsible for the insecticidal actions observed in this study.

The compound with the highest concentration in *P.amarus* extract; sitosterol has been reported to possess some insecticidal properties [28]. The insecticidal properties of the crude extracts of the leaves and flowers of *Anemone pavonina* on *Pheidole pallidula* ants showed that the extract contained as major components; the sitosterol glycopyranoside lipids. Sitosterol is undoubtedly one of the greatest contributors to the insecticidal activity of this *P. amarus* [29]. Another major compound observed in *P. amarus* but not in *S. cayennensis* is Stigmasterol. It is an unsaturated plant sterol present in various medicinal plants. It is utilized in a number of chemical processes which are designed to yield numerous synthetic and semi-synthetic compounds for pharmaceutical industry [30]. Stigmasterol has been reported to inhibit acetyl cholinesterase activity and thus responsible for the larvicidal and repellent properties of *Chromolaena odorata* [31]. It is therefore not surprising that it is one of the active ingredients observed to contribute to the insecticidal ability of *P. amarus*.

5. CONCLUSION

If the major bioactive secondary metabolites observed in these plants are isolated, characterized and formulated into an industrially available form like we have in most synthetic pesticides, we may have in our hands another potent natural pesticide that is affordable, available and environmentally friendly. Field trials are also encouraged to further confirm the findings from this research.

CONSENT

It is not applicable.

ETHICAL DISCLAIMER

As per international standard or university standard written ethical permission has been collected and preserved by the authors.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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Peer-review history:
The peer review history for this paper can be accessed here:
http://www.sciencedomain.org/review-history/25404