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Studies on the Medicinal Plant Acalypha wilkesiana ethanol Extract Phytocomponents by GCMS Analysis

Igwe K. K., Madubuike A.J., Chika Ikenga , Amaku F.J.

Department of Veterinary Physiology, Pharmacology and Biochemistry, Michael Okpara University of Agriculture, Umudike, Nigeria. Department of Veterinary Physiology, Pharmacology and Biochemistry, Michael Okpara University of Agriculture, Umudike, Nigeria. Recare Natural Products, Lagos, Nigeria

Physical Chemistry Research Lab, University of KwaZulu, Natal, Durban, South Africa.

ABSTRACT: The ethanolic extract of the leaves of *Acalypha wilkesiana* plant was studied to find the phytochemical compounds using GCMS analysis. The result of the phytochemical analysis revealed the presence of 12 compounds. Among the 12 compounds, the most abundant were 2-Ethyl-1-hexene with 39.21 peak area %, RT 22.698 and molecular formula C_8H_{16} ; n-Haxadecanoic acid or plamitic acid with 20.92 peak area %, RT:20.92 and molecular formula $C_{16}H_{32}O_2$ and Butane 1,4-diol with 11.58% peak area RT:8.358 and molecular formula of $C_4H_{10}O_2$.which demonstrated various medicinal potentials. Therefore the ethanolic leaf extract of *Acalypha wilkesiana* contain pharmacologically useful active phytochemicals which have effect on progesterone receptors, glucocorticoid receptors, androgen and estrogen receptors with a mild antioxidant and atherosclerotic activity thus could play vital roles in health care programs.

KEYWORDS: Acalypha wilkesiana, Acetophenone, GCMS, n-Hexadecanoic acid, Progesterone receptor.

I. INTRODUCTION

The extract of the herbs has been in use as the main approach to folk medical practitioners in the treatment of ailments and debilitating diseases. The claim that such herbs are efficacious against several ailments and diseases must be backed up by scientific proofs. Twenty five percent of people in the world depend on traditional medicinal plants as drugs for curing various diseases and ailments (1,2,3].Over 6000 plants in India are used in traditional, folk and herbal medicine representing about 75% of the medicinal needs of the developing countries [4]. The side effects associated with synthetic drugs continue to make researchers to look for natural remedies which are safe and effective [5,6]. Our research is therefore being directed towards elucidating potential sources of ethno-medicinal plants using modern scientific analysis like Gas Chromatography-Mass Spectrometry because developments in biotechnology have enhanced investigation of natural compounds faster with more precision than before, leading to isolation of bioactive compounds with health benefits. *Acalypha wikesiana* is one of those ethno medicinal plants with health benefits.

Acalypha wikesiana is a plant (shrub) found worldwide mostly around the tropical of Africa, America and Asia. Its common names are copperleaf and Jecob's coat and it is one of the most widely known and utilized of the family Euphorbiacaece. The genus comprises about 570 species [7] with a layer proportion as needs while others are ornamental plants. The leaves measures 10 - 15cm and heart-shaped with combination of colours like green, purple, yellow, orange, pink or white depending on cultivation. Acalypha wilkesiana is an evergreen shrub usually planted around homes for horticultural purposes. The plant may grow up to 3meters high with erect stems and many branches. Previous scientific evaluation of Acalypha wilkesiana leaves revealed mycotic/antifungal activity [8] and some level of liver toxicity conducted after treatment for 28 days [9]. It looks its best when provided with regular watering during drought and will grow on a wide variety of garden soils, easily propagated by air, layers or cutting [10].



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The leaves of *acalypha wilkesiana* are eaten as vegetables in the management of hypertension [11]. The expressed juice or boiled decoction is used for the treatment of gastrointestinal disorder and fungal infections. Aphids, mites and scales are pest and disease problems on *Acalypha wilkesana* plant [12].

[13] reported the presence of saponins, tannins, anthraquinone and glycoside in the leaves of *Acalypha wilkesiana*. It has antifugal and antibacterial properties [14,15,16,13]. [17] demonstrated that prolonged oral use of *Acalypha wilkesiana* at high dose may be toxic.

This study is to identify the phytocompounds in ethanol extract of *Acalypha wilkesana* responsible for most of these folk claims.

II. MATERIALS AND METHODS

A. Plant Materials

Fresh leaves of *Acalypha wilkesiana* was harvested at Ohafia town in Abia State, Nigeria. The plant leaves were identified by Prof M C Dike at the Taxonomy section of College of Natural Resources and Environmental Management, Michael Okpara University of Agriculture, Umudike, Nigeria.

B. Preparation of Plant Extract

The plant material of *Acalypha wilkesiana* was collected from wild, shade dried for 10 days and pulverized to powder using mechanical grinder. The plant extract was prepared using Soxhlet method described by [18]. Thirty five grams (35 g) of powdered sample was introduced into the extraction chamber of the Soxhlet extractor using methanol as solvent. Temperature was maintained at 70° C throughout the extraction period of 48 hrs. At the end of the extraction period, the extract was concentrated using oven at 35° C to obtain dried extract which was sent for GCMS analysis.

C. GCMS analysis of Acalypha wilkesiana

The characterization of the Phytochemicals in *Acalypha wilkesiana* was done using GC-MS QP2010 Plus (Shimadzu, Japan). The identification of the phytochemicals in the sample was carried out using a QP2010 gas chromatography with Thermal Desorption System, TD 20 coupled with Mass Spectroscopy (Shimadzu). The ionization voltage was 70eV. Gas Chromatography was conducted in the temperature programming mode with a Restek column (0.25 mm, 60 m, XTI-5).The initial column temperature was 80°C for 1min, and then increased linearly at 70°C min⁻¹ to 220°C, held for 3 min followed by linear increased temperature 10°C min⁻¹ to 290°C for 10 min. The temperature of the injection port was 290°C and the GC-MS interface was maintained at 290°C. The sample was introduced via an all-glass injector working in the split mode, with helium carrier gas low rate of 1.2 ml min⁻¹. The identification of compounds was accomplished by comparison of retention time and fragmentation pattern, as well as with mass spectra of the GC-MS.

D. Identification of Phytocompoments in Acalypha wilkesiana

GC-MS Chromatogram of *Acalypha wilkesiana* revealed twelve peaks showing that twelve different compounds were present.

Identity of the active components in the extract was done by comparison of their retention indices, peak area percentage and mass spectra fragmentation pattern with those stored in the database of National Institute of Standards and Technology (NIST) and also with published literature, NIST08.LIB [19], WILEY8.LIB [20], PESTEI-3.LIB and FA-ME.LIB library sources were used for matching the identified components from the plant material. The name, molecular weight, formula, structure and bioactivities of the compounds were ascertained.

III. RESULTS

GCMS chromatogram of the ethanolic extract of *Acalypha wilkesiana* (Figure 1) showed twelve peaks which indicated the presence of twelve phytochemicals constituents. The mass spectra data of *Acalypha wilkesiana* is show in figure 2. The retention time (RT), peak area percentage, molecular weight, molecular formula and bioactivities of *Acalypha wilkesiana* is shown in table 1.



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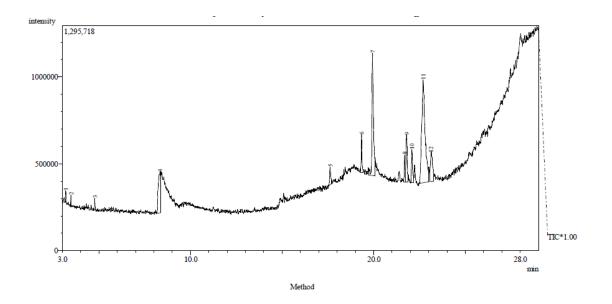
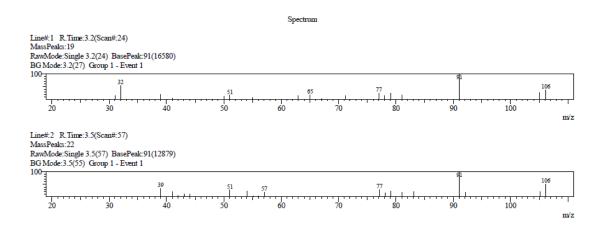


Figure 1: Shows the chromatogram of Acalypha wilkesiana





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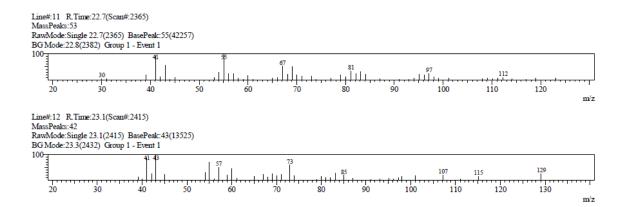


Figure 2: Shows the mass spectra of the twelve phytocompounds identified by GCMS analysis Table1: Shows the names, retention time, peak area percentage, molecular weight, molecular formula and bioactivity of compounds identified in *Acalypha wilkesiana* by GCMS analysis.

S.N	Name of	Retenti	Peak	Molecul	Molecul	Molecular structure	Bioactivity
0	Compound	on time	area %	ar weight	ar formula r		
1	3-Methylene-1- vinyl-1- cyclopentene	3.193	0.58	106.16	C ₈ H ₁₀	CH ₂ H ₂ C	Progesteron e receptor
2	2- Vinylbicyclo[2.1.1] hex-2-ene	3.469	0.47	106.16	C ₈ H ₁₀	CH ₂	Progesteron e receptor
3	Acetophenone or Methyl phenyl ketone	4.771	0.63	120.14	C ₈ H ₈ O	H ₃ C	Hypnotic and anticonvulsa <u>nt</u> under brand name Hypnone.
4	Butane-1,4-diol	8.358	11.53	90.12	C ₄ H ₁₀ O ₂	НООН	Nausea, vomiting, dizziness, sedation, vertigo, and potentially death if ingested in large amounts



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2 Mothul 6 hontor	17.622	2.32	100 01	CILO		Classic
3-Methyl-6-hepten- 1-ol		2.32	128.21	C ₈ H ₁₆ O		Glucocortic oid receptor
Acrylic acid butyl ester	19.331	3.46	128.16	C ₇ H ₁₂ O ₂	H ₂ C	Unknown
n-Hexadecanoic acid or Palmitic acid	19.935	20.92	256.42	C ₁₆ H ₃₂ O 2	0 ••••••••••••••••••••••••••••••••••••	Mild antioxidant and anti- atherosclero tic activity [21]
1,4- Dimethylbenzene or 1,4-Xylene	21.692	3.03	106.16	C ₈ H ₁₀	H ₃ C CH ₃	Inhaling <i>p</i> - xylene can cause dizziness, headache, drowsiness, and nausea. dry skin and redness
Styryl alcohol	21.794	5.97	120.14	C ₈ H ₈ O	ОН	Estrogen receptor; agonist
Phenylethyl alcohol	22.085	4.45	122.16	C ₈ H ₁₀ O	OH	Antiinfectiv e agent and disinfectant
2-Ethyl-1-hexene	22.698	39.21	112.21	C ₈ H ₁₆	H ₃ C CH ₂ CH ₃	Androgen receptor, estrogen receptor agonist
2-Butenyl propionate	23.146	7.41	128.16	C ₇ H ₁₂ O ₂	H ₃ C 0	ACE, angiotensin -converting enzyme,
	Acrylic acid butyl ester n-Hexadecanoic acid or Palmitic acid 1,4- Dimethylbenzene or 1,4-Xylene Styryl alcohol Phenylethyl alcohol 2-Ethyl-1-hexene 2-Butenyl propionate	Acrylic acid butyl ester19.331n-Hexadecanoic acid or Palmitic acid19.9351,4- Dimethylbenzene or 1,4-Xylene21.692Styryl alcohol21.794Phenylethyl alcohol22.085Phenylethyl alcohol22.6982-Ethyl-1-hexene22.6982-Butenyl propionate23.146	Acrylic acid butyl ester 19.331 3.46 n-Hexadecanoic acid or Palmitic acid 19.935 20.92 1,4- Dimethylbenzene or 1,4-Xylene 21.692 3.03 Styryl alcohol 21.794 5.97 Phenylethyl alcohol 22.085 4.45 Phenylethyl alcohol 22.085 39.21 2-Ethyl-1-hexene 22.698 39.21 2-Butenyl 23.146 7.41	Acrylic acid butyl ester 19.331 3.46 128.16 n-Hexadecanoic acid or Palmitic acid or Palmitic acid 19.935 20.92 256.42 1,4- Dimethylbenzene or 1,4-Xylene 21.692 3.03 106.16 Styryl alcohol 21.794 5.97 120.14 Phenylethyl alcohol 22.085 4.45 122.16 2-Ethyl-1-hexene 22.698 39.21 112.21 2-Butenyl propionate 23.146 7.41 128.16	$$	Acrylic acid butyl ester 19.331 3.46 128.16 $C_{7}H_{12}O_{2}$ $H_{2}C - \int_{O} O$ n-Hexadecanoic acid or Palmitic acid 19.935 20.92 256.42 $C_{10}H_{32}O_{2}$ $^{\circ}_{H_{0}}$ 1.4- Dimethylbenzene or 1.4-Xylene 21.692 3.03 106.16 $C_{8}H_{10}$ $H_{3}C - \int_{CH_{3}} C_{H_{3}}$ Styryl alcohol 21.794 5.97 120.14 $C_{8}H_{8}O$ OH Phenylethyl alcohol 22.085 4.45 122.16 $C_{3}H_{10}O$ $\int_{CH_{2}} OH$ 2-Eithyl-1-hexene 22.698 39.21 112.21 $C_{8}H_{16}$ $H_{3}C - \int_{CH_{2}} OH$ 2-Butenyl propionate 23.146 7.41 128.16 $C_{7}H_{12}O_{2}$ $H_{3}C - \int_{CH_{2}} O - \int_{O} OH$

Bioactivity source: www.chemspider.com



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IV. DISCUSSION

The chromatogram of Acalypha wilkesiana leaf indicated the presence of 12 phytocomponents. These compounds were 3-methylene-1-vingl-1-cyclopentene which at retention time of 3.193 had a peak are percentage of 0.58% and 2-Vinylbicyclo (2.1.1) hex -2 – ene which at retention time of 3.459 had a peak area percentage of 0.47% had effect on progesterone receptor. The local effects of progesterone on reproductive organs include the glandular development of the lobular and alveolar tissue of the breast and the cyclic glandular development of the endometrium [22, 23, 24] therefore this plant could be beneficial in the management of pregnancy related cases especially to synchronize estrus.

The compound acetophenone with retention time 4.771 and peak area percentage of 0.63 was found to possess hypnotic and anticonvulsant effect. This compound could be used to induce sleep (hypnosin) or to immobilize reflex as a preanaethetic agent in treatments or surgery. It could also be used to inhibit convulsions acting as a sedative by depressing the central nervous system. Other compounds Butane-1, 4-diol with retention time of 8.358 with peak area percentage of 11.33% and 1, 4 – Dimethyl benzene with retention time of 21.692 and peak area percentage of 3.03 also showed abilities of causing dizziness and sedation thus can act synergistically to potentiate the activity of acetophenone. Acetophenazine, acetophenetidin, and acetophenone group of drugs are known to have a tranquilizing effect [25].

These compounds should be used with caution, because at high doses they could bring side effects like Nausea, vomiting, dizziness, vertigo, headache dry skin and redness and even death [17].

The compound 3- methyl - 6 - hepten - 1 - ol with retention time of 17.622 with peak area percentage of 2.32% had effect on glucocorticoid receptors which could be used to moderate the use of glucose by the cells. [26] in their work agreed that Glucocrticoid hormones stimulate gluconeogenesis by the liver, sometimes producing 6 to 10 fold increase in hepatic glucose production thus being critical to survival during periods of fasting and starvation. [24].

The compound n-Hexadecanoic acid or palmitic acid with retention time 19.935 and peak area percentage of 20.92% had mild antioxidant and anti-atherosclerotic activity [21].

The compound styryl alcohol with retention time 21.794 and peak area percentage of 5.97% had estrogen receptor against activity because of the presence of the benzene ring which could bind alpha or beta estrogen receptor [27].

Also the compound 2-Ethyl – 1-hexene with retention time of 22.698 and peak area percentage of 39.21% which was the most abundant compound in the sample had androgen receptor and estrogen receptor agonist activities.

V.CONCLUSION

From the GCMS analysis of Acalypha wilkesiana, we can conclude that the activities of the extract were hormonal in nature. The influence of the extract was mostly targeted towards steroid hormones as seen in Table 1. From the above analysis Acalypha wilkesiana ethanolic extract could have some tranquilizing and antioxidant activity because of the presence of acetophenone and n-Hexadecanoic acid. The plant extract could also be useful in controlling rennindependant hypertension due to the presence of phytochemical, 2-Butenyl propionate identified by GCMS. Acalypha wilkesiana should be used with caution because high dose could be toxic as demonstrated by [17].

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