



GLOBAL JOURNAL OF SCIENCE FRONTIER RESEARCH: D
AGRICULTURE AND VETERINARY
Volume 16 Issue 2 Version 1.0 Year 2016
Type : Double Blind Peer Reviewed International Research Journal
Publisher: Global Journals Inc. (USA)
Online ISSN: 2249-4626 & Print ISSN: 0975-5896

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Keywords: *acalypha wilkesiana*, acetophenone, GCMS, n-Hexadecanoic acid, progesterone receptor.

GJSFR-D Classification : FOR Code: 060799



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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 C₈H₁₆; n-Hexadecanoic acid or palmitic acid with 20.92 peak area %, RT:20.92 and molecular formula C₁₆H₃₂O₂ and Butane 1,4-diol with 11.58% peak area RT:8.358 and molecular formula of C₄H₁₀O₂, 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.

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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 wilkesiana* is one of those ethno medicinal plants with health benefits. *Acalypha wilkesiana* is a plant (shrub) found worldwide mostly around the tropical of Africa, America and Asia. Its common names are copperleaf and Jacob's coat and it is one of the most widely known and utilized of the family Euphorbiaceae. 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].

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 wilkesiana* plant [12].

[13] reported the presence of saponins, tannins, anthraquinone and glycoside in the leaves of *Acalypha wilkesiana*. It has antifungal 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 wilkesiana* responsible for most of these folk claims.

II. MATERIAL 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.

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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 Phytochemicals 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 AND DISCUSSION

a) 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 shown in figure 2. The retention time (RT), peak area percentage, molecular weight, molecular formula and bioactivities of *Acalypha wilkesiana* is shown in table 1.

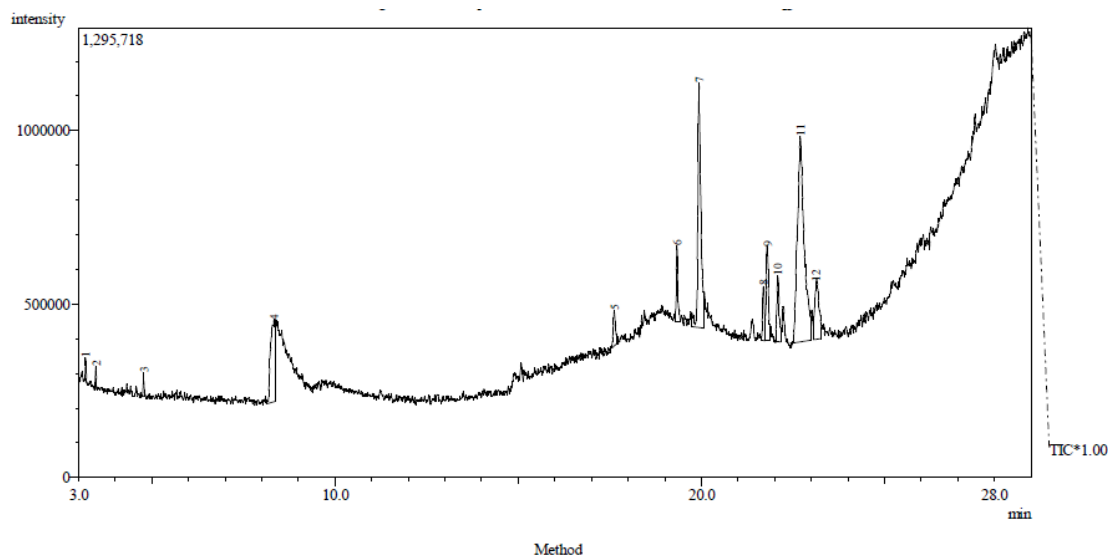


Figure 1 : Shows the chromatogram of *Acalypha wilkesiana*

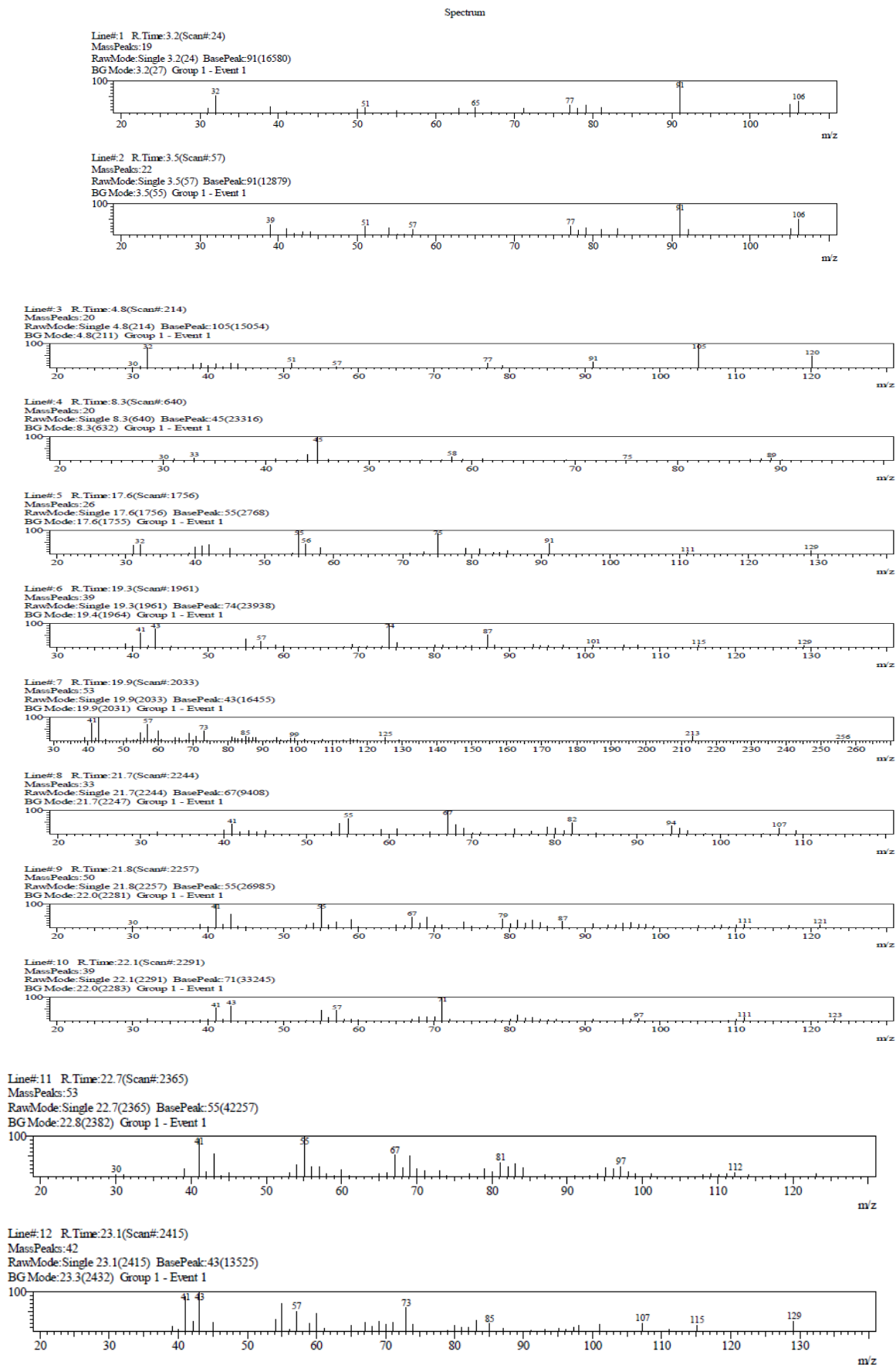
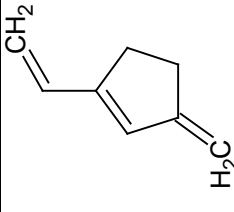
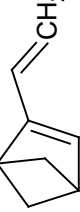
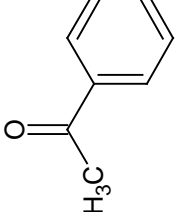
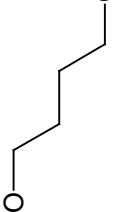
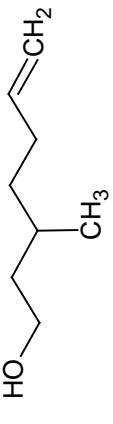
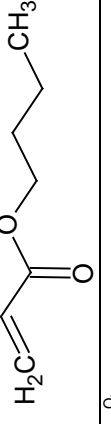

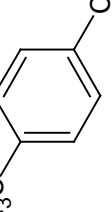
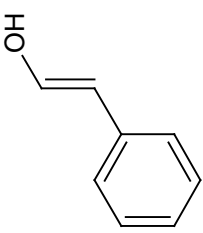
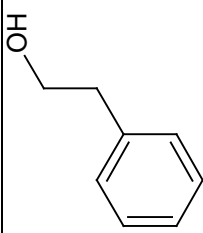
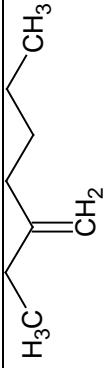
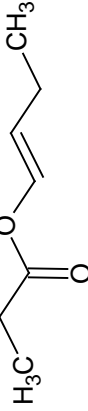


Figure 2 : Shows the mass spectra of the twelve phytocompounds identified by GCMS analysis

Table 1 : Shows the names, retention time, peak area percentage, molecular weight, molecular formula and bioactivity of compounds identified in *Acalypha wilkesiana* by GCMS analysis.

S.No	Name of Compound	Retention time	Peak area %	Molecular weight	Molecular formular	Molecular structure	Bioactivity
1	3-Methylene-1-vinyl-1-cyclopentene	3.193	0.58	106.16	C ₈ H ₁₀		Progesterone receptor
2	2-Vinylbicyclo[2.1.1]hex-2-ene	3.469	0.47	106.16	C ₈ H ₁₀		Progesterone receptor
3	Acetophenone or Methyl phenyl ketone	4.771	0.63	120.14	C ₈ H ₈ O		Hypnotic and anticonvulsant under brand name Hypnone.
4	Butane-1,4-diol	8.358	11.53	90.12	C ₄ H ₁₀ O ₂		Nausea, vomiting, dizziness, sedation, vertigo, and potentially death if ingested in large amounts
5	3-Methyl-6-hepten-1-ol	17.622	2.32	128.21	C ₈ H ₁₆ O		Glucocorticoid receptor
6	Acrylic acid butyl ester	19.331	3.46	128.16	C ₇ H ₁₂ O ₂		Unknown
7	n-Hexadecanoic acid or Palmitic acid	19.935	20.92	256.42	C ₁₆ H ₃₂ O ₂		Mild antioxidant and anti-atherosclerotic activity [21]
8	1,4-Dimethylbenzene or 1,4-Xylene	21.692	3.03	106.16	C ₈ H ₁₀		Inhaling p-xylene can cause dizziness, headache, drowsiness, and nausea. dry skin and redness

9	Styryl alcohol	21.794	5.97	120.14	$C_9H_{10}O$		Estrogen receptor; agonist
10	Phenylethyl alcohol	22.085	4.45	122.16	$C_9H_{10}O$		Antifungal agent and disinfectant
11	2-Ethyl-1-hexene	22.698	39.21	112.21	C_8H_{16}		Androgen receptor, estrogen receptor agonist
12	2-Butenyl propionate	23.146	7.41	128.16	$C_7H_{12}O_2$		ACE, angiotensin-converting enzyme,

Bioactivity source: www.chemspider.com

IV. DISCUSSION

The chromatogram of *Acalypha wilkesiana* leaf indicated the presence of 12 phytochemicals. These compounds were 3-methylene-1-vinyl-1-cyclopentene which at retention time of 3.193 had a peak area 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 preanaesthetic 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 Glucocorticoid 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 rennin-dependant 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].

VI. ACKNOWLEDGEMENT

We appreciate with thanks the research supports from EUNISELL and RECARE natural products.

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