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Chemical and Phytochemical Analyses of Extracts from the Leaves of *Acalypha wilkesiana,* "an Herbal Plant used for the Treatment of Various Skin Disorders"

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Abstract

Acalypha wilkesiana is a tropical herb used for the treatment of skin disorders. Phytochemical studies carried out on the pulverized dried leaves of the plant revealed the presence of alkaloids, cardiac glycosides, anthraquinones, saponins, flavonoids and tannins. The crude 50% methanol extract obtained from the dried leaves was sequentially partitioned into three fractions. Thin layer chromatography of the fractions revealed three (3) components each in hexane fraction; ethyl acetate fraction and butanol fraction of the plant extract. Column chromatography of the fractions was carried out. The infrared analyses of the components revealed the presence of alkenes, hydroxyl group, conjugate carbonyls, esters in the hexane fraction components. Those from the ethyl acetate and butanol fractions contained aliphatic amides or amines with carbonyl and esters attached in the two fractions. Essential oil was extracted from the dried leaves using hydro-distillation method. GC-MS analysis of the essential oil revealed the presence of major compounds which included: n-Hexadecanoic acid 4-Hexen-2-one-3-methyl, Pyrrole and 6-Benzamido-4-benzoyl-1,2,4-triazine-3,5. The presence of the functional groups inferred from IR and GC-MS analyses with the antimicrobial potential of the essential oil may support the use of the plant in the management of skin infections, gastrointestinal disorders and other ailments.

Keywords: *Acalypha wilkesiana,* Phytochemical analyses, Antimicrobial activity, Medicinal properties.

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1.0 Introduction

Most of the World's population use extracts from plants, for their primary health care (Geoffrey, 2009). The plant serves as source of food and medicine giving metabolites, which contain biologically active chemical substances (phytochemicals) such as saponins, tannins, essential oil, flavonoids, alkaloids and other chemical compounds which have therapeutic properties. Many diseases are resistant to synthetic drugs thereby health care practices are now changing from curative to preventive medicine. Alkaloids and vitamins are common phytochemicals popular in preventing diseases. Examples of plants used in the management of different ailments are bitter leaves, *Vernonia amygdalina* used for diabetes, and to stop blood flow in wound (Odugbemi, 2008), lemon grass used as stomachic and diuretic. Some plants are also known to contain essential oils used in different medicinal areas (Sofowora, 2008, Adeyemo *et al*., 2018).

Some *Acalypha* plants are ornamental plants used for gardening because they are attractive and have brilliant colours for example *Acalypha wilkesiana* and *Acalypha hispida. Acalypha* plants are known to have high phytochemical constituents; *Acalypha wilkesiana*, contains carbohydrates, tannins, flavonoids, saponins, essential oil (Adesina *et al.,* 2000). *Acalypha indica* contains compounds like acalyphine, triacetoneamine, cyanogenic glucosides and alkaloids. *Acalypha hispida* also contains some phytochemicals like crude fat, crude proteins, crude fibre and carbohydrates. Omage *et al*., 2014

Acalypha plants are known for their remarkable medicinal values. They are traditionally used in the treatment and/or management of diverse ailments such as diabetes, jaundice, hypertension, fever, liver inflammation, schistosomiasis, dysentery, respiratory problems including bronchitis, asthma and pheumonia as well as skin conditions such as scabies, eczema and mycoses, (Seebaluck *et al.,* 2015) *Acalypha wilkesiana* plant can be used in the management of gastrointestinal diseases, diabetes, skin diseases especially on newly born and on the skin of grown-ups for example, *pityriasis/tinea versicolor* which is very common in people in their twenties. (Adesina *et al.,* 1980). Its ointment is used to treat fungal skin diseases (Oyelami *et al.,* (2003) and can also be used for body pain caused after child birth.

A survey of literature revealed that the FTIR analysis of functional groups for the leaf extracts of *Acalypha wilkesiana* was not done so far, an attempt is made in the present study to analyse the functional groups of phytoactive compounds present, proximate analysis, the GC-MS analysis of the essential oil and as well as antimicrobial activity of the extracts were also investigated. The aim of this research is to investigate the phytochemical and proximate analyses of the leaf extract *Acalypha wilkesiana* and to carry out analyses using GCMS and FTIR on the extracts to identify the chemical compounds responsible for the antimicrobial activities of the plant.

2.0 Experimental

Collection of Plant Materials

Fresh leaves of *Acalypha wilkesiana* were collected from a compound behind Faculty of Education, University of Lagos, Akoka, Lagos, South-West Nigeria around June 2017. The plant was identified and authenticated at Botany Department of the same University with the Voucher specimen number LUH 6497. The fresh plant was air dried and pulverized into coarse powder. Two methods were used to extract from the pulverized material: hydro-distillation and solvent extraction.

Extraction Methods:

Hydro-distillation of Sample

In a typical experiment, 11.45 kg of the fresh leaves were cut into small pieces and air-dried at room temperature in a dust-free environment giving 1.55 kg which was blended to obtain the powder. Batches of 100 g of the powder were each added to 3 L of distilled water in a 5 L round-bottomed flask for each batch. The essential oil was extracted by hydro-distillation and collected into hexane. The hexane solution of the essential oil was concentrated by evaporation at room temperature. Anhydrous sodium sulphate was used to dry the essential oil extract. The colour of the essential oil was light yellow with herbal leave smell. The extraction was carried out in two modes: one was four hours at a stretch where the extract was collected after four hours and the other hourly collection every hour within the four hours (Ogunlesi *et al*., 2009).

GC-MS analysis of the essential oil

The essential oil sample was analysed on a GC-MS (Shimadzu, GC-MS Qp2010 Ultra), with a capillary column size of 30m x 250µm x 0.25µm, film thickness 0.25µm, and packed with OPTIMA -5-MS-using helium as the carrier gas at a flow rate of 1 ml/min. 1 µL of sample was injected. The mass spectrometer (MS) was fitted with Chem-station software for control of the program and processing of the data in MS Library (NIST MS search 2.0).

The injection mode split was at a ratio of 1:1 and injection volume was 0.5µl. Column temperature program was as follows: initially 60^{o}C held at same temperature for 2 minutes, finally increased to 290^{o}C at 9^{o}C/minute and ran for about 30mins, injection temperature was at 250^{o}C, with ion source temperature at 230^{o}C and interface temperature was kept at 300^{o}C. A scan was conducted between 30-300 m/z. Mass spectra were recorded using ionization energy of 70 eV. The molecular ions detected were identified by the MS library (NIST MS search 2.0)

Phytochemical Screening of the Pulverized Sample

The pulverized sample of *Acalypha wilkesiana* was screened for different classes of secondary metabolites. The test carried out included test for alkaloids, cardiac glycosides (cardenolides), anthraquinones, saponins, flavonoids and tannins. The entire tests were carried out using standard methods (Ajaiyeoba *et al.,* 2003; Osadebe *et al.,* 2011; Trease and Evans, 1989). The colour intensity and the precipitate formation were used as analytical responses to these tests since most of these reactions are colour reactions.

Methods of Plant Extraction and Fractionation

The dried and pulverized plant powder (600g) was extracted with 50% methanol in air tight, clean flat bottom flask for 72 hours at room temperature with occasional stirring and shaking. The extract was sieved, and the residue was re-soaked for 24hrs to ensure complete extraction. The extract was concentrated with rotary evaporator at a temperature below 40°C and subsequently dried with a freeze drier. 72.0g of the crude extract was dissolved in 80 mL of distilled water and put in a separating funnel for partitioning. To the water mixture of the crude extract was added 40mL of hexane for the partitioning of the non-polar fraction, the solution was vigorously stirred and allowed to settle until two immiscible layers were formed. The aqueous phase was separated from the organic phase; the organic layer was collected using pipette. This was repeated five times until all components soluble in hexane were extracted and the hexane was colourless. The procedure above was repeated for ethyl acetate to partition the slightly polar fraction and butanol to partition the more polar fraction. The hexane and ethyl acetate fractions were concentrated separately at room temperature while butanol fraction was concentrated using a vacuum oven at a temperature below 40°C.

Yield of the plant was obtained using the formula below:

$$\% \text{ yield} = \frac{\text{Weight of dried extract}}{\text{Weight of wet plant sample}} \times 100$$

The moisture content of the plant was calculated using the formula below;

$$\% \text{ moisture content} = \frac{\text{Amount of water loss}}{\text{Weight of fresh plant sample}} \times 100$$

Thin Layer Chromatography (TLC) of the Fractions

The thin layer chromatography of the three fractions, hexane, ethyl acetate and butanol fractions were carried out using different solvent systems. The solvent system resolved to separate the fractions were: Hexane: Ethyl acetate; 8:2 for the hexane fraction; Hexane: Ethyl acetate: Methanol; 2:7:1 for ethyl acetate fraction and Ethyl acetate: Methanol: Hexane; 1:8:1 for the butanol fraction

The pre-coated TLC plate was used for the conventional analytical TLC. The plant extracts were spotted on the starting point on the plate. The sample was dried and plate later transferred to the development chamber for the separation of the sample. It was later viewed with UV lamp at 254nm wavelength. The R_f values for the components in the hexane fraction were 0.37, 0.60, 0.73 in the ethyl acetate fractions 0.25, 0.45, 0.86; in butanol fractions 0.20, 0.48 0.83.

The three fractions were separated with column chromatography using the resolved solvent systems in the TLC. Components were obtained from each fraction, TLC of the components was carried out and the refractive indices of the components were also obtained to be able to batch the components. The IR spectroscopy of the batched components was carried out. Results are given in Tables 4-7.

3.0 Results

The results of the phytochemical screening are presented in Tables 1 and 2. The result of TLC of the three fractions; hexane, ethyl acetate and butanol in the different solvent systems and the R_f values are presented in Table 4. The results of the FTIR spectroscopy and refractive indices of components from the hexane fraction with column chromatography are presented in Table 5. The results of the FTIR spectroscopy and refractive indices of components from ethyl acetate fraction are presented in Table 6. The results of the FTIR spectroscopy and refractive indices of components from butanol fraction are presented in Table 7.

Table 1: Results of the Phytochemical Screening.

Secondary Metabolites	Reagent	Result
Alkaloids	Dragendoff's	+
	Meyer's	+
	Wagner's	+
Cardiac Glycosides	Keller-killiani	+
Anthraquinones	Chloroform/Ammonia	+
Saponins	Frothing	+
Tannins	Ferric chloride	+
Flavonoids	Magnesium turning/Conc.HCl.	+

Note: + means present

Table 2: Results of the proximate analysis of *Acalypha wilkesiana* compared to proximate analysis of *A. indica, A. marginata, A. hispida* leaves.

Component	Average of 3 *A.wilkesiana* leaves	Std. dev	*A. indica* leaves	*A. hispida* leaves	*A. marginata* leaves
% Moisture content	8.43	0.07	9.49	11.02	10.83
%Crude protein	15.29	0.052	23.98	13.78	18.15
% Crude fat	1.60	2.71948E-16	2.59	6.15	5.60
%Crude fibre	**17.00**	0.02	**8.97**	**10.25**	**11.50**
% Ash	**13.29**	0.035	**12.83**	**10.32**	15.68
% CHO	**61.39**	0.053	**51.10**	**48.48**	**38.24**

CHO: Total Carbohydrate by difference (FAO/WHO/UN, 1991).

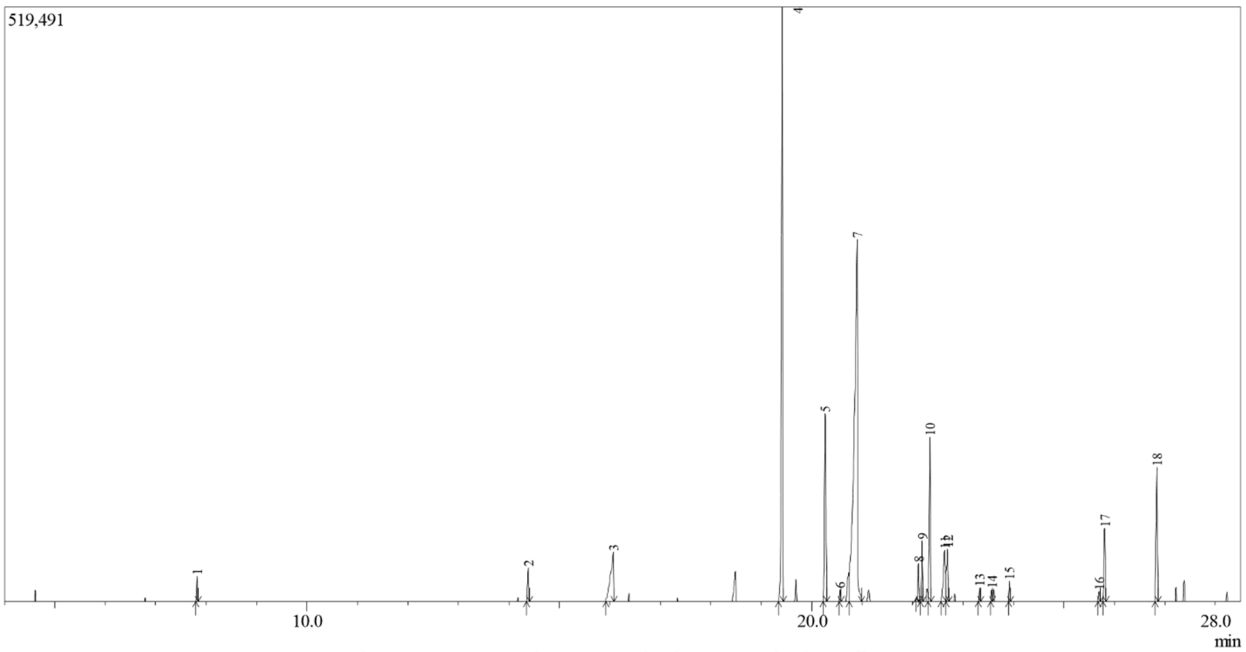


Figure 1: GCMS Chromatogram of the essential oil from *Acalypha wilkisienna*

Table 3: Some major Compounds identified from GC-MS analysis of the four hours stretch essential oils from *Acalypha wilkesiana*.

S/N	Compounds	Retention Time (min)	Abundance (%)	Medicinal and Industrial Uses
1	4-Hexen-2-one,3-methyl	20.27	8.42	Antioxidant, anti-inflammatory, anti-tumor, anticancer and antimicrobial properties (Mc Guinness, 2003)
2	Propanoic acid, anhydride	20.56	0.36	Antibacterial properties and used for the manufacture of pharmaceuticals (Sava *et al.,* 2005)
3	n-Hexadecanoic acid	20.90	37.82	Antibacterial and antioxidant properties and it is also used for the manufacture of soaps, cosmetics and pharmaceuticals hypocholesterolemic, nematicide, pesticide (Sermakkani, and Thangapandian, 2012)
4	1,6-Heptadiene, 2-methyl	23.33	0.42	Anti-Alzheimer, anti-cancer and anti-oxidant properties (Wanninger 2015))
5	Pyrrole	23.57	0.34	Antibiotic, anticancer, antibacterial, antialzheimer, antimalarial and antifungal properties. (Meher *et al.,* 2007)
6	1,3,7-Octatriene	23.918	0.65	Antioxidant properties (Rezaee *et al.,* 2019)
7	2,5-Pyrrolidinedione, 1-(benzoyloxy)	25.70	0.48	Antibacterial and antifungal properties (Meher *et al.,* 2007)
8	6-Benzamido-4-benzoyl-1,2,4-triazine-3,5	25.80	2.87	Antimicrobial, antioxidant, bacteriostatic preservative and also used in nanotechnologies (Hin *et al.,* 2015)

Table 4: TLC Result of Partitioned Fractions

Fraction	Developing solvents	Number of spots
Hexane	Hexane: Ethyl acetate; 8:2	3 (0.37, 0.60, 0.73)
Ethyl acetate	Hexane: Ethyl acetate: Methanol; 2:7:1	3 (0.25, 0.45, 0.86)
Butanol	Hexane: Ethyl acetate: Methanol; 1:1:8	3 (0.20, 0.48 0.83)

Table 5: Infrared Spectroscopy Data of components from hexane fraction

Component	Refractive index	IR Stretching Frequency(cm^{-1})	Group	IR Bending/stretching Frequency (cm^{-1})	Functional group
1	1.3594	2853.64 - 2956.51	sp^3 C-H str.,	1738.36, 1463.03	C=O str., CH_2 bend
2	1.3648	2852.63- 2921.76	sp^3 C-H str.,	1737.62, 1462.67, 1363.81	C=O str., CH_2, and CH_3 bends
3	1.3634	2852.70 and 2921.94	sp^3 C-H str.	1736.63, 1463.64, 1364.09, 1079.86	C=O str., CH_2, and CH_3 bends, C-O bend
4	1.3627	2852.83 and 2921	sp^3 C-H str.	1737.16, 1604.64, 1463.41, 1363.94, 1080.27	C=O str., C=C str., CH_2, and CH_3 bends, C-O bend
5	1.3632	2955.36, 2923.70 and 2853.67	sp^3 C-H str.	1737.74, 1465.55, 1364.57, 1079.64	C=O str., CH_2, and CH_3 bends, C-O bend
6	1.3635	2954.91, 2922.71 and 2853.24	sp^3 C-H str.	1737.26, 1464.02, 1363.95, 1080.46	C=O str., CH_2, and CH_3 bends, C-O bend
7	1.3626	2957.06, 2922.46, 2853.88	sp^3 C-H str.	1737.79, 1463.38, 1364.47, 1079.96	C=O str., CH_2, and CH_3 bends, C-O bend
8	1.3632	2922.12 and 2852.79	sp^3 C-H str.	1736.78, 1464.95 and 1413.47, 1364.13, 1260.07	C=O str., CH_2, and CH_3 bends, C-O bend
9	1.3613	2955.79, 2922.02 and 2852.83	sp^3 C-H str.	1738.55, 1462.66, 1366.01, 1048.17-1230.71	C=O str., CH_2, and CH_3 bends, C-O bend
10	1.3626	2922.25 and 2852.92	sp^3 C-H str.	1737.07, 1462.78, 1365.08, 1079.00-1248.29	C=O str., CH_2, and CH_3 bends, C-O bend

From the IR values obtained, the various components obtained from the hexane fraction after column chromatography were batched as indicated in table 5. Because of similarity in wavenumbers they can be grouped as components 1 and 2 contain carbonyl, 6-7 may contain the ester functional group and 8-10 may contain C-O bend of ether or alcohol.

Table 6: Infra-red Spectroscopy Data of components from ethyl acetate fraction

Component	Refractive index	IR Stretching Frequency(cm^{-1})	Group	IR Bending/stretching Frequency (cm^{-1})	Functional group
1	1.3491	3393.96, 2922.91 and 2853.06	N-H str, sp^3 C-H str.,	1726.01, 1460.72, 1376.82, 1032.99	C=O, CH_2, CH_3 C-O bending
2	1.3662	3399.89, 2923.78 and 2853.62	N-H str, Sp^3 C-H str,	1710.28, 1631.78, 1460.42, 1376.69	C=O, C=C, CH_2, C-H Bending
3	1.3667	3349.82, 2925.59, and 2854.95	O-H str. sp^3C-H str.	1708.43, 1610.65-1515.29, 1439.90, 1204.21	C=O(conj.), C=C, CH_2, C-O bending.
4	1.3309	3298.67, 2926.18 and 2855.10	OH str. sp^3 C-H str.	1701.78, 1608.33-1512.23, 1443.53	C=O (conj.), C=C, CH_2, bending.
5	1.3634	3368.84, 2927.87, 2855.96	OH str. and sp^3 C-H str.	1708.13, 1607.42-1515.17, 1439.40, 1202.88	C=O (conj.), C=C, CH_2, C-O bending.
6	1.3644	3314.37, 2952.55, 2842.61	OH str. and sp^3 C-H str.	1639.98, 1450.34-1408.24, 1111.36-1014.53	C=C CH_2, C-O bending.
7	1.3605	3379.91, 2922.96, 2853.22	N-H str, sp^3 C-H str., CH_2 str.	1735.21,1460.13,1377.49, 1259.66	C=O (ester bend), CH_2, CH_3, C-O bend.
8	1.3623	3341.43, 2112.45	OH str, and C=O	1637.98- 1552.48, 1411.63, 1348.06, 1277.21	C=C, CH_2, CH_3, C-O bend.
9	1.3341	3374.09, 2958.34-2927.33, 2855.18	OH str and sp^3 C-H str,	1710.69, 1637.93, 1379.86, 1261.92	C=O, C=C, CH_2, CH_3, C-O Bending
10	1.3353	3341.07, 2958.54, 2924.84, 2853.17	OH str. and sp^3 C-H str.	1577.16, 1410.80 1382.40, 1349.61	C=C, CH_2, CH_3, C-O Bending

From the IR values obtained, the various components obtained after column chromatography of the ethyl acetate fraction they can be batched as indicated in table 6. Because of similarity in wavenumbers they can be grouped as components 1&2 contain amide as the functional group present, components 3-6 contain carboxylic acid, 7-amide and ester, 8-10 contain carboxylic acid and esters.

Table 7: Infra-red Spectroscopy Data of components from butanol fraction

Component	Refractive index	IR Stretching Frequency(cm^{-1})	Group	IR Bending/stretching Frequency (cm^{-1})	Functional group
1	1.3392	3343.90 and 3247.76, 2925.24, 2853.76	N-H, sp^3 C-H str.	1708.66, 1604.08, 1443.59, 1323.35, 1031.33 and 1193.86.	C=O, C=C, CH_2, CH_3 bends, C-O
2	1.3442	3328.36, 2935.62	OH str. and sp^3 C-H str.	1693.30 and 1611.65, 1440.28, 1333.03, 1036.14	C=O, CH_2, CH_3 C-O bends
3	1.3356	3334.11, 2944.23 and 2832.34	OH str. and sp^3 C-H str.	1725.53, 1448.97 and 1409.23, 1376.94, 1021.47	C=O, CH_2, CH_3, C-O bends
4	1.3373	3331.99, 2943.45 and 2831.81	OH str. and sp^3 C-H str.	1725.71, 1448.86, 1376.50, 1022.13	C=O, CH_2, CH_3, C-O bends
5	1.3328	3299.46, 2947.85 and 2835.58	OH str. and sp^3 C-H str.	1631.72, 1552.63, 1410.24, 1016.17	C=C, C=O, CH_2 C-O bends
6	1.3340	3332.23, 2945.67 and 2833.94	OH str. and sp^3 C-H str.	1724.94, 1641, 1461.71, 1377.42, 1018.32	C=O, C=C CH_2, CH_3, C-O bends
7	1.3345	3334.79, 2924.16 and 2852.73	OH str. and sp^3 C-H str.	1641.11,1462.68,1015.29	C=C, CH_2 C-O bends
8	1.3342	3331.41, 2925.33 and 2852.90	OH str. and sp^3 C-H str.	1640.69, 1409.80, 1014.93	C=C, CH_2 C-O bends
9	1.3338	3348.10, 2944.92 and 2833.52	OH str. and sp^3 C-H str.	1724.88, 1658.04, 1449.36, 1377.67, 1019.6	C=O, C=C CH_2, CH_3, C-O bends
10	1.3307	3329.66, 2923.79 and 2853.13	OH str. and sp^3 C-H str.	1639.58, 1465.86, 1015.68	C=C, CH_2 C-O bends

From the IR values obtained the various components obtained from the butanol fraction can be grouped as component 1 with amide functional group and the –OH which may be from polyphenols (2-10). The –OH may give account of flavonoids that are present from the phytochemical studies.

4.0 Discussion

The result of preliminary phytochemical analysis showed that alkaloids, cardiac glycosides (cardenolides), anthraquinones, saponins, flavonoids and tannins are present as presented in Table 1. Tannins in *Acalypha wilkesiana* helps in the protection of underlying tissue thus; it helps in wound healing (Su *et al.,* 2017). Since flavonoids have the anti-oxidative, anti-inflammatory, anti-mutagenic and anti-carcinogenic properties coupled with the capacity to modulate key cellular enzyme function (Beking*, et al.,* 2010), the presence of flavonoids in the plant supports its usefulness in the management of ailments related to oxidative stress. The qualitative phytochemical result has also shown that leaves of *Acalypha wilkesiana* contain all the secondary metabolites tested for –alkaloids, cardiac glycosides, anthraquinones, tannins, saponins and flavonoids and supported by Ikewuchi *et al.,* 2010.

The result obtained from proximate analysis of the leaves of *Acalypha wilkesiana* in Table 2 establishes that it can be ranked as carbohydrate rich (61.39%) in the leaves due to its relative high carbohydrate content when compared to the other species of the plant. The low moisture content (8.43%) of the leaves would hinder the growth of micro-organisms and the storage life would be high compared to others (9.49%, 11.02%, 10.83*%); Acalypha indica, Acalypha hispida,* and *Acalypha marginata* respectively *(Adeyeye and Ayejuyo,* 2009). The presence of crude protein content of *A. wilkesiana* helps it to build or repair tissues/cells. It has very high crude fibre content, which supports its usefulness in the management of digestive disorders. Epidemiological evidences suggest that increased fibre consumption may contribute to a reduction in the incidence of certain diseases like colon cancer, coronary heart disease, diabetes, high blood pressure, obesity and various digestive disorders. Dietary fiber has been associated with alterations of the colonic environment that protect against colorectal diseases (SACN, 2008). Since the *Acalypha* plant is edible it can also be very useful in the management of ailments that are associated with the colonic environment. The ash content is related to the inorganic elements which can help in the body fluid.

The TLC (thin layer chromatography) result of the three serial partitioned fractions (hexane, ethyl acetate and butanol) revealed that each of the fractions has three (3) spots when viewed in UV lamp (254 nm).

The column chromatography of the fractions was carried out, into many components in test tubes and these were apparently batched according to the refractive indices and TLC of components that gave mostly similar R_f.

The FTIR spectroscopic studies revealed different characteristic peak values with various functional compounds in the extracts. The infrared analysis of the components from hexane fraction (Table 5), confirmed that the components contained the aliphatic carbonyl (aldehyde, esters) As the broad -OH absorption peak is missing, functional groups with C=O stretch and CH_2 bends, some also contained C-O bend are present. Rajiv *et al.2016* analysed *Myristica dactyloides* fruit extracts by FTIR and reported that the functional groups of carboxylic acids, aromatics, alkanes, alcohols, phenols, aliphatic amines, alkenes and

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amine groups in the fruit extracts and also reported that the strong absorption band were observed around 3373–3422 cm^{-1} may be due to the presence of bonded N-H/C-H/O-H stretching of amines and amides). This supports the presence of amides in the ethyl acetate fraction.

The components from ethyl acetate fraction (Table 6), revealed the presence of amides or amines with carbonyl functional groups. The spectra with range of 1735.21 cm^{-1} to 1738.55cm^{-1} indicate the components are aliphatic compounds with carbonyl of carboxylic acid attached. The components spectra that have broad peaks of O-H $_{stretch}$ of hydrogen bonded hydroxyl groups which are indications of alcohols attached to alkene groups. In addition, the -C-O absorption (ether linkage) at the range of 900cm^{-1} to 1200cm^{-1} was very diagnostic. The IR spectra of some components also showed absorption stretching representing the -CH=CH and –OH functional groups respectively. The IR spectra from butanol fraction showed the presence of –NH and C=O, -OH, C=O, CH_2 and C-O, C=C. These functional groups are indicative of the presence of amide, CH_3.

All the samples of essential oil had pungent herbal smell and light yellow colours irrespective of their hours of extraction.
The major peaks as observed in the GC-MS spectrum in Figure 1 and the result in Table 3 revealed the presence of some medicinal compounds like pyrrole, 1,3,7-Octatriene, 2,5-Pyrrolidinedione, 1-(benzoyloxy), 6-Benzamido-4-benzoyl-1,2,4-triazine-3,5,; 4-Hexen-2-one,3-methyl, Propanoic acid, anhydride and also n-Hexadecanoic acid. These compounds have been known to be very strong and effective antifungal, antioxidant, antibacterial, antimalarial and anti-inflammatory, hence, the high antimicrobial activity of the essential oil.

Conclusion

The results of this study indicate that the 50% aqueous methanol extract and several of the fractions, components and the essential oil obtained from the leaves of *A. wilkesiana* exhibit significant antimicrobial activity against several microorganism including gram-positive and gram-negative bacteria as well as fungal organism as revealed also by Oladunmoye 2006. The phytochemical studies had revealed the physiological use of the plant as reported by Osagie *et al* 2014.

The presence of the functional groups from FTIR result, the constituents obtained from the GC-MS analysis of the essential oil which included included: n-Hexadecanoic acid (an antibacterial and antioxidant, Sermakkani, and Thangapandian, 2012), 4-Hexen-2-one-3-methyl (an anti-oxidant and anticancer), Pyrrole (anticancer and antibacterial) and 6-Benzamido-4-benzoyl-1,2,4-triazine-3,5 (antimicrobial and antioxidant) and the antimicrobial potential of the essential oil may support the use of the plant as a treatment for skin infections, gastrointestinal disorders, pneumonia, diabetes, ulcer and cancer.

Conflict of Interest

All authors hereby declare that the work has not been published before and that the publication was read through and the final version approved by all the authors. The authors disclose that there is no actual or potential conflict of interest including any financial, personal or other relationships in any way with other people or organizations within three years of beginning the submitted work that could inappropriately influence, or be perceived to influence, the work.

Author Contributions

Conception: [EO, SN]
Design: [EO, SN]
Execution: [EO, SN, HI]
Interpretation: [EO, SN]
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