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Research Article

GCMS and FTIR analysis of ethanol and methanol leave extract of *Urena lobata* (Caesar weed) for bioactive phytochemical constituents

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Abstract

This study was designed to apply the highly sophisticated biological and chemical characterization techniques–(GC-MS) Gas chromatography-mass spectrometry and FTIR spectroscopy to screen for bioactive phytocompounds present in ethanol and methanol extracts of *U. lobata* .The leaves of the plant were collected from a farm in Umuode village in Osisioma Ngwa local government area of Abia state, Nigeria. Ethanol and methanol extracts of the leaves were prepared and analyzed using Buck M910 Gas chromatography system with HP-5MS column (30m in length x250μm in diameter x 0.25μm in thickness film). Analysis of functional groups in the phytocompounds were done using Buck scientific M530 USA FTIR.12 vibrational functional groups were revealed by the FTIR screening in the ethanol and methanol extracts of U. *lobata* leaves. Some of the functional groups were indicative of alcohols, phenols, aromatic compounds, unsaturated hydrocarbons, vinyl ethers, amines, isonitriles and aliphatic compounds. GC-MS analysis of the ethanol leaf extract revealed 41 bioactive compounds with the following being most abundant, 9-octadecenoic acid(16.8%), dodecenoic acid(13.43%),n-hexadecanoic acid(11.73%), octadecanoic acid (9.78%), 1-docosene(9.57%) while the methanol extract revealed 47 bioactive compounds with the following having the highest abundance, n-hexadecanoic acid (26.65%) and (9.11%), dodecanoic acid(6.89%), 1-docosene(6.06%), erucic acid(4.09%).These phytocompounds and many others present in the leaf have been reported to possess multiple therapeutic activities. This therefore explains the use of this leaf in ancient medicine to treat numerous disease conditions. It further implies that the leaf could be exploited for the formulation of therapeutic molecules.

Keywords: *Urena lobata*, GC-MS, FTIR, Spectroscopy, Phytocompounds, Functional group, Phytoconstituents

INTRODUCTION

Plants, both in their natural state and as extracts, have provided significant benefits to human health for centuries. The pharmacological benefits of these plants come from the wide variety of secondary and primary metabolites they contain, each of which has its own complicated chemical makeup and is responsible for their pharmacological effects[1]. For their low or nonexistent price tags and lack of negative side effects, natural remedies routinely top the list of those recommended over today's synthetic pharmaceuticals. The chemical make-up of these plants is useful not only in traditional therapy but also in the characterization and finding of novel sources of phyto-components for pharmacological research and development[2, 3]. Recently, the use of Gas chromatography-mass spectrometry and FTIR spectroscopy for target identification of functional groups and determination of these different phyto components that are present in medicinal plants even in small concentrations has expanded substantially [1]. GC/MS remains superior among effective, rapid, and precise methods for identifying plant-based substances such as amino acids, alkaloids, esters, long-chain hydrocarbons, steroids, organic acids, nitro compounds and alcohols[4,3].

The sub-shrub *Urena lobata* is a member of the Malvaceae family. They attain heights of 0.6-3m and diameters of 7cm at the base[5]. The plant is mostly found in the tropics and sub-tropics of South and North America, Philippines, Asia, Indonesia as well as in several African nations like Nigeria, Senegal, Ghana and the Democratic Republic of the Congo[6]. In Nigeria, *U. lobata* is also popular and known with different local names among different ethnic groups. The Igbos of Eastern Nigeria particularly those in Imo state call it Achara /Udo[7]the Hausas call it Rama-rama, in Efik, it is known as Rideri, the Binis and some other parts of Edo state call it Oronhon while the Yorubas of Western Nigeria call the plant different names such as; Ilasa-omode, Akeri, Ilasa-agbunrin[8]. *U. lobata* is employed in forkloric medicine in the treatment of dysentery, hematemesis, edema, carbunicle, bleeding due to trauma, leucorrhea, gonorrhea, fever, pain, cold, numbness resulting from rheumatism and other disease conditions[9]. The inhabitants of Nigeria's Katsina State utilize the leaves and petals of this plant to make local delicacies and also offer them to lactating women to increase their milk supply[10]. Numerous studies have reported significant pharmacological activities of various extracts of this plant parts to include anti-inflammatory[11], anti-diabetic[12], anti-tumor[13],antimicrobial[14], anti-oxidant and cytotoxic[15],wound healing effect[16], antidiarrheal[11], neuro-pharmacological activities[17], anti-fertility in female rats as a result of reduced myometrial and epimetrial thickness accompanied by reduced uterine diameter[18]as well as reversibly inhibiting spermatogenesis and steroidogenesis serving as male contraceptives[19].

In order to better understand the therapeutic effects of *Urena lobata*, this study used GC/MS and FTIR to identify the many bioactive phytoconstituents and functional groups present in ethanol and methanol extracts of the plant.

MATERIALS AND METHODS

Collection and processing of plant materials

Healthy and mature leaves of the plant were freshly picked from a farm in Umuode village in Osisioma Ngwa L.G.A of Abia state, Nigeria and identified as *Urena lobata.*

Extraction of plant material

The picked leaves underwent a thorough cleaning with distilled water before being air dried for two weeks. Using an electric blender (ES-BL-090/350W/220-240V/50Hz/China), the dry leave materials were crushed into a fine powder. Soxhlet extraction was performed on the dried and powdered materials using ethanol and methanol as extraction solvents. A rotary evaporator(Heidolph Rotavapor, Germany) was used to filter and concentrate the extracts. When the dried extracts were ready for analysis, they were put into airtight containers, corked, and kept in a refrigerator at 4°C.

Gas Chromatography –Mass Spectrometry (GC-MS) Analysis

Using a BUCK M910 Gas chromatograph with an HP-5MS column (30 m in length, 250µmin diameter, and 0.25µm in film thickness), we were able to conduct the GC-MS analysis of the bioactive components. High-energy electrons were used in a system of electron ionization for spectroscopic detection by GC-MS (70 eV). Helium gas, purified to 99.995% purity, was employed as the carrier gas at a flow rate of 1 mL/min. We started with a temperature of 50°C, ramped it up by 3°Cevery minute, and held it there for around 10 minutes.

At last, a 10°C/min increase in temperature brought the total to 300 °C. In splitless mode, 1µm of the produced 1% extracts diluted with acetonitrile was injected. Based on the chromatographic peak area, the relative amount of bioactive chemicals in each extract was calculated and expressed as a percentage. Based on GC retention time on HP-5MS column and spectrum matching with software data of standards, bioactive chemicals were identified from various extracts (Replib and Mainlab data of GC–MS systems). The name, molecular weight and structure of the compounds of the test extracts were determined

Fourier Transform Infrared Spectroscopic (FT-IR) Analysis

A Buck Scientific M530 USA FTIR was used for the analysis. A deuterated triglycine sulfate detector and a potassium bromide beam splitter were used in this device. The spectra were acquired and adjusted with the help of the Gram A1's software. About 1.0g of samples and 0.5ml of nujol were used; they were carefully mixed and then placed on the salt pellet. FTIR spectra were acquired during the measurement in frequency ranges of 4,000 - 600 cm-1 and co-added at 32 scans and 4 cm-1 resolution. Transmitter values were shown as FTIR spectra.[20].

RESULTS AND DISCUSSION

Among the 41 phytocompounds identified by GC-MS in the ethanol leave extract of *U. lobata* (Table 1), the following were found to be most predominant: 9-Octadecenoic acid, (E)-(16.80%), Dodecanoic acid (13.43%), n-Hexadecanoic acid (11.73%), Octadecanoic acid (9.78%), 1-Docosene (9.57%), Cyclohexadecane, 1,2-diethyl-(4.85% and 2.03%).), Oleic acid (4.74%), Phenol,3,5-bis(1,1-dimethylethyl) (2.42%), Tetradecanoic acid (2.14%). Conversely, the least abundant bioactive phytocompounds in the extract were Nonadecyltrifluoroacetate (0.12%), 6,11-Dimethyl-2, 6,10-dodecatrien-1-ol (0.16%), Decane (0.19%), Dodecyl acrylate (0.22%), 1-Eicosanol (0.22%) and Methyl stearate (0.24%). In a similar study, [76]Fadillah and his groupfrom Jawa Barat, Indonesia investigated the active compounds in both 50% and & 70% ethanol extracts of *U. lobata*. Their findings revealed 5 and 17 bioactive compounds in 50% and 70% ethanol extract of *U. lobata* leaves respectively. They went further to report high concentration of n-Hexadecanoic acid (19.97%) as well as Oleic acid (9.42%) in 50% ethanol leave extract of *U. lobata.* Again, the most prevalent bioactive constituent in the 70% ethanol leave extract of *U. lobata* was discovered to be n-Hexadecanoic acid (18.91%).These findings are comparable with ours, particularly in methanol extract which had, n-Hexadecanoic acid (26.65%) as the most abundant bioactive compound, nevertheless, their study revealed the presence of other bioactive phytocompounds which were not identified in ours and vice versa. As demonstrated in table 5, ethanol leave extract of *U.lobata* has been shown to contain a number of biologically active phytocompounds with diverse biological and pharmacological effects. 9-Octadecenoic acid has been reported to exhibit antimicrobial activities[31].

Dodecanoic acid (also kmown as lauric acid), a medium-chain as well as saturated fatty acid found abundantly in oils of coconut and palm kernel is known to exhibit antibacterial and antioxidant activities[32]. Among many other bioactivities, n-hexadecanoic acid (also known as palmitic acid) has inflammatory, antioxidant, hypocholesteromic, and cancer-preventive properties38. Some common and natural sources of n-Hexadecanoic are palm oil, butter, palm kernel oil, milk, meat and cheese. 1-Docosene has antibacterial activities[43]. Albrathy also reported the antimicrobial activities of Oleic acid, a common monounsaturated fatty acid seen naturally in numerous animal an vegetable fats and oil and in plants such as *Gladiolus italicus* and *Prune mume*[43].

In addition to the above mentioned biological roles, oleic acid has been shown to have antifungal, anti-inflammatory, antioxidant, and antibacterial properties[41]. The phenolic compound, Phenol,3,5-bis(1,1-dimethylethyl) possesses antibacterial and antimicrobial activities[30,29].Least abundant bioactive compounds such as; 1-Eicosanol, methyl stearate, Dodecyl acrylate and 6,11-Dimethyl-2,6,10 dodecatrien-1-ol are not exempted in exhibiting different biological activities as many researchers have also demonstrated their biological activities. For example, 1-Eicosanol, an alcohol has been reported to possess antimalarial, antifungal, and antioxidant activities[31]while Methyl stearate has proven to be a potent anti-inflammatory,intestinal lipid metabolic regulator, GABA aminotransferase inhibitor,antinociceptive, anthelmintic, and a potent gastric inhibitor[42]. On the other hand, Dodecyl acrylate, a typical acrylate ester is known to be antioxidant and antimicrobial[36]while 6,11-Dimethyl-2,6,10 dodecatrien-1-ol has been researched to possess antimicrobial activities[46].

The most bioactive phytocompounds (47) were found in the methanol extract of *U. lobata.* This was illustrated by the GC-MS chromatogram of the methanol extract, which is shown in Figure 2.The following were the prevailing bioactive phytocompounds identified; n-Hexadecanoic acid (26.65% and 9.11%), Dodecanoic acid (6.89%), 1-Docosene (6.06%), Erucic acid(4.09%), 1-Octadecene (3.54%), 9-Octadecenoic acid (3.45%), Tetradecanoicacid (3.01%) and

Diisooctylphthalate (2.97%).The least abundant compounds were; 3-Eicosene (2.97%), 6-Octadecanoic acid methyl ester(0.14%), Dodecane(0.15%), 1-Docosene(0.17%), Trifluoroacetic acid pentadecyl ester (0.18%), Cyclohexane, 1,1`-(1,4-butanediyl)bis (0.22%), Tridecanoic acid,12-methyl-methyl ester (0.22%) and Cyclotetradecane (0.23%).These bioactive compounds and many more in methanol extract of *U. lobata* have been reported to possess many biological activities just like those identified in ethanol extract of the leaf . One of them is Erucic acid; an omega-9 and monosaturated fatty acid that is common among brassicaceae family of flowering plants has been reported to regulate mesenchymal stem cell differentiation into osteoblasts and adipocyte[72]. Another study reported it to have a broad-spectrum antiviral activity against Influenza A. Virus (IAV), anti-inflammatory and pro-inflammatory amplification effect as well as the ability to inhibit NF-κB and p38 MAPK[73]. 1-Octadecene, a long –chain hydrocarbon and an alkene which has been found in appreciable quantity in *Vacciniummacrocarpon*, a North American variant of cranberry known to be used by women in the treatment of recurrent urinary tract infection has also shown excellent antibacterial, antioxidant and anticancer activities[39,59.60]. 9-Octadecenoic acid has been reported to possess antimicrobial activities[31].An ample amount of 9-Octadecenoic acid is found naturally in *Eleutherococcus sessiliflorus* and *Dipteryxlacunifera*, an oleaginous leguminous plant native to Piaui and Maranhao state in North East of Brazil[77]. Diisooctyl phthalate has antimicrobial, antifouling[74]and antibacterial activities[75]. 3-Eicosene has also shown to be antimicrobial, antihyperglycemic, cytotoxic, antioxidant and insecticidal[35]. Another fatty acid methyl ester (FAME) that has been shown to have important biological activities is 6-octadecanoic acid methyl ester. It has been researched to possess strong analgesic, anti-inflammatory and antipyretic activities[68]. Pantadecanoic acid, 9-Hexadecanoic acid and Heptadecanoic acid are among the fatty acids uniquely found in methanol extract of *U.lobata* leaves with tremendous biological significance. Pantadecanoic acid, an essential fatty acid found in butter whose biological activities promote long-term metabolic and cardiovascular health[78]has been shown to possess anticancer activity[61]. 9-Hexadecanoic acid from methanol extract of *Tribolium castaneum* also showed good antibacterial activity against *Escherichia coli*, a Gram-negative, enteropathogenic bacterium implicated in diarrheal disease and urinary tract infections[65]. Heptadecanoic acid has antioxidant properties[66].

Tables 3 and 4 show the findings of the FTIR investigation of the ethanol and methanol leaf extracts of *U. lobata*.FTIR, a vibration spectroscopic technique[79]is renowned for its ability to pin-point important functional groups embedded in plant extracts, biological and synthetic compounds. For identifying types of bonds (functional groups) in compounds, FT-IR remains unmatched[80]. In this study, the FTIR spectra of ethanol and methanol extract shown in figure 3 and 4, unraveled twelve peaks indicative of 12 functional groups. For the ethanol extract, the peaks are in the range of 3704.812, 3498.917, 3181.879, 3013.238, 2782.866, 2665.173, 2571.721, 222.731/2018, 1622.32, 1416.411, 1235.417 and 843.5038 cm^{-1} whereas the peaks of the methanol extract are in the range of; 3664.219/3415.311, 3155.608, 3056.931, 2915.574, 2814.952, 2500.399, 2117.752, 1830.701, 1614.028, 1393.314, 1295.721 and 852.1639cm^{-1}. The 12 functional groups revealed in ethanol extract of the leave are: alcohols/phenols, aromatic amines, primary amides, aromatic/unsaturated hydrocarbons, ether/amine, aldehydes, phosphorus oxyacids, alkynes, primary amines and trisubstituted benzene. In the methanol extract, the functional groups revealed were: alcohols/phenols, amino acids, aromatic/saturated hydrocarbons, aliphatic compounds, hydrohalides, isonitriles, β-lactones, vinyl ethers, t-butyl groups, sulfones and primary amines. The following vibrational functional groups: alcohols/phenols, aromatic/unsaturated hydrocarbons, ether/amine are all common to both extracts and produce, OH stretch, =CH-H stretch and CH stretch respectively. With its hydrogen-bonding capabilities, the OH group is likely responsible for the inhibitory effect against pathogenic microbial agents observed in both methanol and ethanol extract of the leave[80].

Figures 5 and 6 illustrate, respectively, the percentage abundance of the classes of bioactive chemicals found in the ethanol and methanol extracts. In the ethanol extract, fatty acids had a total abundance of 57.58%, alkene, 12.58%, cycloalkane, 11.62%, monounsaturated fatty acids (MUFA), 6.78% while alkane had a total abundance of 4.31%. The least abundant class of bioactive compounds was acrylate ester (0.22%) and alcohol (0.22%). For the methanol extract, as seen in ethanol extract, fatty acid has a total abundance of 56.15%, followed by alkene (20.01%), MUFA (10.02%), phthalate esters (2.97%) and delta-lactams (2.66%). The least abundant class of bioactive compounds were cycloalkane (0.8%) and fatty acid esters (0.18%).

Table 7 showed the bioactive compounds common to both ethanol and methanol extracts of *U. lobata*.The fatty acids were more prevalent in the methanol extract (26.65%) than in the ethanol (11.73%), including n-Hexadecanoic acid, 9-Octadecenoic acid, Dodecanoic acid, and Tetradecanoic acid. In ethanol extracts, 9-octadecenoic acid predominated (16.8%) compared to methanol extracts (3.45%).Dodecanoic acid was prevalent in ethanol extract (13.43%) compared to the methanol extract (6.89%). The varied concentrations of these bioactive compounds in the two extracts underscored the importance of choosing the right extraction solvents during research bearing in mind the bioactive compound of target.

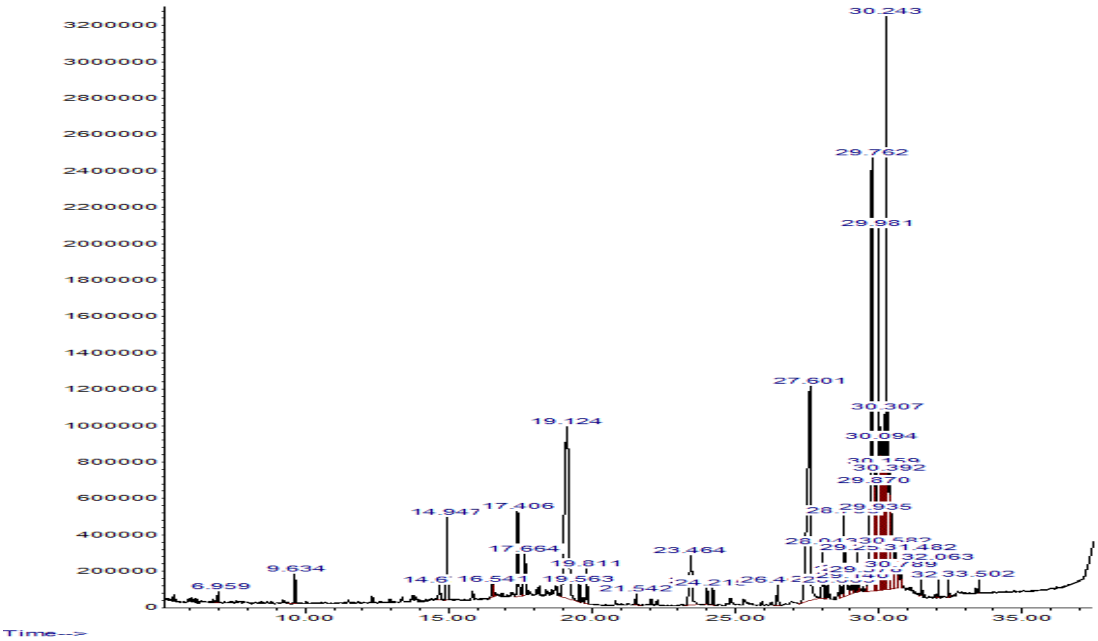


Figure 1: GC-MS Chromatogram of ethanol extract of *U. lobata* leaves

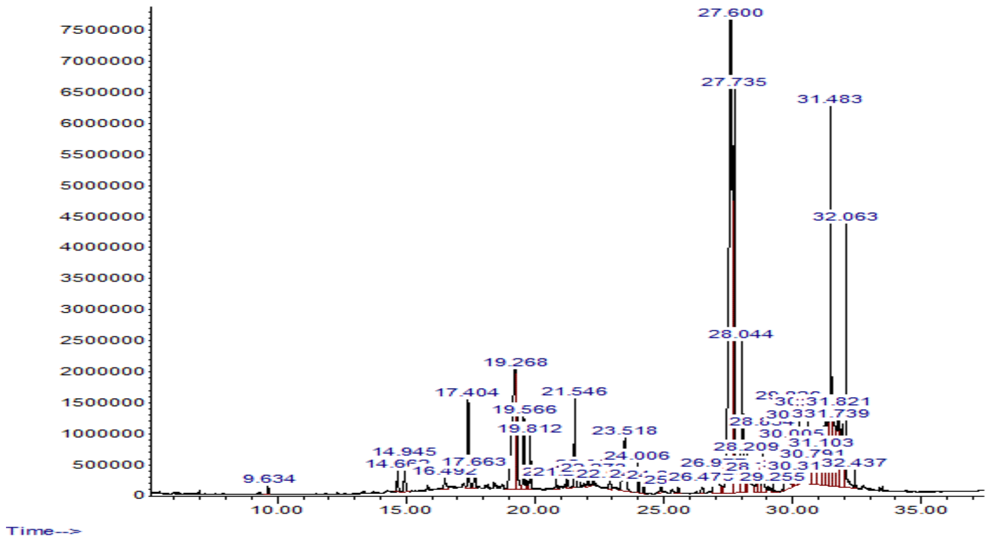
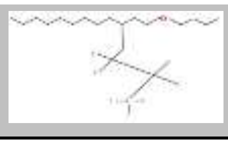
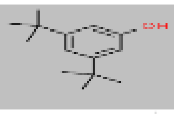
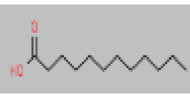
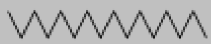
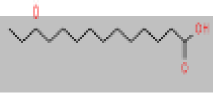
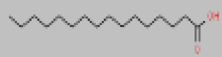
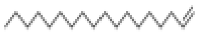
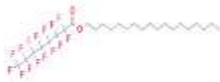
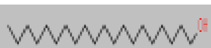


Figure 2: GC-MS Chromatogram of methanol extract of *U. lobata* leaves

TABLE 1: BIOACTIVE COMPOUNDS PRESENT IN *U. LOBATA* ETHANOL LEAVE EXTRACT BY GC-MS

PK	RT	Area %	Bioactive compound	Structure	MF	MW
1	6.9 59	0.19	Decane		**$C_{10}H_{22}$**	142.28
2	9.634	0.61	Dodecane		**$C_{12}H_{26}$**	170.33
3	14.674	0.39	Cyclopentane,1-hexyl-3-methyl-		**$C_{12}H_{24}$**	168.32
4	14.947	1.75	Tetradecane		**$C_{14}H_{30}$**	198.39
5	16.519	0.28	3-Heptafluorobuty Roxydodecane		**$C_{20}H_{35}F_7O_2$**	424.00

6	16.541	0.31	Silane, trichlorodocosyl-		$C_{12}H_{25}Cl_3Si$	303.77
7	17.406	2.42	Phenol, 3,5-bis(1,1-di methylethyl)		$C_{14}H_{22}O$	206.32
8	17.664	1.11	Undecanoic acid, 10-methyl-, methyl ester		$C_{13}H_{26}O_2$	214.34
9	19.124	13.43	Dodecanoic acid		$C_{12}H_{24}O_2$	200.32
10	19.563	0.39	9-Eicosene, (E)-		$C_{20}H_{40}$	280.53
11	19.811	0.72	Hexadecane		$C_{16}H_{34}$	226.44
12	21.542	0.22	Dodecyl acrylate		$C_{15}H_{28}O_2$	240.38
13	23.464	2.14	Tetradecanoic acid		$C_{14}H_{28}O_2$	228.37
14	24.003	0.38	9-Eicosene, (E)-		$C_{20}H_{40}$	280.53
15	24.215	0.37	Octadecane		$C_{18}H_{38}$	254.49
16	26.469	0.47	Pentadecanoic acid, 14-methyl- ,methyl ester		$C_{17}H_{34}O_2$	270.45
17	27.601	11.73	n-Hexadecanoic acid		$C_{16}H_{32}O_2$	256.42
18	28.043	1.11	1-Octadecene		$C_{18}H_{36}$	252.47
19	28.207	0.26	Methoxyacetic acid, 2-tetradecyl ester		$C_{17}H_{34}O_3$	286.45
20	28.669	0.43	Pentadecafluorooctanoic acid, octadecyl ester		$C_{26}H_{37}F_{15}O_2$	666.5
21	28.796	2.02	9-Octadecenoic acid		$C_{18}H_{34}O_2$	282.46
22	29.042	0.40	Cyclohexane, butyl-		$C_{10}H_{20}$	140.26
23	29.140	0.22	1-Eicosanol		$C_{20}H_{42}O$	298.54
24	29.253	0.52	cis-13-Octadecenoic acid, methyl ester		$C_{19}H_{36}O_2$	296.48
25	29.319	0.25	cis-Vaccenic acid		$C_{18}H_{34}O_2$	282.5
26	29.578	0.24	Methyl stearate		$C_{19}H_{38}O_2$	298.5
27	29.762	16.80	9-Octadecenoic acid		$C_{18}H_{34}O_2$	282.46
28	29.870	2.03	Cyclohexadecane, 1,2-diethyl-		$C_{20}H_{40}$	280.53

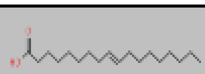
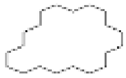
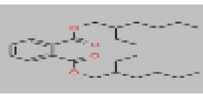
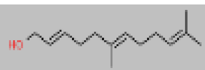
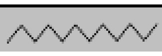
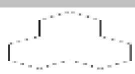
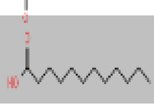
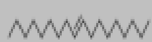
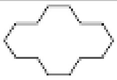
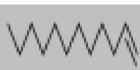
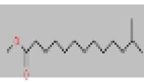
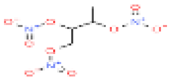
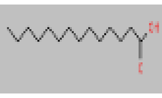
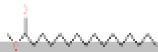
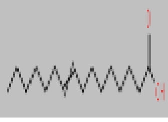
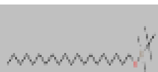
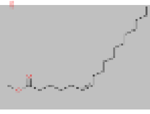
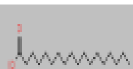
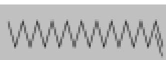
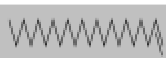
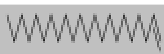
29	29.935	0.91	9-Octadecenoic acid		$C_{18}H_{34}O_2$	282.46
30	29.981	9.78	Octadecanoic acid		$C_{18}H_{36}O_2$	284.47
31	30.094	4.74	Oleic Acid		$C_{18}H_{34}O_2$	282.5
32	30.159	1.79	Oleic Acid		$C_{18}H_{34}O_2$	282.5
33	30.243	9.57	1-Docosene		$C_{22}H_{44}$	308.58
34	30.307	4.34	Cycloeicosane		$C_{20}H_{40}$	280.53
35	30.392	4.85	Cyclohexadecane, 1,2-diethyl-		$C_{20}H_{40}$	280.53
36	30.582	1.63	5-Eicosene, (E)-		$C_{20}H_{40}$	280.53
37	30.789	0.27	3-Eicosene, (E)-		$C_{20}H_{40}$	280.53
38	31.482	0.35	Heptadecylheptafluorobutyrate		$C_{21}H_{35}F_7O_2$	452.49
39	32.063	0.31	Bis(2-ethylhexyl) phthalate		**$C_{24}H_{38}O_4$**	390.55
40	32.437	0.12	Nonadecyltrifluoroacetate		$C_{21}H_{39}F_3O_2$	380.52
41	33.502	0.16	6,11-Dimethyl-2,6,10-dodecatrien-1-ol		$C_{14}H_{24}O$	208.34

TABLE 2: BIOACTIVE COMPOUNDS PRESENT IN *U. LOBATA* METHANOL LEAVE EXTRACT BY GC-MS

Pk	RT	Area %	Bioactive compound	Structure	MF	MW
1	9.634	0.15	Dodecane		$C_{12}H_{26}$	170.33
2	14.662	0.43	5-Tetradecene, (E)-		$C_{14}H_{28}$	196.37
3	14.945	0.61	Tetradecane		$C_{14}H_{30}$	198.38
4	16.492	0.35	Cyclododecane		$C_{12}H_{24}$	168.31
5	17.404	1.86	2,4-Di-tert-butylphenol		$C_{14}H_{22}O$	206.32
6	17.663	0.42	Undecanoic acid, 10-methyl-, methyl ester		$C_{13}H_{26}O_2$	214.34
7	19.221	6.89	Dodecanoic acid		$C_{12}H_{24}O_2$	200.31
8	19.268	2.53	Dodecanoic acid		$C_{12}H_{24}O_2$	200.31
9	19.566	1.41	9-Octadecene, (E)-		$C_{18}H_{36}$	252.47

10	19.812	0.96	Hexadecane		$C_{16}H_{34}$	226.44
11	20.815	0.22	Cyclohexane,1,1'-(1,4-butanediyl) bis-		$C_{16}H_{30}$	222.40
12	21.243	0.23	Cyclotetradecane		$C_{14}H_{28}$	196.37
13	21.546	1.59	1-Dodecene		$C_{12}H_{24}$	168.32
14	22.063	0.31	Heptadecane		$C_{17}H_{36}$	240.46
15	22.273	0.22	Tridecanoic acid, 12-methyl-, methyl ester		$C_{15}H_{30}O_2$	242.39
16	22.904	0.26	1,2,4-Butanetriol, trinitrate		$C_4H_7N_3O_9$	241.11
17	23.518	3.01	Tetradecanoic acid		$C_{14}H_{28}O_2$	228.37
18	24.006	0.53	1-Octadecene		$C_{18}H_{36}$	252.47
19	24.214	0.24	Heptadecane,2 methyl-		$C_{18}H_{38}$	254.49
20	24.880	0.23	Pentadecanoic acid		$C_{15}H_{30}O_2$	242.39
21	25.557	0.18	Trifluoroacetic acid, pentadecyl ester		$C_{17}H_{31}F_3O_2$	324.42
22	26.479	0.25	Hexadecanoic acid, methyl ester		$C_{17}H_{34}O_2$	270.45
23	26.977	1.64	9-Hexadecenoic acid		$C_{16}H_{30}O_2$	254.40
24	27.600	26.65	n-Hexadecanoic acid		$C_{16}H_{32}O_2$	256.42
25	27.735	9.11	n-Hexadecanoic acid		$C_{16}H_{32}O_2$	256.42
26	28.044	3.54	1-Octadecene		$C_{18}H_{36}$	252.47
27	28.209	0.94	Heptacosane,1-chloro-		$C_{27}H_{55}Cl$	415.17
28	28.594	0.46	Heptadecanoic acid		$C_{17}H_{34}O_2$	270.45
29	28.703	0.57	Butyl eicosyl ether		$C_{26}H_{56}OSi$	412.80
30	28.854	1.50	trans-13-Octadecenoic acid		$C_{18}H_{34}O_2$	282.46
31	29.255	0.14	6-Octadecenoic acid, methyl ester, Z		$C_{19}H_{36}O_2$	296.48
32	29.826	3.45	9-Octadecenoic acid		$C_{18}H_{34}O_2$	282.46
33	30.005	0.68	Octadecanoic acid		$C_{18}H_{36}O_2$	284.47

34	30.091	0.28	Hexadecanoic acid, 1,1-dimethylethyl ester		$C_{20}H_{40}O_2$	312.53
35	30.245	0.56	1-Docosene		$C_{22}H_{44}$	308.58
36	30.310	0.10	3-Eicosene, (E)-		$C_{20}H_{40}$	280.53
37	30.599	1.59	cis-Vaccenic acid		$C_{18}H_{34}O_2$	282.46
38	30.791	0.74	1-Docosene		$C_{22}H_{44}$	308.58
39	31.103	1.67	cis-Vaccenic acid		$C_{18}H_{34}O_2$	282.46
40	31.249	2.67	Palmitoleic acid		$C_{16}H_{30}O_2$	254.40
41	31.382	2.66	2-Piperidinone,N-[4-bromo-n-butyl		$C_9H_{16}BrNO$	234.13
42	31.483	6.06	1-Docosene		$C_{22}H_{44}$	308.58
43	31.621	2.84	1-Docosene		$C_{22}H_{44}$	308.58
44	31.739	2.04	1-Docosene		$C_{22}H_{44}$	308.58
45	31.821	4.09	Erucic acid		$C_{22}H_{42}O_2$	338.60
46	32.063	2.97	Diisooctyl phthalate		$C_{24}H_{38}O_4$	390.55
47	32.437	0.17	1-Docosene		$C_{22}H_{44}$	308.58

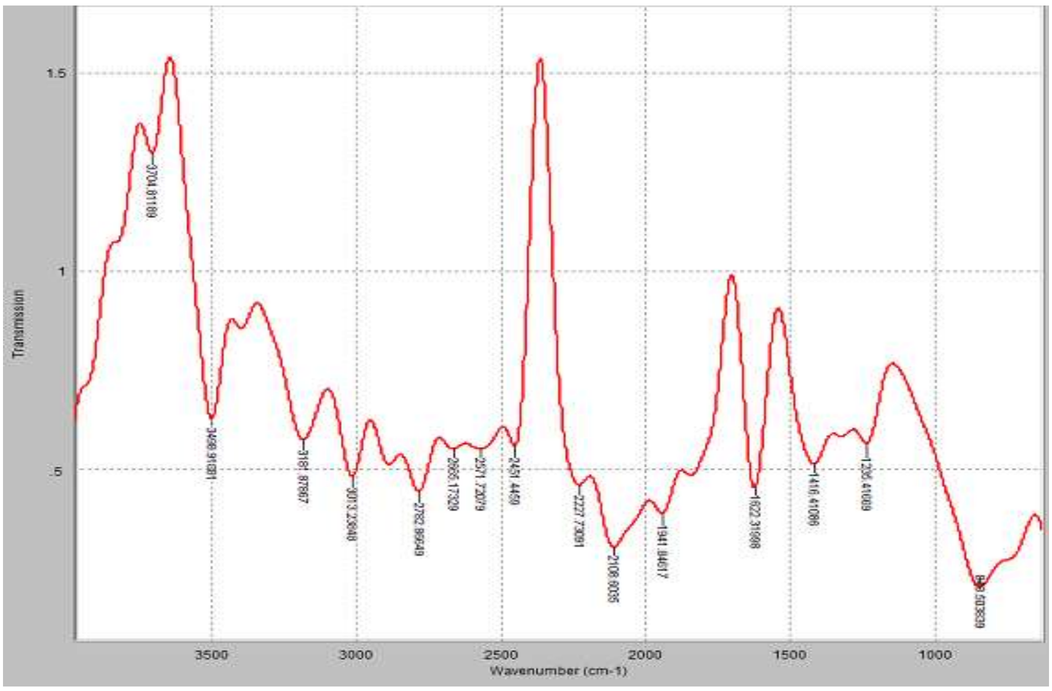


Figure 3: FTIR Spectrum of ethanol extract of *U. lobata* leaves

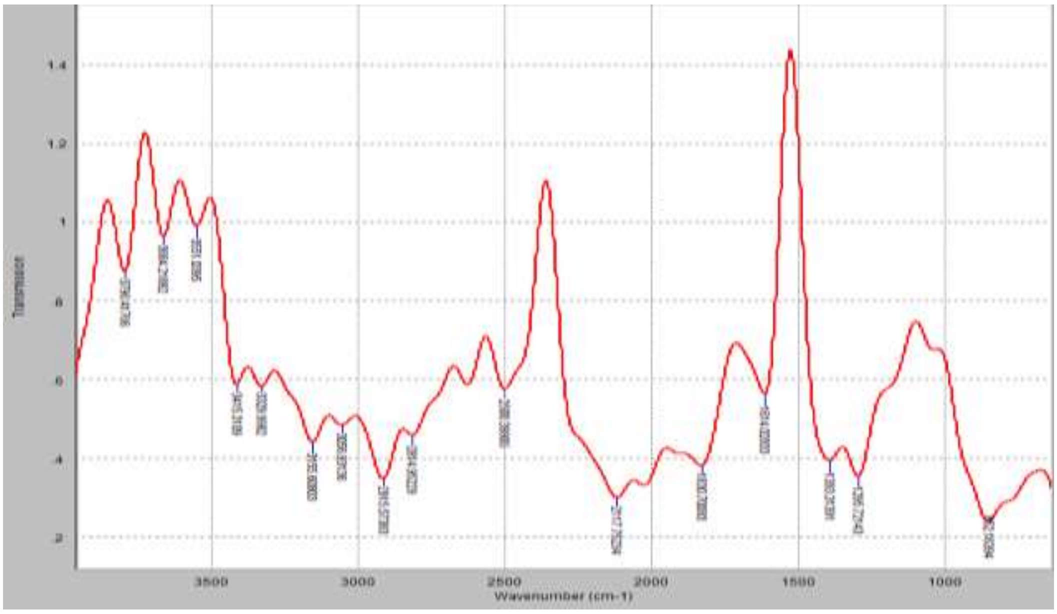


Figure 4: FTIR Spectrum of methanol extract of *U. lobata* leaves

Table 3: FTIR peak values and functional groups of ethanol extract of *U.lobata* leaves

	Wavenumber (cm^{-1})	Functional group/mode of vibration	Inference
1	3704.812	-OH, OH Stretching	OH in Alcohols and Phenols
2	3498.917	-NH_2, NH Stretch	NH in primary amines, amides and aromatic amines,
3	3181.87	-NH_2,NH_2Symmetric stretch	NH_2 in primary amides
4	3013.238	= CH, = CH-H Stretching	= CH in unsaturated and aromatic hydrocarbons
5	2782.866	-CH_3, CH Stretch	CH_3 attached to N or O
6	2665.173	-CHO, CH Bending	CH in Aldehydes
7	2571.721	-OH, OH Stretching	OH in phosphorus Oxyacids
8	2227.731 2108	C ≡ C, C ≡ C Stretch	C ≡ C in Alkynes
9	1622.32	N-H, NH Deformation	NH in Primary amides
10	1416.411	C-N, C-N Stretch	C-N in Primary amides
11	1235.417	C-N, C-N Stretch	C-N in Primary amides
12	843.5038	1,3,5-trisubstituted benzenes, CH out-of-plane deformation	In tri-substituted benzenes

Table 4: FTIR peak values and functional groups of methanol extract of *U.lobata* leaves

S/N	Wavenumber (cm^{-1})	Functional group/mode of vibration	Inference
1	3664.219 3415.311	-OH , OH stretch	OH in Phenols and Alcohols
2	3155.608	NH_3^+, Antisymmetric stretch	NH in amino acids
3	3056.931	=CH, =CH-H stretch	CH in unsaturated and aromatic hydrocarbons
4	2915.574	-CH and –CH_2-, -CH_3- Antisymmetric and symmetric stretching	CH in Aliphatic compounds
5	2814.952	-CH_3, CH Stretch	CH_3 attached to N or O
6	2500.399	NH_3^+, NH Stretching	NH in Amine in hydrohalides
7	2117.752	N≡C, N≡C Stretch	N≡C in isonitriles

8	1830.701	C=O, C=O Stretch	C=O in β-lactones
9	1614.028	C = C, C = C Stretch	C = C in Vinyl ethers
10	1393.314	t-butyl groups, CH_3 deformations	t-butyl groups
11	1295.721	SO_2, SO_2 Antisymmetric stretch	SO_2 in Sulfones
12	852.1639	R-NH_2, NH_2 wag	NH_2 in Primary amines

TABLE 5: ACTIVITIES OF BIOACTIVE COMPOUNDS PRESENT IN *U. LOBATA* ETHANOL LEAVE EXTRACT

S/N	COMPOUNDS	CLASS OF COMPOUND	BIOLOGICAL ACTIVITY
1	Decane	Alkane	Possess alarm pheromonal activity [21] Possess tumor-promoting activity[22] Antifungal and Antibacterial[23]
2	Dodecane	Alkane	Neurotrophic action[24]
3	Cyclopentane, 1-hexyl-3-methyl-	Alkane	No activity reported
4	Tetradecane	Alkane	Possess tumor-promoting and cocarcinogenic activity[22,25] Antimicrobial, Diuretic &Antituberculosis[26] Antifungal[27] Antibacterial[28]
5	3-Heptafluoro-butyroxydecane	Alkane	No activity reported
6	Silane, trichlorodocosyl	Organosilicon	No activity reported
7	Phenol, 3,5-bis(1,1-dimethylethyl)	Phenol	Antimicrobial activity[29] Antibacterial activity[30]
8	Undecanoic acid, 10-methyl, methylester	FAME	Antioxidant, increase aromatic amino acid decarboxylase activity[31]
9	Dodecanoic acid	Fatty acid	Antibacterial and Antioxidant activities[32]
10	9-Eicosene	Fatty acid	Antimicrobial[33]
11	Hexadecane	Alkane	Antifungal, antibacterial and antioxidant activities[34] Cytotoxicity, antimicobial, antioxidant, antipyretic, anthelmentic, anti-infammatory, antidiarrhea, antidiabetic[35]
12	Dodecyl acrylate	Acrylate ester	Antioxidant and antimicrobial activities[36]
13	Tetradecanoic acid	Fatty acid	Antifungal, antioxidant, cancer preventive, nematicide, hypocholestrolemic, lubricant[31]
14	9-Eicosene	Fatty acid	Antimicrobial[33]
15	Octadecane	Alkane	Antibacterial and antifungal activity[26] Posess anti-inflammatory, detoxification,cough, lung disease, fever ,cold ,antioxidant, antisepsis and anticorrosion activities, [35]
16	Pentadecanoic acid, 14-methyl-methylester	FAME	Antioxidant[37]
17	n-Hexadecanoic acid	Fatty acid	Anti-inflammatory, antioxidant, cancer as well as hypocholesterolemic acid preventive activities[38] Nematicide, pesticide, lubricant, antiandrogenic activity[31]
18	1-Octadecene	Alkene	Antibacterial, antioxidant and anticancer activity39
19	Methoxyacetic acid, 2-tradecylester	Fatty-acid ester	Antibacterial activity[40]
20	Pentadecafluorooctanoic acid, octadecyl ester	Fatty-acid ester	No activity reported

21	9-Octadecenoic acid	Fatty acid	Antimicrobial activity[31]
22	Cyclohexane,butyl	Cycloalkane	No activity reported
23	1-Eicosanol	Alcohol	Antimalarial, antifungal and antioxidant activities[31]
24	Cis-13-Octadecenoic acid, methyl ester	FAME	Therapeutic uses in medicine and surgery[41]
25	Cis-Vaccenic acid	MUFA	Anti-hypocholesterolemic and anti-inflammatory activities[41]
26	Methyl Stearate	FAME	Antihelminitic(nematodes), inhibits GABA aminotransferase, antinociceptive, regulates intestinal lipid metabolism, anti-inflammatory, inhibits gastric acid activities[42]
27	9-Octadecanoic acid,	Fatty acid	Antimicrobial activity[31]
28	Cyclohexadecane, 1,2-diethyl	Cycloalkane	No activity reported
29	9-Octadecenoic acid	Fatty acid	Antimicrobial activity[31]
30	Octadecanoic acid	Fatty acid	Antimicrobial activity[31]
31	Oleic acid	MUFA	Antimicrobial activity[43] Antifungal, anti-inflammatory, antioxidant and antibacterial activities[41]
32	Oleic acid	MUFA	Antimicrobial activity[43] Antifungal, anti-inflammatory, antioxidant and antibacterial activities[41]
33	1-Docosene	Alkene	Antibacterial activity[43]
34	Cycloeicosane	Cycloalkane	No activity reported
35	Cyclohexadecane, 1,2 –diethyl	Cycloalkane	No activity reported
36	5-Eicosene	Alkene	Antimicrobial activity[44]
37	3-Eicosene	Alkene	Antimicrobial, anti-hyperglycemic, cytotoxic activity, antioxidant and insecticide activities[35]
38	Heptadecylheptafluorobutyrate	Ester	No activity reported
39	Bis-(2-ethylhexyl) pthalate	Phthalate Ester	Antimicrobial and cytotoxic activities[45]
40	Nonadecyltrifluoroacetate	Ester	No activity reported
41	6,11-Dimethyl-2,6,10-dodecatrien-1-ol	Phenol	Antimicrobial activity[46]

TABLE 6: ACTIVITIES OF BIOACTIVE COMPOUNDS PRESENT IN *U. LOBATA* METHANOL LEAVE EXTRACT

Pk	Bioactive compound	Class of compound	Biological activity
1	Dodecane	Alkane	Antibacterial activity Antifungal activity[47] Enhances antifungal activity. Possess tumor-promoting Activity[25]
2	5-Tetradecene	Alkene	No activity reported
3	Tetradecane	Alkane	Possess tumor-promoting and cocarcinogenic activity[25];Antimicrobial,Diuretic &Antituberculosis[22]; Antifungal[26-28,] Antibacterial, Antipyretic and bronchitis [35]
4	Cyclododecane	Cycloalkane	Serves as a hydrophobic mask, melt and as a temporary consolidant agent[48]
5	2,4-Di-tert-butylphenol	Phenol	Antibacterial, Antimalarial, Anticancer, Antifungal activities[49-52]
6	Undecanoic acid, 10-methyl-, methyl ester	FAME	Antioxidant, increase aromatic amino acid decarboxylase activity[31]
7	Dodecanoic acid	Fatty acid	Antibacterial and Antioxidant activities[32]
8	Dodecanoic acid	Fatty acid	Antibacterial and Antioxidant activities[32]

9	9-Octadecene	Alkene	Antifungal, antioxidant, anticancinogenic and antimicrobial activity[53-54]
10	Hexadecane	Alkane	Antifungal, antibacterial and antioxidant activities[34]Cytotoxicity, antimicrobial, antioxidant, antipyretic, anthelmintic, anti-inflammatory, antidiarrheal, antidiabetic[35]
11	Cyclohexane,1,1'-(1,4-butanediyl) bis-	Cycloalkane	No activity reported
12	Cyclotetradecane	Cycloalkane	Antimicrobial[55]
13	1-Dodecene	Alkene	Antibacterial activity[56]
14	Heptadecane	Alkane	Antibacterial activity[57]
15	Tridecanoic acid, 12-methyl-, methyl ester	FAME	No activity reported
16	1,2,4-Butanetriol, trinitrate	Nitrate ester	An energetic plasticizer[58]
17	Tetradecanoic acid	Fatty acid	Antifungal, antioxidant, cancer preventive, nematicide, hypocholestrolemic, lubricant[31]
18	1-Octadecene	Alkene	Antibacterial activity; Antioxidant activity[39,59] Anticancer activity[60,39]
19	Heptadecane,2 methyl-	Alkane	No activity reported
20	Pentadecanoic acid	Fatty acid	Anticancer[61]
21	Trifluoroacetic acid, pentadecyl ester	Fatty acid ester	No activity reported
22	Hexadecanoic acid, methyl ester	FAME	Promotes aortic dilation[62],Promotes membrane autolysis[63],Inhibits phagocytosis and influences nitric oxide production in some cells[64]
23	9-Hexadecenoic acid	Fatty acid	Antimicrobial[65]
24	n-Hexadecanoic acid	Fatty acid	Cancer preventing activities, anti-inflammatory,antioxidant, hypocholesterolemic [38], Nematicidal, pesticidal, lubricant and anti-androgenic[31]
25	n-Hexadecanoic acid	Fatty acid	Cancer preventing activities, anti-inflammatory,antioxidant, hypocholesterolemic[38], Nematicidal, pesticidal, lubricant and anti-androgenic[31]
26	1-Octadecene	Alkene	Antibacterial, antioxidant and anticancer[39]
27	Heptacosane,1-chloro-	Chloroalkane	No activity reported
28	Heptadecanoic acid	Fatty acid	Antioxidant[66]
29	Butyl eicosyl ether	Ether	No activity reported
30	trans-13-Octadecenoic acid	Fatty acid	Anti-inflammatory activity[67]
31	6-Octadecenoic acid, methyl ester	FAME	Possess strong analgesic, anti-inflammatory and antipyretic activity[68]
32	9-Octadecenoic acid	Fatty acid	Antimicrobial[31]
33	Octadecanoic acid	Fatty acid	Antimicrobial[31]
34	Hexadecanoic acid, 1,1-dimethylethyl ester	FAME	No activity reported
35	1-Docosene	Alkene	Antibacterial activity[43]
36	3-Eicosene	Alkene	Antimicrobial, anti-hyperglycemic, cytotoxic activity, antioxidant, insecticide activity[35]
37	cis-Vaccenic acid	MUFA	Anti-hypocholesterolemic and anti-inflammatory[41]
38	1-Docosene	Alkene	Antibacterial activity[43]
39	cis-Vaccenic acid	MUFA	Anti-hypocholesterolemic and anti-inflammatory[41]

40	Palmitoleic acid	MUFA	Anti-inflammatory; Increases insulin sensitivity, inhibits hepatic steatosis.[69] Ameliorates development of hypertriglyceridemia and hyperglycemia, reduces body weight increase Down-regulates mRNA expression of proinflammatoryadipocytokine genes in mice[70].
41	2-Piperidinone,N-[4-bromo-n-butyl	Delta-lactams	Antimicrobial activity[71]
42	1-Docosene	Alkene	Antibacterial activity[43]
43	1-Docosene	Alkene	Antibacterial activity[43]
44	1-Docosene	Alkene	Antibacterial activity[43]
45	Erucic acid	MUFA	Regulates mesenchymal stem cell differentiation into osteoblasts and adipocyte[72];Broad-spectrum antiviral activity against IAV[73;] Anti-inflammatory/Pro- inflammatory amplification effect.InhibitsNF-κB and p38 MAPK
46	Diisooctyl phthalate	Phthalate ester	Antimicrobial and antifouling[74] Antibacterial[75]
47	1-Docosene	Alkene	Antibacterial activity[43]

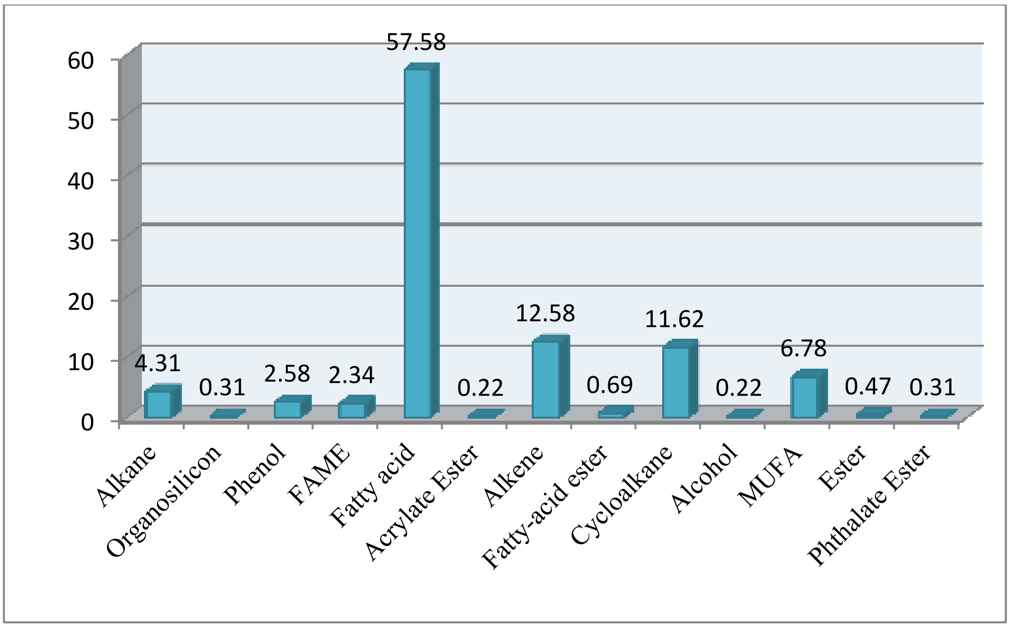


Figure 5: Percentage Abundance of Classes of phytocompounds in of *U. lobata* ethanol leave extract

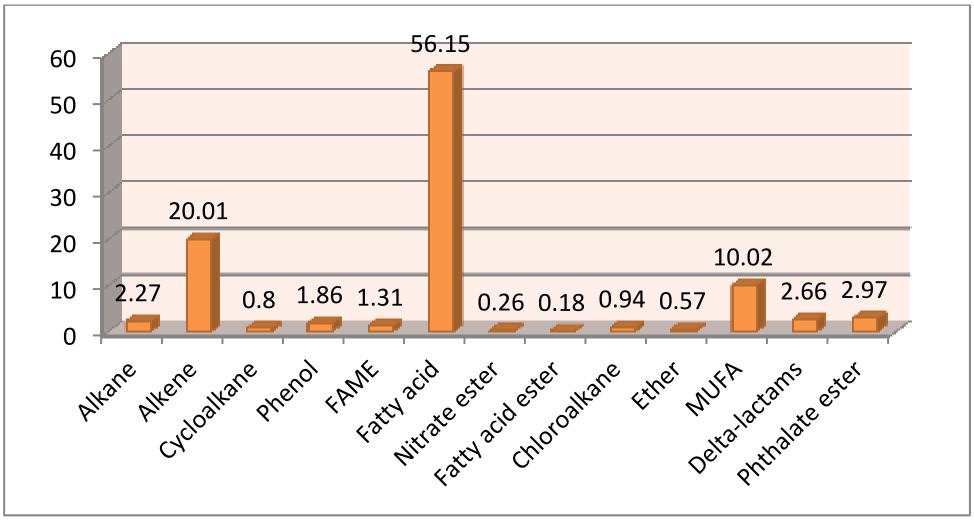


Figure 6: Percentage Abundance of Classes of phytocompounds in *U. lobata* methanol leave extract

Table 7: Bioactive compounds common to ethanol and methanol extracts of *U.lobata* leaves

Bioactive compounds		Percentage Abundance in extraction solvents (%)	
		Ethanol	**Methanol**
1.	Dodecane	0.61	0.15
2.	Undecanoic acid, 10-methyl-, methyl ester	1.11	0.42
3.	Dodecanoic acid	13.43	6.89, 2.53
4.	Hexadecane	0.72	0.96
5.	Tetradecanoic acid	2.14	3.01
6.	n-Hexadecanoic acid	11.73	26.65, 9.11
7.	1-Octadecene	1.11	3.54
8.	9-Octadecenoic acid	16.8, 2.02, 0.91	3.45
9.	cis-Vaccenic acid	0.25	1.59, 1.67
10.	Octadecanoic acid	9.78	0.68
11.	1-Docosene	9.57	2.84,0.56,0.74,6.06,2.04,0.17
12.	3-Eicosene, (E)-	0.27	0.1

Figure 7: Whole plant of *U. lobata*

CONCLUSION

GC-MS and FT-IR investigation of ethanol and methanol leave extracts of *Urena lobata* demonstrated the abundance of bioactive phytochemicals and their inherent functional groups. These bioactive phytocompounds have a wide range of biological and therapeutic actions, lending credence to the plant's ethnomedicinal use for the treatment of various disease conditions amongst ethnic groups.

CONFLICT OF INTEREST

No conflict of interest exist

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