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GC-MS EVALUATION OF BIOACTIVE COMPOUNDS AND ANTIBACTERIAL ACTIVITY OF THE OIL FRACTION FROM THE STEM BARK OF *BRACHYSTEGIA EURYCOMA* HARMS

OKENWA UCHENNA IGWE* and DONATUS EBERE OKWU

Department of Chemistry, Michael Okpara University of Agriculture, Umudike, P.M.B. 7267 Umuahia, Abia State, NIGERIA

ABSTRACT

The ethanolic extract of the stem bark of *Brachystegia eurycoma* yielded reddish coloured substance (5.68 g). The extract was subjected to GC-MS studies. Nineteen phyto-constituents were identified with 4[1], 5-dihydroxy-7-methoxy flavones (21.97%) constituting the bulk of the oil, followed by 9-octadecenoic acid (12.4%). Other compounds indentified include 9,12-octadecadienoic acid (10.16%), hexadecanoic acid (9.50%), 9-octadecenoic acid ethyl ester (9.50%), hexadecanoic acid ethyl ester (4.59%), (4-(2-methyl-piperidine-1-sulfonyl)-phenyl)-(2-methyl-piperidin-1-yl)-methanone (3.93%), 2-O-methl-D-Mannopyranosa (3.61%), 2-hydroxy-5-methylisophthaladehyde (3.28%), 1,2,3-trihydroxyphenol (2.30%), ethyl 2-hydroxybenzyl sulfone (2.30%), 1,2,4-trimethyl-3-nitrobicyclo (3.3.1) nonan-9-one (2.30%), butanal, 4-hydroxy-3-methyl (1.97%), 1,6-anhydro-beta-D-glucopyranose (1.97%), 1,2-benzenedicarboxylic acid, 2-ethoxy-2-oxoethyl ester (1.64%), N,N-diethyl-4-(morpholine-4-sulfonyl)-benzenesulfanamide (1.64%) and 9-octadecenamide (1.31%). The extract showed antibacterial activity against *Escherichia coli*, *Salmonella typhi* and *Staphylococcus aureus*. These results suggest the use of the extract from the stem bark of *Brachystegia eurycoma* in the treatment of typhoid fever, wounds and infections in herbal medicine in Nigeria.

Key words: *Brachystegia eurycoma*, GC-MS analysis, Bioactive Compounds, Antibacterial activity, Phyto-chemicals.

INTRODUCTION

The invaluable use of phyto-chemicals in herbal medicine is as old as man. Most synthetic drugs being used today have their origin from naturally occurring plant chemicals[1]. These chemicals show pharmacological and bioactive activity which may include antioxidant, anti-inflammatory, anticancer, anti-allergic, antibacterial, antiviral, anti-plasmodic and

*Author for correspondence; E-mail: okescoty@yahoo.com; okenwauigwe@gmail.com

estrogenic affects [2]. Bioactive compounds in plants are ubiquitous and a lot of them are yet to be discovered, studied and documented. As part of our effort to research on the bioactive constituents of Nigerian vegetation, *B. eurycoma* has been selected for study.

Brachystegia eurycoma grows mainly along the river banks or swamps in Western and Eastern Nigeria. It also grows on well-drained soils[3]. It is a large tree with irregular and twisted spreading branches. The seed has a roundish flat shape with brown colour and hard hull. The fruit ripens from September to January and is released by explosive mechanism[4]. The fruits occur as broad lathery dark purplish brown pods containing between four and six brown shiny flat disc-like seeds. The plant also possesses a rough fibrous bark, which peels off in patches and often gives out brownish buttery exudates[5,6]. The leaves from *Brachystegia eurycoma* make an excellent browse material for cattle, sheep and goats. In agro-forestry, the tree is suitable as a shade tree and ornamental plant especially in the dry season when the trees produce masses of coloured young foliage. The fruit pods make good fuel wood. Timber sold in international market is also produced from *B. eurycoma*[6]. The seeds of *Brachystegia eurycoma* help in softening bulky stools and have been associated with the protection against colon and rectal cancer[7]. The exudate is used in faster healing of wounds[8]. The exudate in right combination with mucin and honey is used for wound healing, prevention of bacteria infection, scar formation and promotes regeneration of hair follicles[8].

As part of our chemical studies on Nigerian medicinal plants, we hereby describe the chemical constituents of the volatile extract of *Brachystegia eurycoma* and also evaluate the antibacterial activity of the extract against some pathogenic bacteria for possible development of new drugs for the prevention and treatment of infections.

EXPERIMENTAL

GC analysis were carried out in SHIMADZU JAPAN gas chromatography 5890-11 with a fused GC column (OV-101) coated with polymethyl silicon (0.25 mm x 50 m) and the conditions were as follows: Temperature programming from 80-200^{o}C held at 80^{0}C for 1 minute, rate 5^{0}C/min and at 200^{o}C for 20 min. FID temp. 300^{o}C, injection temp. 250^{o}C, carrier gas nitrogen at a flow rate of 1 mL/min, split ratio 1 : 75. GC-MS (Gas chromatography mass spectrometry) analysis was conducted using GCMS-QP 2010 Plus Shimadzu Japan with injector temperature of 230^{o}C and carrier gas pressure of 100 Kpa. The column length was 30 m with a diameter of 0.25 mm and the flow rate of 50 mL/min. The eluents were automatically passed into a mass spectrometer with a dictator voltage set at 1.5 KV and sampling rate of 0.2 sec. The mass spectrum was also equipped with a computer fed mass spectra data bank. Hermle Z 233 M-Z centrifuge Germany was used. Reagents and

solvents like ethanol, chloroform, diethyl ether, hexane, were all of analytical grade and were procured from Merck, Germany. The nutrient agar was purchased from scharian chemie (APHA) Spain.

Plant materials

Brachystegia eurycoma barks were harvested from the tree plant located at Umuovo village Stream in Old Umuahia, Umuahia South Local Government Area of Abia State, Nigeria. The harvested barks (2 Kg) were then dried on the laboratory bench for 30 days.

Extraction of plant materials

The barks of *Brachystegia eurycoma* were milled into a uniform and fine powder by a mechanically driven attrition mill. The powdered plant sample (300 g) was successfully extracted with 2 L of benzene (8 hrs/3 times/80^{o}C) followed by 2 L of ethanol (8 hrs/3 times/ 65^{o}C). The extracts were concentrated under reduced pressure and the supernatant reddish extract was decanted (6.68 g) after complete removal of the solvent. The extract was centrifuged at 10,000 rpm for 20 minutes and the clear supernatant extract was subjected to systematic GC-MS analysis.

Component identification

The components of the extract were identified by matching the peaks with computer Wiley MS libraries and confirmed by comparing mass spectra of the peaks and those from literature[9].

Bioassay

The *in vitro* antibacterial activity of the extract was carried out for 24 h culture of three selected bacteria. The bacteria organisms used were *Escherichia coli, Salmonella typhi* and *Staphylococcus aureus.* All the test organisms were clinical isolates of human pathogens obtained from stock cultures at the Central Laboratory Services Unit of National Root Crops Research Institute, Umudike, Abia State, Nigeria. With the aid of a single hole punch office paper perforator, circular discs of 5 mm diameter were cut from whatman No. 1 filter paper. The paper discs were boiled in distilled water for an hour to remove any residual preservatives. The boiled paper discs were allowed to drain dry and they were wrapped in aluminum foil and sterilized in an autoclave at 121^{o}C for 15 minutes. They were however used within 48 hours of production. The sensitivity of each test microorganism to the extract was determined using the Disc Diffusion Technique[10,11]. A loopful of each test sample organism was aseptically transferred into the surface of a sterile solid medium, appropriate for the test organism. Using a flamed glass hockey, the innoculeum was spread evenly over

the surface of the medium. Then with the aid of a flamed pair of forceps, the extract bearing paper discs was carefully placed on the surface of the inoculated medium at some distance from one another. The inoculated plates were incubated for 24 hours in an incubator at 37^{o}C. They were examined daily for growth and for the presence of inhibition zones around the paper discs. The level of sensitivity was determined by the diameter of the inhibition zones as measured with a transparent millimeter rule. The minimum inhibitory concentration (MIC) was determined by comparing the different concentrations of the extract having different zones and selecting the lowest concentration.

RESULTS AND DISCUSSION

The ethanol extract of *Brachystegia eurycoma* stem bark showed nineteen peaks from the chromatogram of the extract. These peaks indicated the presence of nineteen compounds (1-19) in the extract (Fig. 1). The molecular formula, percentage constituents and molecular masses of the compounds are shown in Table 1. These compounds comprise mainly flavonoids, fatty acids, esters, alkaloids, sulphones, aldehydes, phenols and carbohydrates. The composition of the extract was; flavonoid (21.97%), phenols (5.25%), alkaloids (9.18%), sulphone (2.30%), aldehydes (5.25%), esters (18.38%), carbohydrates (5.58%) and fatty acids (32.12%).

Compound **1** was identified as 1,2-benzenediol and has molecular formula of $C_6H_6O_2$ (m/z 110) with base peak at m/z 110. The molecular ion mass also represented the base peak of the compound. The compound comprised 2.3% of the volatile extract. Compound **2** was a sulphone named ethyl 2-hydroxybenzyl sulphone. It has molecular formula of $C_9H_{12}O_3S$ (m/z 200) and base peak at m/z 107 which resulted because of the cleavage of a $C_3H_7O_2S$ group from the compound. The compound comprised 2.30% of the volatile extract. Compound **3** was identified as 1,2,3-trihydroxybenzene (pyrogallol) with a molecular formular of $C_6H_6O_3$ (m/z 126) and a base peak at m/z 126. The base peak at m/z 126 was due to the molecular ion of the compound. The compound comprised 2.95% of the volatile extract. Compound **4** was an aldehyde identified as 4-hydroxy-3-methylbutanal with a molecular formula of $C_5H_{10}O_2$ (m/z 102) and a base peak at m/z 56 due to the loss of a C_3H_4O molecule fro the compound. Its composition in the volatile extracts was 1.97%. Compound **5** was a carbohydrate molecule identified as 1,6-anhydro-beta-D-glucopyranose. It has a molecular formula of $C_6H_{10}O_5$ (m/z 162) and a base peak of m/z 60 due to the loss of a $C_2H_4O_2$ group from the compound. The compound comprised 1.97% of the extract. Compound **6** was an ester identified as 1,2-benzenedicarboxylic acid, 2-ethoxy-2-oxoethyl ethyl ester with a molecular formula of $C_{14}H_{16}O_6$ (m/z 280) and a base peak at m/z 149. The base peak was a result of cleavage of a $C_9H_9O_2$ fragment from the compound. It comprised 1.64% of the volatile extract. Compound 7 was a carbohydrate molecule identified as 2-O-

methylhexose with a molecular formula of $C_7H_{14}O_6$ (m/z 194) and a base peak at m/z 87. The compound comprised 3.61% of the volatile extract. Compound **8** has a molecular formula of $C_9H_8O_3$ (m/z 164). It was identified as 2-hydroxy-5-methylisophthalaldehyde and comprised 3.28% of the volatile extract. It has a base peak at m/z 136. Compound **9** was a fatty acid identified as n-hexadecanoic acid (palmitic acid) with a molecular formula of $C_{16}H_{32}O_2$ (m/z 256) and a base peak at m/z 43 which occurred as a result of the cleavage of a C_3H_7 (propyl group) fragment from the compound. The compound comprised 9.50% of the volatile extract.

Compound **10** comprised 4.59% of the volatile extract. It was identified as hexadecanoic acid ethyl ester with a molecular formula of $C_{18}H_{36}O_2$ (m/z 284) and a base peak at m/z 88. The base peak occurred as a result of McLafferty re-arrangement leading to the detachment of CH_2=C (OH)OC_2H_5 group from the compound. Compound **11** comprised 2.62% of the volatile extract with a molecular formula $C_{17}H_{32}O_2$ (m/z 268) and base peak at m/z 55. The compound was identified as 7-hexadecenoic acid methyl ester. The base peak observed at m/z 55 was due to the cleavage of a $C_{14}H_7$ group from the compound. Compound **12** was identified as 9,12-octadecadienoic acid with a molecular formula of $C_{18}H_{32}O_2$ (m/z 280) and a base peak at m/z 67. The compound comprised 10.16% of volatile extract. Compound **13** was identified as 9-octadecanoic acid with a molecular formula of $C_{18}H_{34}O_2$ (m/z 282) and a base peak at m/z 55 due to the loss of a C_4H_7 fragment from the compound. It comprised 12.46% of the volatile extract. Compound **14** was identified as 9-octadecenoic acid ethyl ester with a molecular formula of $C_{20}H_{38}O_2$ (m/z 310) and base peak at m/z 55 due to the loss of a C_4H_7 fragment from the compound. It comprised 9.50% of the volatile extract. Compound **15** has a molecular formula of $C_{19}H_{28}N_2O_3S$ (m/z 364) and was identified as (4-(2-methyl-piperidine-1-sulfonyl)-phenyl)-(2-methyl-piperidine-1-yl)-methanone. The compound has a base peak at m/z 349 due to the detachment of $C_{18}H_{25}N_2O_3$ S. It comprised 3.93% of the volatile extract. Compound **16** comprised 1.64% of the volatile extract. It was identified as N,N-diethyl-4-(morpholine-4-sulfonyl)-benzenesulfanamide and has a molecular formula of $C_{14}H_{22}N_2O_5S_2$ (m/z 362) and a base peak at m/z 347. The base peak was as a result of the cleavage of $C_{13}H_{19}N_2O_5S_2$ from the compound. Compound **17** was identified as 4^1, 5-dihydroxy-7-methoxy flavone with a molecular formula of $C_{16}H_{12}N_2O_5$ (m/z 284) and a base peak at m/z 284. It means that the molecular ion mass was responsible for the base peak. The compound comprised 21.97% of the volatile extract. The exceptional high quantity of Compound **17** is noteworthy. Compound **18** comprised 1.31% of the volatile extract and was identified as 9-octadecenamide with a molecular formula of $C_{18}H_{35}NO$ (m/z 281) and a base peak at m/z 59. Compound **19** was identified a 1,2,4-trimethyl-3-nitrobicyclo (3.3.1) nonan-9-one with a molecular formula of $C_{12}H_{19}NO_3$ (m/z 225) and a base peak at m/z 95. The compound comprised 2.30% of the volatile extract.

Table 1: GC-MS analysis of ethanol fractions from the stem barks of *Brachystegia eurycoma*, showing the fragment ion peaks and retention time

Chromato-gram peak	Compound name	Molecular formula	Mol. weight	Retention time (mins)	Peak height (cm)	Percentage content (%)	Fragment peak (m/z) and % abundance
1	O-Hydroxyphenol	$C_6H_60_2$	110	12.6	0.7	2.30	27 (5%), 39 (5%), 53 (14%), 64 (33%) 81 (19%), 92 (14%), 110 (100%)
2	Ethyl 2-hydroxybenzyl sulfone.	$C_9H_{12}0_3S$	200	15.5	0.7	2.30	50 (5%), 62 (2%), 77 (24%), 107 (100%).
3	1,2,3-Trihydroxybenzene	$C_6H_6O_3$	126	16.1	0.9	2.95	40 (29%), 55 (19%), 68 (19%), 70 (19%), 97 (95%), 126 (100%)
4	Butanal, 4-hydroxy-3-methyl.	$C_6H_{10}0_2$	102	16.8	0.6	1.97	15 (2%), 27 (19%), 41 (71%), 56 (10%), 72 (19%), 85 (10%)
5	1,6-Anhydro-beta-D-glucopyranose.	$C_6H_{10}0_5$	162	17.4	0.6	1.97	41 (10%), 43 (19%), 60 (100%), 73 (38%), 97 (14%)
6	1,2-Benzenedicarboxylic acid, 2-ethoxy-2-oxoethyl ethyl ester.	$C_{14}H_{16}0_6$	280	19.4	0.5	1.64	27 (10%), 29 (43%), 50 (14%), 65 (10%), 76 (33%), 93 (5%), 104 (29%), 121 (5%), 132 (5%), 149 (100%), 177 (38%), 235 (19%)
7	2-0-methyl-D-mannopyranosa.	$C_7H_{14}0_6$	194	21.0	1.1	3.61	27 (10%), 31 (29%), 45 (24%), 59 (14%), 74 (71%), 87 (100%), 116 (10%) 145 (55%)

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Chromato-gram peak	Compound name	Molecular formula	Mol. weight	Retention time (mins)	Peak height (cm)	Percentage content (%)	Fragment peak (m/z) and % abundance
8	2-Hydroxy-5-methylisophthaladehyde	$C_9H_8O_3$	164	22.7	1.0	3.28	27 (10%), 39 (19%), 51 (24%), 63 (10%), 77 (48%), 90 (57%), 107 (38%), 118 (5%), 136 (100%), 164 (43%)
9	n-hexadecanoic acid	$C_{16}H_{32}O_2$	256	24.5	2.9	9.50	7 (19%), 41 (76%), 43 (100%), 60 (90%), 73 (92%), 83 (14%), 98 (10%), 115 (10%), 129 (20%), 157 (5%), 171 (5%), 185 (5%), 213 (20%)
10	Hexadecanoic acid,ethyl ester	$C_{18}H_{36}0_2$	284	25.9	1.4	4.59	27 (5%), 41 (24%), 57 (20%), 73 (15%), 88 (100%), 101 (57%), 115 (5%), 129 (2%), 143 (5%), 157 (14%), 239 (5%)
11	7-Hexadecenoic acid, methyl ester	$C_{17}H_{32}0_2$	268	27.3	0.8	2.62	41 (90%), 55 (100%), 69 (48%), 74 (67%), 87 (43%), 98 (29%), 123 (14%), 137 (10%), 152 (10%), 194 (10%)
12	9,12-Octadecadienoic acid	$C_{18}H_{32}0_2$	280	27.6	3.1	10.16	29 (24%), 41 (71%), 67 (100%), 81 (67%), 95 (43%), 109 (20%), 123 (10%), 135 (10%), 149 (10%), 163 (5%)
13	9-Octadecenoic acid	$C_{18}H_{34}0_2$	282	27.7	3.8	12.46	27 (14%), 41 (71%), 55 (100%), 69 (71%), 83 (57%), 97 (23%), 98 (25%), 125 (10%), 151 (5%), 264 (14%)

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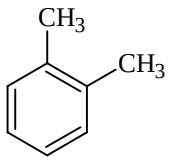
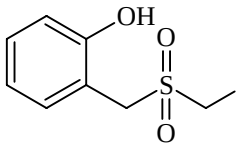
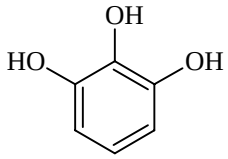
Chromato -gram peak	Compound name	Molecular formula	Mol. weight	Retention time (mins)	Peak height (cm)	Percentage content (%)	Fragment peak (m/z) and % abundance
14	9-Octadecenoic acid ethyl ester	$C_{20}H_{38}0_2$	310	28.1	2.9	9.50	41 (81%), 559 (100%), 69 (57%), 83 (43%), 88 (52%), 101 (29%), 123 (10%), 137 (5%), 151 (5%), 180 (10%), 222 (10%), 264 (14%)
15	(4-(2-methyl-piperidine-1-sulfonyl)-phenyl)-(2-methyl-piperidin-1-yl)-methanone	$C_{18}H_{28}N_2O_3S$	364	32.8	1.2	3.93	41 (10%), 55 (14%), 76 (24%), 84 (5%), 104 (52%), 125 (5%), 132 (2%), 146 (5%), 146 (5%), 174 (29%), 187 (5%), 202 (20%), 218 (5%), 250 (5%), 266 (43%), 349 (100%)
16	N,N-Diethyl-4-(morpholine-4-sulfonyl)-benzenesulfanamide	$C_{14}H_{22}N_2O_5S_2$	362	333	0.5	1.64	41 (5%), 56 (14%), 76 (5%), 86 (10%), 140 (5%), 197 (10%), 226 (14%), 290 (10%), 347 (100%)
17	Flavone,4^1,5-dihydroxy-7-methoxy	$C_{16}H_{12}0_5$	284	35.5	6.7	21-97	29 (2%), 41 (5%), 69 (5%), 83 (5%), 95 (10%), 110 (5%) 118 (5%), 138 (5%), 166 (5%), 241 (14%), 255 (30%), 284 (100%)
18	9-Octadecenamide	$C_{18}H_{35}N0$	281	30.2	0.4	1.31	41 (30%), 55 (40%), 59 (100%), 72 (62%), 86 (10%), 98 (10%), 112 (10%), 126 (10%), 140 (5%), 154 (5%), 281 (10%)
19	1,2,4-Trimethyl-3-nitrobicyclo (3.3.1) nonan-9-one	$C_{12}H_{19}N0_3$	225	46.1	0.7	2.30	27 (19%), 41 (76%), 55 (57%), 69 (86%), 81 (62%), 95 (100%), 109 (43%), 121 (10%), 135 (5%), 151 (14%), 179 (5%)



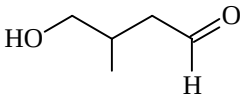
[1**] O-Hydroxyphenol**



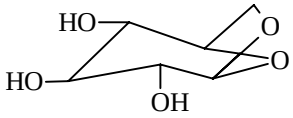
[2**] Ethyl 2-hydroxybenzyl sulfone**



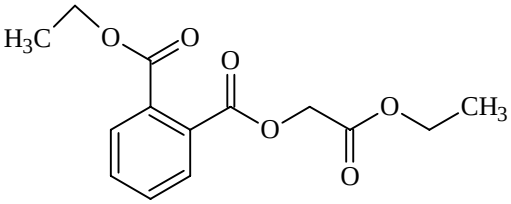
[3**] 1,**2,3**-Trihydroxybenzene (Pyrogallol)**



[4**] Butanal, 4-hydroxyl-3-methyl**



[5**] 1,**6**-Anhydro-beta-D-glucopyranose**



[6**] 1,**2**-Benzenedicarboxylic acid, 2-ethoxy-2-oxoethyl ethyl ester**

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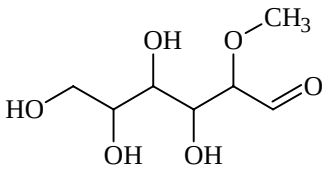
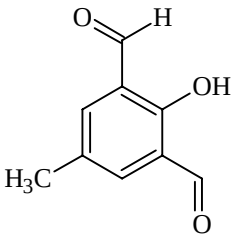
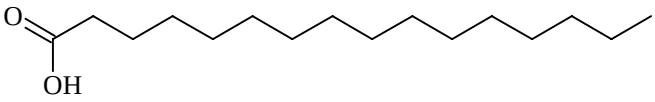


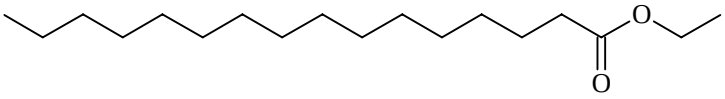
[7**] 2-O-Methyl-D-mannopyranosa**



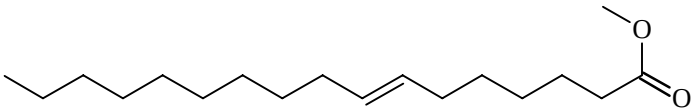
[8**] 2-Hydroxy-5-methylsophthladehyde**



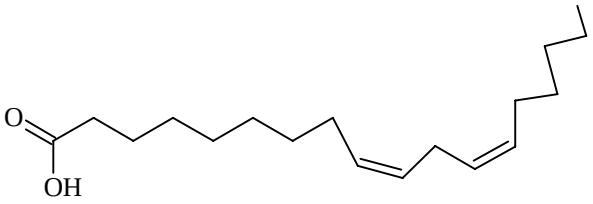
[9**] n-Hexadecanoic acid**



[10**] Hexadecanoic acid, ethyl ester**



[11**] Hexadecanoic acid, methyl ester**



[12**] 9,**12**-Octadecadienoic acid**

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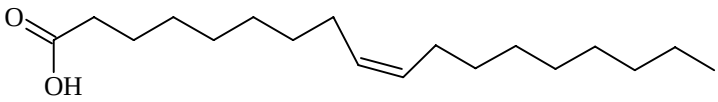
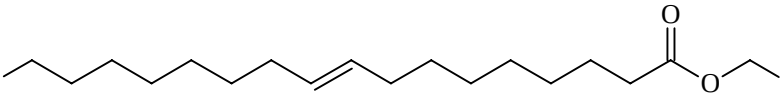
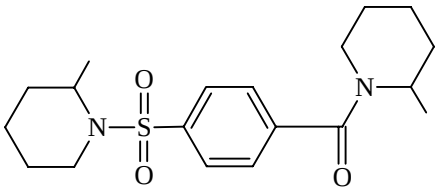


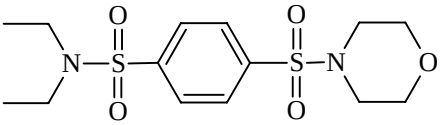
[13**] 9-Octadecadienoic acid**



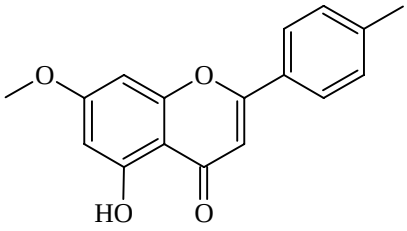
[14**] 9-Octadecedienoic acid ethyl ester**



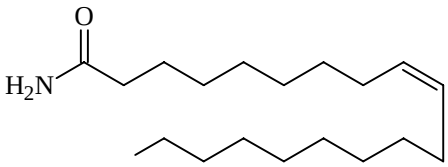
[15**] (**4-(2-**Methyl-piperidine-1-sulfony)-phenyl)-(**2-**methyl-piperidin-1-yl)-methanone**



[16**] N,N-Diethyl-4-(morpholine-4-sulfonyl)-benzenesulfanamide**

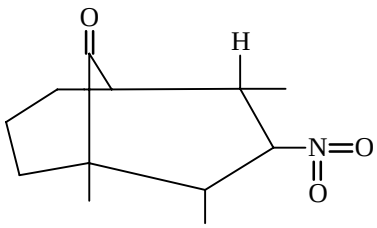


[17] Flavone, 4[1],5-dihydroxy-7-methoxy



[18**] 9-Octadecenamide**

Cont…



[19**] 1,**2,4-**Trimethyl-3-nitrobicylo (**3.3.1**) nonan-9-one**

Fig. 1: Structures of the compounds from GC-MS analysis of the extract from the stem bark of *Brachystegia eurycoma*.

4[1],5-dihydroxy-7-methoxy flavone was the highest of all the nineteen components of the volatile extract of the bark of *Brachystegia eurycoma*. It is a flavonoid compound. The presence of flavones and other phenolic compounds in the extract suggest its therapeutic potentials in phyto-medicine for the treatment of a lot of diseases and infections. The flavones seem to be the most powerful flavonoids for protecting the body against reactive oxygen species (ROS)[12]. Flavonoids can prevent injury caused by free radical in various ways. One way is the direct scavenging of free radicals[13]. Because of high reactivity of the hydroxyl group of phenols and flavonoids, radical are made inactive[13]. The plant might be used in the treatment of atherosclerosis. Flavonoids have been reported to have antioxidant, anti-inflammatory, anti-microbial, anti-allergic, anti-carcinogenic, immune-stimulating and estrogenic effects[14,15]. The detection of flavonoids in the stem bark of *Brachystegia eurycoma* gives evidence to the use of the extract in the treatment of wounds and infections caused by bacteria and fungi.

Fatty acids and alcohols in the plant undergoes esterification reaction to form esters which frequently exudes out of the plant as resins and it may be used in treating wounds and skin infections[16]. Alkaloid constitutes 9.18% of the extract. Many of them are toxic to organisms and are used as anti-microbial agents. Alkaloids often have pharmacological effects and are used as anesthetics, stimulants, analgesics and anti-malarial[17]. Organosulphur compounds (OSCs) prevent or slow down the carcinogenic process induced by a variety of chemical carcinogens. These include inhibition of the carcinogens, dermatitis and other minor wounds[18]. Organosulphur compounds have been reported to have numerous beneficial health effects including protection from oxidative damage[19].

The detection of N,N-diethyl-4-(morpholine-4-sulfonyl)-benzensulfanamide (an alkaloid) in the bark of *Brachystegia eurycoma* is very interesting. Sulfanamides often referred to as sulfa drugs are a group of drugs that were originally designed to treat

infections caused by bacteria and fungi[20]. The most common infectious disease for which sulfa drugs are used is in the treatment of urinary tract infections (UTIS)[20]. Sulfonamides may also be prescribed to treat bronchitis, shigellosis, ear infections, foxoplasmosis, traveler's diarrhea, nocardia infection, bacteria pneumonia and certain eye infections[20]. The presence of the sulfonamide suggests wide applicability of the extract in phyto-medicine. The detection of carbohydrate molecules in the bark of *Brachystegia eurycoma* could mean that the extract while being used as medicine might also provide energy for body metabolism and at the same time help to cushion the side effects which might occur from the various bioactive constituents of the back extract.

The extract from the stem bark of *Brachystegia eurycoma* successfully inhibited *E. coli*, *S. typhi* and *S. aureus* (Table 2). It exhibited highest antibacterial activity against *S. typhi*. The minimum inhibitory concentration (MIC) of the extract was 50%. The microorganisms tested were human commensals and have been incriminated in the infection of wounds[21]. These findings suggest the use of *Brachystegia eurycoma* extracts in the treatment of wounds, typhoid fever and gonorrhea. The antibacterial activity exhibited by the extracts could be a synergistic effect of flavonoids, alkaloids, phenols and sulfonamides on the organism. The mechanism of inhibitory action may be due to impairment of variety of enzymes system including those involved in energy production, interference with the integrity of the cell membrane and structural component synthesis[21]. The use of the extract from the stem bark of *Brachystegia eurycoma* in treatment of wounds and infections has been authenticated by this research. The extract possesses a high potency against bacterial infections by inhibiting their growth and accelerating wound healing.

Table 2: Inhibitory effects of the extract from the stem bark of *Brachystegia eurycoma*

Pathogens	Concentration (%)			MIC (%)
	50	75	100	
Escharichia coli	9.33	12.33	15.00	50
Salmonella typhi	10.67	12.33	14.33	50
Staphylococcus aureus	9.33	11.67	14.50	50

MIC= Minimum inhibitory Concentration

Figures are in mm and include the diameter of the paper disc (5 mm). Data are means of triplicate determinations.

REFERENCES

1. J. C Winston, Herbal Remedies that Promote Health and Prevent Disease, CRC Press, Boca Raton, London (2001) pp. 179-204.
2. D. E. Okwu, Nigerian Medicinal Plants II, Med. and Arom. Plant Sc. Biotech., **26**, 1-4 (2006).
3. D. E. Okwu and E. Okoro. Phyto-chemical Composition of *Brachystegia eurycoma* and *Mucuna flagellipes* Seeds, J. Med. and Arom. Plant Sc. Biotech., **26**, 1-4 (2006).
4. A. Uzomah and R. N. Ahihigwo, Studies on the Rheological Properties and Functional Potentials of Achi (*Brachystegia eurycoma*) and Ogbono (*Irvingia gabonesis*) Seed Gums, Food Chemistry, **67**, 217-228 (1999).
5. R. N. J. Keay, Trees of Nigeria, Oxford University Press, New York (1989) p. 112.
6. N. J. Enwere, Foods of Plant Origin, Afron Orbis Publication Ltd, Nsukka, Nigeria (1998) pp. 64-65.
7. M. C. Ndukwu, Determination of Selected Physical Properties of *Brachystegia eurycoma* Seeds. Res. Agric. Eng., **55(4)**, 165-169 (2009).
8. M. U. Adiukwu and T. C. Enebeke, Evaluation of Snail Mucin Dispersed in *Brachystegia eurycoma* Gum Gel as a Wound Healing Agent, Animal Research International, **4(2)**, 685-697 (2007).
9. A. J. D. Brittto and P. A. S. Selvakymari, GC-MS Study of the Oil Fraction of Methanol Extract of the *Corm Typhonium roxburghii*, J. Med. and Arom. Plant Sc., **28**, 578-579 (2006).
10. M. Pelczar and E. C. S. Chan, Laboratory Exercises in Microbiology, Black Dot. Inc., New York (1977) p. 425.
11. M. Cheesbrough, Medical Laboratory Manual for Tropical Countries, Educational Publishers (2000) p. 447.
12. B. Stravic, T. I. Matula and R. Klassen, Effect of Flavonoids on Mutagenicity and Bio-Availability of Xenobiotics in Foods, in Phenolic Compounds in Food and their Effects on Health. Antioxidants and Cancer Prevention, ACS Synip. Ser., **11**, 507 (1992).
13. R. J. Nijveldt, E. Nood and D. Hoorn, Flavonoids: A Review of Probable Mechanism of Action and Potential Applications, Am. J. Clin. Nutr., **78**, 418-425 (2001).

14. C. T. Ho, Q. Ceri, H. Shi, K. Q Zhang and R. T. Rosen, Anti-Oxidative Effect of Polyphenol Extract Prepared from Various Chinese Teas, Prev. Med., **21**, 520 (1992).

15. D. E. Okwu and Omodamiro, Effect of Hexane Extract and Phyto-chemical Content of *Xylopia aethiopia* and *Ocimum Gratissimum* on Uterus of Guinea Pig, Bio-Research, **3**, 40-44 (2005).

16. D. E. Okwu and B. U. Ighodara, GC-MS Evaluation of Bioactive Compounds and Antibacterial Activity of the Oil Fraction from the Leaves of *Alstonia boonei* De Wild. Der Pharma Chemical, **2(1)**, 261-272 (2009).

17. R. H. Thomson, the Chemistry of Natural Products, Chapman and Hall Publishers (1994) pp. 218-219.

18. D. E. Okwu and N. Ukanwa, Isolation and Characterization of Flavonoids Chakones and Aanthocynidines from *Bridelia Ferruginea* Benth., E-J. Chem., **1(2)**, 21-28 (2010).

19. C. Dwivedi, L. M John, D. S. Schmids and F. N. Encjineer, Effects of Oil-Soluble Organosulphur Compounds from Garlic on Doxorubicin-Induced Lipid Peroxidation. Anti-cancer Drugs, **9**, 291-294 (1998).

20. Health and Medicinal Information Produced by Doctors, www.medicinenet.com-google search (2011).

21. D. E. Okwu and F. N. 1. Morah, the Potentials of *Garcinia Kola* Seed as a Source of Nutraceuticals, J. Med. Arom. Plant. Sci., **28**, 605-611 (2006).

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