

Identification of the Bioactive Phytochemicals in the Ethanol Leaf Extract of *Alafia Barteri* (Apocyanaceae)

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ABSTRACT: *Alafia barteri* is a plant species belonging to the family Apocyanaceae. It is native to the West and Central Africa, stretching from Guinea Bissau to Cameroon, Congo and Nigeria. It is valued for its effectiveness in the traditional medicine system in Nigeria and other African countries. The objective of the present study was to investigate the phytochemical constituents of the leaf of *Alafia barteri* using gas chromatography and mass spectroscopy analysis (GC-MS). The GC-MS analysis revealed the presence of 70 phytochemical components in the ethanol leaf extract of *Alafia barteri*. The prevailing bioactive compounds include: 2-pyrrolidinone (14.03%), cyclotetrasiloxane (12.56%), 9,12-octadecanoic acid(8.12%), n-Hexadecanoic acid(7.97%). The compounds were identified by comparing measured mass spectral data with those in NIST 14 Mass Spectral Library. The results of the present study suggest that the plant leaf can be used as a valuable source in the field of herbal drug discovery.

KEY WORDS: *Alafia barteri*, gas chromatography-mass spectrometry, phytochemicals

INTRODUCTION

Plants whose roots, stems, leaves and seeds have therapeutic effects are referred to as medicinal plants. Medicinal plants have been used in traditional medicine for the prophylactic or therapeutic treatments of different diseases over the years and so have attracted interest. The World Health Organization (WHO) estimates that 80% of the population rely on use of medicinal plants. They serve as sources of potent drugs due to the presence of some bioactive components and as such are useful for drug development. Many species of medicinal plants have been reported to possess pharmacological activities due to the presence of phytochemicals.

There is a growing evidence of a correlation between a medicinal plant's phytochemical constituents and its pharmacological activity (Turker and Usta, 2008). Screening of active compounds from plants has led to the invention of new medicinal drugs and they have an efficient protection against various diseases (Sheeja and Kuttan 2007). *Alafia barteri* Olive (Apocynaceae) commonly known as *Alafia* chewing stick and guinea fowl's crest is a vigorous climbing shrub with stems that scramble over the ground or climb up into trees. Locally named *agbari-etu* and *ibo-agba* (yoruba), *ota nza*(igbo) *momunimo*, *ndambi*, it is native to the West and Central Africa, stretching from Guinea Bissau to Cameroon, Congo and Nigeria (Irvine, 1961). It is valued for its importance in the traditional medicine system in Nigeria and other African countries. Previous literature evidences reveal that the leaf extracts possess antibacterial and antifungal activities (Adekunle and Okoli, 2002; Hamid and Aiyelaagbe, 2011). The root and leaf decoctions are used for toothache and eye infections (Odugbemi, 2008); fiber from the stem of the plant serves as tying material for roofs; aqueous leaf extract displayed potent antiplasmodial activity (Lasisi *et al.*, 2012). The root extract showed analgesic effect (Ishola *et al.*, 2015) while the stem extract of the plant has also been reported to show anti-proliferative activity (Hamid and Aiyelaagbe, 2017). Furthermore, the leaf extracts reduced the blood glucose level of the diabetic animals and maintained historenal architecture (Atilade, *et al.*, 2018); its effect on spermatogenesis and steroidogenesis has been studied by Adalakun *et al.*, 2018. The ethanol leaf and root extracts of *Alafia barteri* have also been shown to possess anti-inflammatory activity using the carrageenan induced paw edema model (Sofidiya and Akindele, 2014; Ishola *et al.*, 2015). In addition, the ethanol leaf and root extracts was found to possess antiarthritic potential (Kolawole *et al.*, 2021). Gas chromatography-mass spectrometry (GC-MS) is an analytical technique for separating mixtures of volatile organic compounds and identifying each constituent compound in plant samples (Eleanora *et al.*, 2018). In the last few years, gas chromatography mass spectrometry (GC-MS) techniques have been used for the identification of bioactive components in both plant and non-plant species. A detailed literature review on the plant in investigation has shown that so far there are no published reports, related to the possible chemical components of "*Alafia barteri*" ethanol leaf extract. Hence, the present study aims at investigating the possible chemical components present in the ethanolic leaf extract of *Alafia barteri* by subjecting it to GC-MS analysis, providing a scientific basis for its traditional use.

MATERIALS AND METHODS

Collection and authentication of the plant material

The complete plant material of *Alafia Barteri* was collected in Ile-Ife, Osun State, Nigeria in January 2021 identified at the Department of Botany, University of Lagos by Dr. Nodza George, where a voucher specimen number: LUH 8789 was allotted to it.

Preparation of plant extracts

The leaves were washed, air dried; blended into powdery form. 60 grams was weighed into a glass jars containing 300ml of ethanol. The mixture was then macerated at room temperature for 72 hours with occasional vigorous stirring. Filtration was carried out using a muslin cloth and the filtrate was concentrated using a rotary evaporator. The concentrated extracts were then transferred to air-tight containers, corked and preserved in the refrigerator at 4°C until required for the experiment.

Gas Chromatography-Mass Spectrometry Analysis

GC-MS analysis was performed using 8860A gas chromatograph coupled to 5977C inert mass spectrometer with electron impact source (Agilent Technologies). The stationary phase of separation of the compounds was carried out on HP-5 capillary column coated with 5% of Phenyl Methyl Siloxane (30 m length × 0.32 mm diameter × 0.25 µm film thickness) (Agilent Technologies). The carrier gas was helium used at a constant flow rate of 1.573 ml/min, an initial nominal pressure of 1.9514 psi and at an average velocity of 46 cm/s. One microliter of the samples were injected with 50:1 Split mode at an injection temperature of 300°C. Purge flow was 21.5 ml/min at 0.50 min with a total gas flow rate of 23.355ml/min; gas saver mode was switched on. The oven was initially programmed at 40°C (1 min), then ramped at 10°C/min to 270°C (4 min). Run time was 30.25 min with a 3 min solvent delay. The mass spectrometer was operated in electron-impact ionization mode at 70eV with ion source temperature of 230°C, quadrupole temperature of 150°C and transfer line temperature of 280°C. Scanning of possible compounds was from m/z 50 to 550 amu at 2.62s/scan scan rate and were identified by comparing measured mass spectral data with those in NIST 14 Mass Spectral Library. The names and molecular weights of the components of the test materials were ascertained finally.

RESULTS

The GC chromatogram of the extract presented in Figure1 shows the retention time in the column and the detected peaks which correspond to the bioactive compounds present in the extract. The bioactive compounds present in the ethanol leaf extract of *Alafia barteri* are shown in Table 1. The identification was based on their elution order in the column. The elution time and the amount of these bioactive compounds were also presented. Based on abundance, the top 4 major compounds present in the ethanol leaf extract include: 2-pyrrolidinone(14.03%), cyclotetrasiloxane(12.56%), 9,12-octadecadienoic acid(8.12%), n-hexadecanoic acid(7.97%).

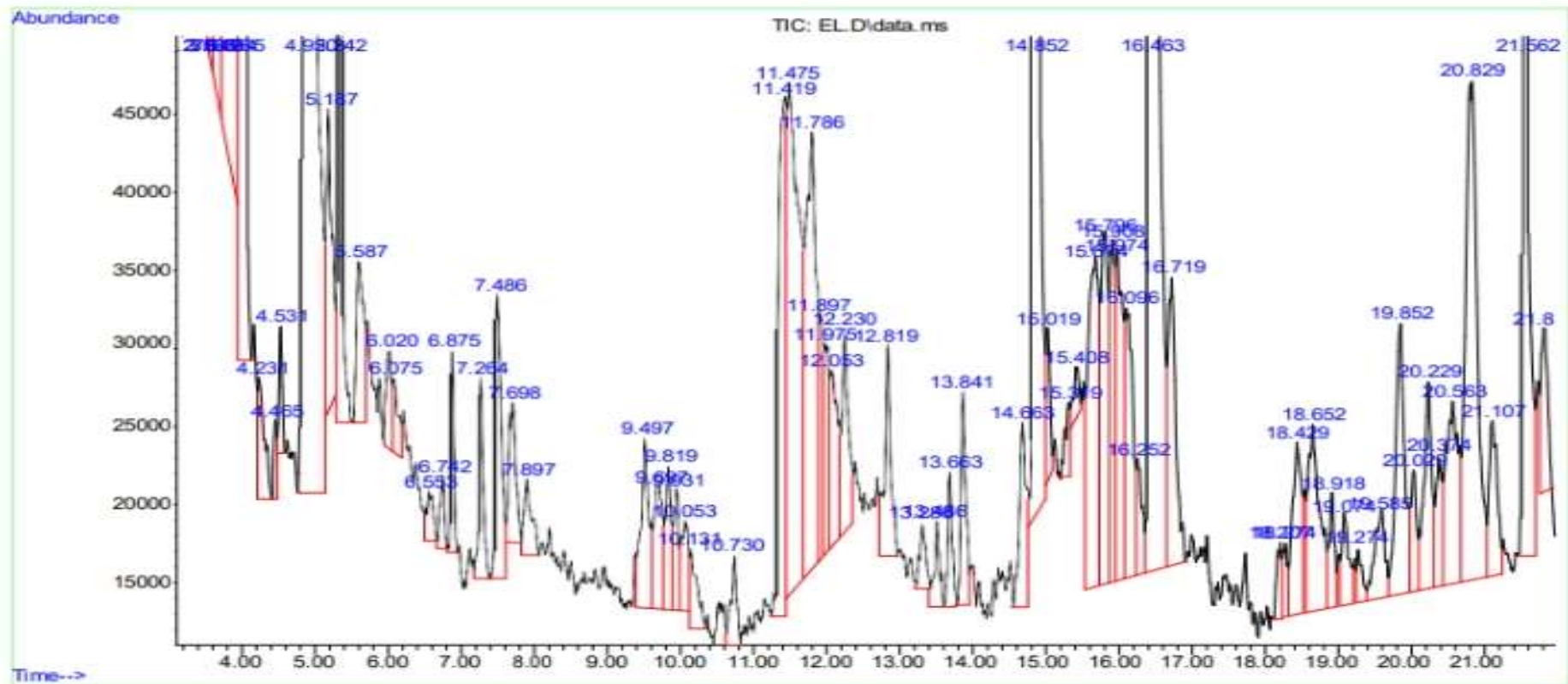


Figure 1: Phytocomponents in the ethanol leaf extract of *Alafia barteri* by GC-MS

Table 1: Phytocomponents identified in the ethanol extract of *Alafia barteri* leaves

Peak No	Retention Time,	Area %	Compound	Molecular Formula	Molecular Weight
1	3.276	12.56	Cyclotrasiloxane 2,6-Dihydroxyacetophenone 2TMS derivative		152.15g/mol)
2	3.565	0.37	Fluoroethyl acrylate Undecanone	C ₅ H ₇ FO ₂ C ₁₁ H ₂₂ O ₂₅	118.11 136.21
3		0.99	2,4-dimethylpent-3-yl ester 2-Pentyn-1-ol Formic acid, methylpropyl ester	C ₅ H ₁₀ O	102.13
4	4.99	4.99	1-Hexene, 1-chloro-, 2-(E)- Dimethyl ether 2-Aminocyclohexanol	C ₆ H ₁₁ Cl C ₈ H ₁₄ O C ₆ H ₁₃ NO	118.61 126.19 115.17
5	3.965	3.80	3-mercapto-Cyclohexanol,	C ₁₈ H ₁₆ O ₅	160.28
6	4.231	0.42	Oxirane, (3-methylbutyl)-2-Undecanone, 6,10-dimethyl-3-Methoxy-4,5-methylenedioxy- N-methylamphetamine	C ₂ H ₄ O	48.08
7	4.465	0.15	p-Cresol 1,2-Cyclooctadiene	C ₇ H ₈ O	108.14

				C ₈ H ₁₂	108.18
8	4.531	0.21	Mequinol, 2-Cyclopenten-1-one, 2,3,4-trimethylPhenol, 2-methoxy-	C ₇ H ₈ O ₂ C ₅ H ₆ O	124.14 82.10
9	4.920	14.03	2-Pyrrolidinone	C ₄ H ₇ NO	85.1045
10	5.187	0.92	Piperazine	C ₄ H ₁₀ N ₂	86.12
11	5.342	1.27	Cyclopentasiloxane, decamethyl-	C ₁₀ H ₃₀ O ₅ Si ₅	370.77
12	5.587	0.65	Propanenitrile, 3-(5-diethylamino-1-methyl-3-pentynyloxy)- 2,3,4,6-Tetrafluorophenyl isothiocyanate 4-Bromo-2,6-difluoroaniline	C ₃ H ₅ N	55.0785
13	6.020	0.31	Hexanoic acid	C ₆ H ₁₂ O ₂	
14	6.075	0.20	alpha.-Pyrrolidinopropiophenone Benzoic acid	C ₆ H ₅ COOH	122.12
15	6.553	0.17	3-Chloro-1,1,2,2-tetrafluoropropan 2-Propylthiazole 4-Hexanoic acid, 2-methylpropyleste	C ₃ H ₃ ClF ₄ C ₆ H ₉ NS C ₈ H ₁₄ O ₂	136.48 127.21 142.20

16	6.742	0.17	1-Tri(isobutyl)silyloxytridecane Silane, Diethyl (trans-4 methylcyclohexyloxy) undecyloxy 1H-Pyrazole-1-acetamide, 4-iodo-N- (4-pyridinylmethyl)-	$C_{18}H_{36}O_2Si$	312.56
17	6.875	0.33	9-Borabicyclo[3.3.1]nonane, 9-(1-ethylpropyl)- 4-(2,6,6-trimethyl-2-cyclohexen-1-yl)-Buten-2-one, 2,5-Dichloro-4-methoxy-pyridin-3-ylamine	$C_{13}H_{25}B$ $C_{13}H_{20}O$ $C_5H_4Cl_2N_2O$	192.2 192.3 170.00
18	7.264	0.47	Succinic acid,hept-2-yl oct-1-en 3-yl ester	$C_{19}H_{34}O_4$	326.47
19	7.486	0.95	Cyclotetrasiloxane, octamethyl- trans-4-(2-(5-Nitro-2-furyl)vinyl)-2-quinolinamine 1,1,3,3,5,5,7,7-Octamethyl-7-(2-methylpropoxy) tetrasiloxan-1-ol	$C_8H_{24}O_4Si_4$ $C_{15}H_{11}N_3O_3$ $C_{12}H_{34}O_5Si_4$	296.62 281.27 370.74
20	7.698	0.53	Acetoxyacetic acid, tetradecyl ester Nitrophthalhydrazide Succinic acid, isobutyl ester	$C_{18}H_{34}O_4$ $C_8H_5N_3O_4$ $C_{12}H_{22}O_4$	314.46 207.14 230.30

21	7.897	0.31	N-Methyl-1-adamantaneacetamide	$C_{13}H_{21}NO$	207.31
			2-quinoxalinamine 3-chloro-N-ethyl-	$C_{10}H_{10}ClN_3$	207.66
			1,2,5-Oxadiazole-3-carbonitrile	C_3HN_3O	95.06
22	9.497	0.75	3-Ethyl-3-methoxypyrazine	$C_7H_{10}N_2O$	138.17
			2,5-Dihydroxybenzaldehyde	$C_7H_6O_3$	138.12
			3-Methoxy-5 methylphenol	$C_8H_{10}O_2$	138.16
23	9.697	0.57	2,3-Methylenedioxyphenol	$C_7H_6O_3$	138.12
			1,2,4,5-Benzenetetramine	$C_6H_{10}N_4$	138.17
			2,5-Cyclohexadiene-1,4-dione, dioxime	$C_6H_6H_2O_2$	138.12
24	9.819	0.44	2,3-Dihydroxybenzaldehyde	$C_7H_6O_3$	138.12
			Norethindrone , O-methyloxime	$C_{21}H_{29}NO_2$	327.5
25	9.931	0.32	5-Fluoro-2-(trifluoromethyl)benzamide	$C_8H_5F_4NO$	207.13
			Terephthalic acid,		

			Carbendazim	C ₈ H ₆ O ₄ C ₉ H ₉ N ₃ O ₂	166.13 191.19
26	10.053	0.34	2(5H)-Furanone,4,5,5-trimethyl-3-(3-methyl-2-methylenebutyl)- dimethyl-mercapto-arsine 2,5Dihydroxybenzaldehyde	C ₁₃ H ₂₀ O ₂ C ₂ H ₇ AsS C ₇ H ₆ O ₃	208.30 138.06 138.12
27	10.131	0.27	6-(2-Aminoethoxy)-2H-1,3-benzodioxole Cyclohexa-2,5-diene-1,4-dione,	C ₉ H ₁₁ NO ₃ C ₈ H ₈ O ₃	181.19 152.13
28	10.730	0.28	1-Benzazirene-1-carboxylic acid, 2,2,5a-trimethyl-1a-[3-oxo- 1-butenyl] perhydro-, methyl ester 1H-Indole, 5-methyl-2-phenyl-	 C ₁₅ H ₁₃ N	 207.27
29	11.419	2.27	Benzeneacetic acid, 2,5-dihydroxy-5,5 Dimethyl-3-vinyl cyclohex-2-en-1-one 4(5H)-Benzofuranone, 6,7-dihydro-3,6-dimethyl-, (R)-	 C ₁₀ H ₁₂ O ₂	 164.20

30	11.475	3.29	Benzeneacetic acid, 2,5-dihydroxy-Formic acid phenyl ester 4(5H) Benzofuranone, 6,7-dihydro-3,6-dimethyl-, (R)-	(C ₉ H ₁₀ BrNO ₂ ; C ₁₀ H ₁₂ O ₂	136.1479) 164.20
31	11.786	2.50	Benzeneacetic acid, 2,5-dihydroxy-2-Methyl -6,7-dihydro-5H-benzofuran-4-one Benzo[b]thiophene-2-ol	C ₉ H ₁₀ O ₂ C ₈ H ₆ OS	150.17 150.20
32	11.897	0.52	Benzeneacetic acid, 2,5-dihydroxy-Oxirane, (phenoxymethyl)-2-Methyl -6,7-dihydro-5H benzofuran-4-one	C ₉ H ₁₀ O ₂	150.17
33	11.975	0.51	Benzeneacetic acid, 2,5-dihydroxy-Oxirane (phenoxymethyl) -5,6,7,8-Tetrahydro-1,2,4 benzotriazine-3-amine	C ₇ H ₁₀ N ₄	150.18

34	12.053	0.70	Benzeneacetic acid,		
			2,5,dihydroxy-		
			5,6,7,8-Tetrahydro-1,2,4-benzotriazine-3-amine	C ₇ H ₁₀ N ₄	150.18
			Benzo[b]thiophene-2-ol	C ₈ H ₆ OS	150.20
35	12.230	0.71	Benzeneacetic acid,		
			2,5-dihydroxy-2,5-Methano-1H-inden-7(4H)-one, hexahydro		
			-Propanoic acid, phenyl ester	C ₁₀ H ₁₄ O	150.22
36	12.319	0.66	Benzeneacetic acid,		
			Tricyclo[4.2.2.0(1,5)]decan-4-one		
			Borolo[1,2-a]borine, octahydro-	C ₁₀ H ₁₆ O	152.24
				C ₈ H ₁₅ B	122.02
37	13.286	0.21	2(3H)-Naphthalenone, 4,4a,5,6,7,8-hexahydro		
			Borabicyclo[3.3.1]nonane, 9-[3(dimethylamino)propyl]-		
			4,5,6,6a-Tetrahydro-2(1H)-pentalenone	C ₁₀ H ₁₃ O	150.22
				C ₁₃ H ₂₆ BN	207.2

38	13.486	0.23	2-methyl-5-(4-morpholinyl)-		
			Benzoic acid, 2,4-dihydroxy-6-methyl-, methyl ester	$C_9H_{10}O_4$	182.17
			Oxirane, (phenoxyethyl)-	$C_9H_{10}O$	150.18
39	13.663	0.32	Cyclohexa-2,5-diene-1,4-dione	$C_6H_2O_2$	106.08
			1-Octen-3-yne	C_8H_{12}	108.18
			Oxirane, (phenoxyethyl)-	$C_9H_{10}O$	150.18
40	13.841	0.60	Pentadecanoic acid	$C_{15}H_{30}O_2$	242.02
			Ethyl n-butyl disulphide	$C_6H_{14}S_2$	150.3
			Bicyclo[3.2.2]nona-2,6-dien-5-ol-4-one	$C_9H_{10}O_2$	150.17
41	14.663	0.78	3-Myristinoyl-glycinamide	$C_{16}H_{28}N_2O_2$	280.41
			3-pyridinamine, 2-[(4-methyl-4H-1, 2,4-triazol-3-yl)thio]-	$C_8H_9N_5S$	207.25
			Ethyl n-butyl disulphide	$C_6H_{14}S_2$	150.3

42	14.852	7.97	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	256.42
43	15.019	0.30	Pyrido[2,3-d]pyrimidine,	$C_7H_5N_3$	131.13
			[1,2,4]Triazolo[1,5-a]pyrimidine,	$C_5H_4N_4$	120.11
			3-ethylsulfanyl-5,7-dimethyl Dodecahydropyrido[1,2-b]isoquinolin-6-one	$C_{13}H_{21}NO$	207.31
44	15.319	0.20	Bicyclo[6.1.0]non-1-ene	C_9H_{14}	122.21
			[1,2,4]Triazolo[1,5-a]pyrimidine-6-carboxylic acid, dihydro-7-imino-, ethyl ester	$C_8H_9N_5O_2$	207.19
			Bicyclo[5.2.0]non-1-ene	C_9H_{14}	122.21
45	15.408	0.15	Tricyclo[4.2.1.1(2,5)]decane	$C_{10}H_{16}$	136.23
			1-Cyclopentenylphenylmethane	$C_{12}H_{14}$	158.24
			Pentacyclo[6.3.0.0(2,7).0(4,11).0(5,9)]undecan-3-one	$C_{11}H_{12}O$	160.22

46	15.674	1.94	Stigmasta-4,22-diene	$C_{29}H_{46}O$	280.41
			4-Myristynoyl-glycinamide	$C_{16}H_{28}N_2O_2$	
47	15.796	1.46	Stigmasta-4,22-diene	$C_{29}H_{46}O$	410.7
			Ergosta-4,7,22-trien-3.beta.-ol	$C_{28}H_{44}O$	396.6
			Stigmasta-5,22-dien-3-ol, acetate, 2(3.beta.)-	$C_{31}H_{50}O_2$	454.73
48	15.908	0.98	7-Chlorobicyclo[4.1.0]hept-3-ene	C_7H_9Cl	128.6
			4,7-Methano-1H-indene, octahydro-	$C_{10}H_{16}$	136.23
			Stigmasta-4,22-diene	$C_{29}H_{46}O$	410.7
49	15.974	1.21	Stigmasta-4,22-diene	$C_{29}H_{46}O$	410.7
			1,2,4-Oxadiazole, 3-(1,3-benzodioxol-5-yl)-5-[(4-iodo-1H-pyrazol-1-yl)methyl]- Stigmasta-5,22-dien-3-ol, acetate, (3.beta.)-	$C_{31}H_{50}O_2$	454.73
50	16.096	1.10	Ergost-4,7,22-trien-3.alpha.-ol	$C_{28}H_{42}O$	394.6
			Stigmasta-3,5-diene		

			1,2,4-Oxadiazole,	C ₂₉ H ₄₈	396.7
				C ₂ H ₂ N ₂ O	70.05
51	16.096	0.35	4-Allyl-5-furan-2-yl-2,4-dihydro-[1,2,4]triazole-3-thione 1,2,5-Oxadiazol-3-amine, 4-(3-methoxyphenoxy)- 4-Dehydroxy-N-(4,5-methylenedioxy-2 nitrobenzylidene)tyramine	C ₉ H ₉ N ₃ OS	
52	16.463	8.12	9,12-Octadecadienoic acid (Z,Z)- 9,12-Octadecadienoic acid (Z,Z)- 9,17-Octadecadienal, (Z)-	C ₁₈ H ₃₂ O C ₁₈ H ₃₂ O	280.45 264.45
53	16.719	1.19	2(1H)-Naphthalenone,octahydro-4a-methyl-7-(1-methylethyl)-, (4a. alpha, 7.beta,8a.beta.)- 5-Methyl-Z,Z-3,13-octadecadienol 6-4,7,7-Trimethylbicyclo[2.2.1]heptan-2-one O-allyl oxime	C ₁₄ H ₂₄ O C ₁₉ H ₃₆ O C ₁₃ H ₂₁ NO	208.18 280.5 207.31
54	18.207	0.21	1,2,5-Oxadiazol-3-amine, 4-(3-methoxyphenoxy)- 3,5Dimethylbenzaldehydethiocarbamoylhydrazone [1,2,4]Triazolol[1,5-a]pyrimidine-6-carboxylic acid, 4,7-dihydro-7-imino-, ethyl ester	C ₃ H ₃ N ₃ O	85.07
55	18.274	0.18	3,5-Dimethylbenzaldehydethiocarbamoylhydrazone Isolongifolan-8-ol	C ₁₀ H ₁₃ N ₃ S C ₁₅ H ₂₆ O	207.3 222.37

56	18.429	0.86	[1,2,4]Triazolo[1,5-a]pyrimidine-6-carboxylic acid, 4,7-dihydro-7-imino-, ethyl ester	$C_8H_9N_5O_2$	207.19
			3,5Dimethylbenzaldehydethiocarbamoylhydrazone	$C_{10}H_{13}N_3S$	207.3
57	18.652	1.18	Ethanone, 2-(2-benzothiazolylthio) -1-(3,5-dimethylpyrazolyl)-	$C_{14}H_{13}N_3OS_2$	303.4
58	19.918	0.34	1H-Indole-2-carboxylic acid, (4-ethoxyphenyl)-3-methyl-4-oxo-4,5,6,7-tetrahydro-, isopropyl ester Indole-2-one,		
			2,3-dihydro-N-hydroxy-4-methoxy-3,3-dimethyl 2-(n-Propyl)oxybenzylidene acetophenone		
59	19.074	0.37	3,5-Dimethylbenzaldehydethiocarbamoylhydrazone	$C_{10}H_{13}N_3S$	207.3
			Silicic acid, diethyl bis(trimethylsilyl) ester	$C_{10}H_{28}O_4SiO_3$	
			4-Allyl-5-furan-2-yl-2,4-dihydro-[1,2,4]triazole-3-thione	$C_9H_9N_3O_3$	207.25
60	19.585	0.15	1,2,5-Oxadiazol-3-amine,		
			4-(3-methoxyphenoxy)- 3,5Dimethylbenzaldehydethiocarbamoylhydrazone 2-Myristynoyl-glycinamide		

61	19.585	0.49	3,5Dimethylbenzaldehyde l thiocarbamoylhydrazone	$C_{10}H_{13}N_3S$	207.3
			[1,2,4]Triazolo[1,5-a]pyrimidine-6-carboxylic acid, 4,7-dihydro-7-imino-, ethyl ester	$C_8H_9N_5O_2$	207.19
62	19.825	1.34	7-Methyl-4-(2,6,6-trimethylcyclohex-1-enyl)but-2-en-1-ol	$C_{14}H_{24}O$	208.35
			5-Methyl-6-(5-methyl-2-thiazolin-2 ylamino)pyridine	$C_{10}H_{13}N_3S$	207.30
			Benzoic acid, 4-(1,3-dioxolan-2-yl)-, methyl ester	$C_{11}H_{12}O_4$	208.21
63	20.029	0.37	1H-Indole-2-carboxylic acid, (4-ethoxyphenyl)-3-methyl-4-oxo-4,5,6,7-tetrahydro-,	$C_{10}H_9NO_2$	175.18
64	20.229	0.88	8-Isopropyl-6a,7,10b-trimethyl-dodecahydro-benzo[f]chromene-7,8-dicarboxylic acid, dimethyl ester	$C_{23}H_{38}O_5$	394.6
			4-Dehydroxy-N-(4,5 methylenedioxy-2-nitrobenzylidene)tyramine	$C_{16}H_{14}H_2O_4$	298.29
65	20.374	0.45	1,2,5-Oxadiazol-3-amine,	$C_2H_3N_3O$	85.07
			2 Butenenitrile,	C_4H_5N	67.09

			2-chloro-3-(4-methoxyphenyl)- 3-Indole-2-one, 2,3-dihydro-N-hydroxy-methoxy 3,3-dimethyl	C ₁₁ H ₁₃ NO ₃	207.23
66	20.563	1.22	1,2,5-Oxadiazol-3-amine,	C ₂ H ₃ N ₃ O	85.07
67	20.829	3.23	Stigmastan-6,22-dien, 3,5-dedihydro-Stigmasterylosylate Cholesta-6,22,24-triene, 4,4-dimethyl-	C ₂₉ H ₄₈	396.7
68	21.107	0.70	1-Benzazirene-1-carboxylic acid 2,2,5a-trimethyl-1a-[3-oxo-1-buteny l] perhydro-, Indole-2-one,2,3-dihydro-N-hydroxy-4-methoxy-3,3-dimethyl 1H-Indole-2-carboxylic acid, (4-ethoxyphenyl)-3-methyl-4-oxo-4,5,6 ,7-tetrahydro-, isopropyl ester	C ₁₅ H ₂₃ NO ₃ C ₁₁ H ₁₃ NO ₃	265.35 207.23
69	21.562	2.39	Cholesta-3,5-diene	C ₂₇ H ₄₄	368.64
70	21.807	0.60	Pyridine-4-carboxylic acid, benzeneacetaldehyde, .alpha.-(methoxymethylene)-4-nitro Furan-2-carboxylic acid, [4-(4-methoxyphenyl)-tetrahydropyran-4-ylmethyl]amide	C ₅ H ₄ N C ₁₀ H ₉ NO ₄ C ₁₉ H ₁₈ N ₂ O ₃ S	123.11 207.12 354.4

DISCUSSION

The presence of various secondary class metabolites identified puts these results in line with earlier preliminary screening carried out on the leaf extract of *Alafia barteri* (Adefisan *et al.*, 2021), which revealed the presence of secondary metabolites compounds group of saponins, alkaloids and triterpenoids, flavonoids, anthraquinones, tannins, and cardiac glycosides, in which it was noted that they are defense chemical compounds of plants produced in the plant tissue.

In this study, 70 components were identified in the ethanol leaf extract of *Alafia barteri*. 2-Pyrodilone is an organic compound with pharmaceutical and cosmetic importance. It is also used in industrial settings as a polar solvent for different applications. Cyclotetrasiloxane, a type of silicone is used as an emollient, for cosmetic uses. Hexadecanoic acid, ethyl ester is a fatty acid ester which has been reported to have nematocidal, pesticide, lubricant, anti-androgenic, flavor, hemolytic 5-alpha reductase inhibitor, antioxidant and hypo-cholesterolemic properties (Komansilan *et al.*, 2012). Worthy of note is the presence of N-methylamphetamine which serves as a CNS stimulant (Gentry *et al.*, 2004), as well as piperazine which has been reported to have been antihelmintic. 9,12-Octadecadienoic acid, ethyl ester is a linoleic acid which has hypocholesterolemic, 5-alpha reductase inhibitor, antihistaminic, insectifuge, anti-eczemic, and anti-acne properties. (Aneesh *et al.*, 2013).

The important pharmacological activities of *A. barteri* ethanol leaf extracts may be attributed to the presence of these bioactive components.

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