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Research Article
Chemical Investigation and Therapeutic Significance of Essential Oils of Nigerian *Plumeria acuminata* Ait.

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Abstract

Background and Objective: *Plumeria acuminata* has a long history of use in traditional medicine which includes treatment of rheumatism, pruritic skin lesion, toothache, asthma, cracks on the feet sole crack, heart stroke, dysentery and diarrhoea. This study was designed to extract essential oils from the leaf, flower and root of *P. acuminata*, analyse the oils and evaluate their cytotoxicity and antimicrobial and antioxidant activities. **Materials and Methods:** Essential oils obtained from leaf, flower and root of *P. acuminata* by hydrodistillation were analyzed using the Gas Chromatography-Mass Spectrometry technique. The toxicity, antimicrobial and antioxidant activities of the oils were evaluated via brine shrimp lethality, agar well-diffusion and DPPH free radical-scavenging assays, respectively. **Results:** A total of 26, 85 and 12 constituents were identified in the leaf, flower and root oils corresponding to 70.0, 99.8 and 97.4% of the whole oils, respectively. The major components were β-eudesmol, 43.0% (root), nonanal, 20.0% (flower), palmitic acid, 27.6% (root) and linalool, 16.1% (leaf). The oils exhibited high toxicity with LC_{50} less than 100 ppm, indicating they are biologically active. They also showed potent antimicrobial activities with an inhibition zone range of 1.7±0.2-22.1±0.1 mm when compared with the activities of standard drugs: Gentamycin (8.0±0.0-12.5±0.1 mm) and ketoconazole (10.3±0.4-21.0±1.4 mm). However, low antioxidant activities were demonstrated by the oils (IC_{50} = 441-695 μg mL^{-1}) concerning the positive control, α-tocopherol (81.58 μg mL^{-1}) and butylated hydroxyanisole (45.11 μg mL^{-1}). **Conclusion:** The findings from this study suggest that *P. acuminata* essential oils are biologically active and could be natural sources of antibiotics.

Key words: *Plumeria acuminata*, essential oil, toxicity, antimicrobial, antioxidant

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Plants have continued to be sources of medicine to humans since time immemorial. The medicinal values of plants are related to their bioactive components known as phytochemicals. Previous works have reported essential oils as the most attractive among these phytochemicals because they contain multifunctional compounds which possess diverse biological activities[1]. As a result, essential oils have shown a variety of biological activities which include antibacterial, antifungal, antiseptic, anticancer, anti-inflammatory, antiviral, insecticidal, ovicidal, growth-inhibitory, repellence, antifeedant, spasmolytic, sedative, analgesic and anesthetic[2-4]. However, there remain large numbers of essential oil-bearing plants that are yet to be investigated for their chemical constituents and biological activities.

Plumeria acuminate Ait. (Family: Apocynaceae) is a medicinal plant native to South America, Central America, Mexico and the Caribbean. The plant is widely cultivated because of its fragrance and the flowers that beautify the environment. Nevertheless, the plant is also used in folk medicine for the treatment of rheumatism, printing skin lesions, toothache, asthma, cracks on the feet sole, heatstroke, constipation, fever, dysentery and diarrhea[2]. Early reports showed that the methanol extract of the plant exhibited significant antipyretic, anti-inflammatory, antinociceptive, antimicrobial, antioxidant and free radical scavenging activities[2]. In addition, antimicrobial activity and chemical compositions of the leaf and flower essential oils from the genus *Plumeria* have been reported[5-8]. Other phytochemicals besides essential oils have also been investigated by Gupta *et al.*[9] but there is a dearth of information on the biological activities of the essential oils from *P. acuminata*.

The present study aimed at evaluating the chemical constituents, toxicity, antimicrobial and antioxidant activities of essential oils extracted from the leaf, flower and root of Nigerian *P. acuminata* and also compare the chemical compositions with those results from previous studies. Toxicity, antioxidant and antimicrobial activities of these oils as well as the chemical composition of the root oil are been reported for the first time.

MATERIALS AND METHODS

Study area: This study was carried out from May, 2017 to September, 2018.

Plant preparation and extraction of essential oils: The plant was collected at Masaba Road, University of Ibadan (GSP Coordinates: 7°23'0.2"N/3°54'28.8"E) and authenticated at Forestry Research Institute of Nigeria where a voucher specimen (FHI 112495) was deposited. The leaves and roots were air-dried and pulverized while the flowers were used fresh due to their perishable nature. The 3 plant parts were subjected to hydrodistillation separately in an all-glass modified clevenger apparatus for 3 hrs according to the British Pharmacopeia specification as described by Alade *et al.*[10]. The isolated volatile oils were dried over anhydrous sodium sulphate. The oils were stored in sealed vials and kept in the refrigerator before analysis and bioassay.

Analysis of the essential oils: The essential oils were analyzed through Gas Chromatography-Mass Spectrometry (GC-MS) using a Shimadzu GC-MS-QP2010 equipped with a ZB-5 fused silica capillary column (Phenomenex, Torrance, California, USA) with a (5% phenyl)-polydimethylsiloxane stationary phase and a film thickness of 0.25 µm, operated in the electron-impact mode of 70 eV, scan rate (3.0 scan s^{-1}) and scan range (40-400 atomic mass units). The injector's initial temperature was 50°C increased at a rate of 2°C min^{-1} to 260°C. A 5% w/v solution of each sample in CH_2Cl_2 was prepared and 0.1 µL was injected with a splitting mode of 30:1 using helium as carrier gas with a column head pressure of 552 kPa and flow rate of 1.37 mL min^{-1}.

Identification of constituents was based on a comparison of their mass spectra with the literature and those of reference compounds stored in the home database library[10]. The relative percentage of each constituent was calculated by the integration of GC peak areas.

Toxicity test: The essential oils were subjected to a brine shrimp lethality assay in other to evaluate their toxicity. The assay was carried out using the method described by Hamidi *et al.*[11] with a slight adjustment. *Artemia salina* eggs (brine shrimp) were hatched into larvae after 48 hrs of soaking them in seawater under sunlight. Ten nauplii were added into three replicates of 10, 100 and 1000 ppm solution of essential oils dissolved in Dimethylsulphoxide (DMSO). After 24 hrs at room temperature, the numbers of survived and dead larvae were counted and the values were subjected to Probit regression analysis to evaluate their toxicity in terms of lethality concentration (i.e., LC_{50}), which is the concentration that kills 50% of the larvae[12].

Antimicrobial assay: The essential oils were tested against seven bacteria (*Escherichia coli*, *Staphylococcus aureus*, *Salmonella typhi*, *Leclercia adecarboxylata*, *Morganella morganii*, *Citrobacter freundii* and *Klebsiella pneumoniae*) and 3 fungi (*Aspergillus niger*, *Fusarium solani* and *Candida albicans*) using agar well diffusion method. All microbes were clinical isolates obtained from University College, Ibadan, Nigeria. Diluted overnight cultures (10^{-2} CFU mL^{-1}) of the selected microorganisms were inoculated each into a sterile agar inside the Petri dish. Bacteria and fungi strains were maintained on Mueller-Hinton agar and Sabouraud dextrose agar, respectively. Wells of uniform diameter were created in the seeded agar plates using an 8 mm cork borer. The wells were labelled according to the number of the essential oil samples (dissolved in DMSO) with their serial dilutions (1, 10 and 100 µg mL^{-1}). The oil samples were then introduced into each well accordingly and allowed to diffuse into the seeded agar for 1 hr before incubating at 37°C for 24 hrs for bacteria and at 25-32°C for 48 hrs for fungi after which the inhibition zones were observed and recorded. Each assay was carried out in triplicate and the results were reported as Mean±Standard deviation. Gentamycin (10 µg mL^{-1}) and ketoconazole (200 µg) were used as positive references, respectively for bacteria and fungi.

DPPH free radical-scavenging assay: The DPPH free radical-scavenging ability of the essential oils was studied to assess the antioxidant properties of the oils using the method described by Saleh *et al.*[13] with some modifications. In this assay, 1.5 mL of different concentrations of all the essential oil samples (5, 25 and 100 mg mL^{-1}) was separately mixed with 1.5 mL of 0.2 mM DPPH using methanol as solvent. These were incubated in the dark for 20 min at room temperature. The absorbance at 517 nm was recorded as $A_{(sample)}$ using CE 2021, 2000 series double beam UV-Vis spectrophotometer. The absorbance of the blank, $A_{(blank)}$ was also recorded using the same procedure but without essential oil. Each experiment was carried out in triplicate and the free radical-scavenging activities of the oils were calculated as percentage inhibition using the formula:

$$\text{Inhibition (\%)} = \frac{A_{(blank)} - A_{(sample)}}{A_{(blank)}} \times 100$$

The values of the percentage inhibitions were plotted against the observed concentrations using Microsoft excel to obtain the IC_{50} value which was the concentration required for 50% inhibition[14]. Activities of the oils were compared with control antioxidants butylated hydroxyanisole (BHA) and α-tocopherol.

RESULTS

Chemical compositions of the essential oils: The percentage yields of the leaf, flower and root essential oils of *P. acuminata* were 0.18, 0.13 and 0.08%, respectively. Table 1 shows the retention indices (RI) of the compounds present in the oils in order of their elution from a ZB-5 fused silica capillary column with their percentage composition (peak area). The percentage composition was obtained by the ratio of the individual area (TIC: Total ion count) of the component with the total TIC area of all the peaks and then multiply by 100. The major components in the leaf essential oil were linalool (16.1%), phytol (6.8%), (*E*)-β-ionone (6.1%) and (*E*)-hexadecatrienal (5.4%). Linalool was also an abundant component of the floral essential oil (12.6%), but the fatty aldehydes nonanal (20.5%) and eicosanal (10.6%) were also abundant. In contrast, the root essential oil was dominated by β-eudesmol (43.0%), palmitic acid (27.6%) and 11-hydroxy-(8*E*)-dodecenoic acid lactone (14.9%). The gas chromatograms of the essential oils are given in Fig. 1-3. The x-axis of the chromatograms represents the retention time (minutes) which is the time taken by each constituent to reach the mass spectrometer detector from the injector. The y-axis shows the relative abundance of the oils' constituents which quantifies the amount of ion produced relative to the total ion count of all peaks. The classes of compounds identified include monoterpenes, sesquiterpenes, diterpenes and apocarotenoids. In addition, alkanes, aldehydes, alcohol, ketones, amide, ester, fatty acid and aromatic compounds of non-terpene derivatives were present. The compounds identified are shown in Table 1 and Fig. 1-3.

Toxicity against *Artemia salina*: The ability of the essential oils to kill *Artemia salina* larvae in many cases translates to toxicity and is correlated to the acute oral toxicity assay in mice[12]. The lethality of the volatile oils was found to be concentration-dependent because maximum mortalities (100%) were observed at 1000 ppm and it decreased as the concentration decreased. The toxicity of the oils against brine shrimps in terms of Lethal Concentration (LC_{50}) obtained from the mortality rate of the shrimps is presented in Table 2. Log-Probit analysis of the brine shrimp lethality data revealed median Lethal Concentrations (LC_{50}) of 6.90, 23.20 and 61.22 ppm, respectively, for the leaf, floral and root essential

Table 1: Chemical constituents of Nigerian *Plumeria acuminata* essential oils

		Percentage of composition		
RI	Constituents	Leaf	Flower	Root
800	Octane	-	4.4	-
851	2*E*-Hexanal	1.0	-	-
874	Ethylbenzene	2.0	-	-
879	*o*-Xylene	2.2	-	-
901	Heptanal	-	0.5	-
905	4-Methyl-3-hexanol	3.0	-	-
928	α-Pinene	1.8	0.2	-
959	Benzaldehyde	-	0.8	-
961	1-(1-Methyl-cyclopentyl)-ethanone	4.8	-	-
976	1-Octen-3-ol	-	0.5	-
986	2-Pentylfuran	2.1	-	-
1001	Octanal	-	0.7	-
1026	Limonene	2.0	0.3	-
1033	(*Z*)-β-Ocimene	-	0.1	-
1041	Benzeneacetaldehyde	-	0.9	-
1043	(*E*)-β-Ocimene	-	2.3	-
1067	1-Octanol	-	0.6	-
1084	(*E*)-Linalool oxide	-	0.7	-
1088	2-Nonanone	-	0.2	-
1092	Methyl benzoate	-	1.0	-
1098	Linalool	16.1	12.6	-
1107	*n*-Nonanal	0.9	20.5	-
1119	(*Z*)-*p*-Menth-2-en-1-ol	1.1	-	-
1135	Non-3-en-2-one	-	2.7	-
1138	(*E*)-*p*-Menth-2-en-1-ol	0.7	-	-
1157	(2*E*)-Nonenal	-	0.3	-
1183	α-Terpineol	0.8	1.2	-
1189	Methyl salicylate	-	0.6	-
1204	Decanal	-	0.1	-
1218	β-Cyclocitral	1.1	-	-
1221	Nerol	-	0.2	-
1223	Citronellol	-	0,2	-
1247	Geraniol	-	0.9	-
1260	(*2E*)-Decenal	-	0.1	-
1262	Nonanoic acid	-	0.2	-
1265	Geranial	-	0.1	-
1285	Hydroxycitronellal	-	0.2	-
1289	2-Undecanone	-	0.1	-
1295	1-Nitro-2-phenyl ethane	-	0.3	-
1316	(2*E*,4*E*)-Decadienal	-	0.1	-
1343	α-Terpinyl acetate	-	tr	-
1361	(2*E*)-Undecenal	-	tr	-
1372	α-Copaene	1.7	0.1	-
1375	(*E*)-β-Damascenone	-	0.2	-
1389	β-Elemene	1.8	-	-
1414	β-Caryophyllene	1.0	0.1	-
1418	(*E*)-α-Ionone	-	tr	-
1433	(*E*)-α-Bergamotene	0.7	0.1	-
1443	11-Hydroxy-(8*E*)-dodecenoic acid lactone	-	-	14.9
1444	Geranyl acetone	-	0.1	-
1451	α-Humulene	-	0.1	-
1456	(*E*)-β-Farnesene	0.6	0.1	-
1457	4,6,8,10-Tetramethyltridecane	-	0.1	-
1461	γ-Decalactone	-	tr	-
1479	Germacrene D	0.6	-	-
1483	(*E*)-β-Ionone	6.1	0.1	-
1499	Viridiflorene	2.5	-	-
1500	(*E,E*)-α-Farnesene	-	0.3	-

Table : Continue

RI	Constituents	Percentage of composition		
		Leaf	Flower	Root
1504	β-Bisabolene	-	tr	-
1508	Tridecanal	-	0.1	-
1514	δ-Cadinene	-	0.1	-
1557	(*E*)-Nerolidol	-	4.3	-
1569	Dendrolasin	-	0.1	-
1574	Caryophyllene oxide	-	0.2	0.7
1605	Cedrol	-	0.5	-
1609	Tetradecanal	-	0.1	-
1628	γ-Eudesmol	-	-	1.7
1639	Hinesol	-	-	1.0
1647	β-Eudesmol	-	-	43.0
1651	α-Eudesmol	-	-	1.2
1673	1-Tetradecanol	-	0.2	-
1676	Mustakone	1.1	-	-
1681	*epi*-α-Bisabolol	-	0.2	-
1683	α-Bisabolol	-	0.2	-
1700	Heptadecane	-	0.4	-
1711	Pentadecanal	-	1.8	0.3
1754	Myristic acid	-	0.4	-
1762	Benzyl benzoate	-	0.2	-
1800	Octadecane	-	0.2	-
1832	Neophytadiene	-	0.1	-
1837	Phytone	-	0.7	-
1850	2-Methylbenzyl benzoate	-	0.2	-
1881	(*Z*)-Hexadecatrienal	-	0.6	-
1888	(*E*)-Hexadecatrienal	5.4	0.4	-
1900	Nonadecane	-	8.6	-
1915	Heptadecenal	-	0.3	-
1959	Palmitic acid	1.1	2.7	27.6
2000	Eicosane	-	0.2	-
2016	Octadecanal	-	0.5	-
2079	1-Octadecanol	-	0.4	-
2100	Heneicosane	-	2.4	-
2102	Phytol	6.8	0.5	-
2118	Nonadecanal	-	1.3	-
2123	Linoleic acid	-	0.2	-
2129	Oleic acid	-	-	3.7
2221	Eicosanal	-	10.6	-
2283	1-Eicosanol	-	1.0	-
2300	Tricosane	-	0.5	-
2322	Heneicosanal	-	0.2	-
2349	(9*Z*)-Octadecenamide	-	0.3	1.3
2424	Docosanal	-	0.8	-
2484	1-Docosanol	-	0.3	0.6
2500	Pentacosane	-	0.5	1.4
2700	Heptacosane	-	2.6	-
	Monoterpene hydrocarbons	3.8	2.9	-
	Oxygenated monoterpenes	18.7	16.2	-
	Sesquiterpenes hydrocarbons	8.9	1.1	-
	Oxygenated sesquiterpenes	1.1	5.7	47.6
	Apocarotenoids	7.2	0.1	-
	Diterpenes	6.8	1.3	-
	Non terpenes	23.5	72.7	49.8
	Percentage identified	70.0	99.8	97.3
	Number identified	25	85.0	12

tr: Trace and RI: Retention indices from the analyses value

oils. Values of LC_{50} less than 100 ppm are known to be toxic, values between 100-500 ppm are moderately toxic and 500-1000 ppm are less toxic while above 1000 ppm are non-toxic. High toxicity against brine shrimps also shows that the tested samples contain biologically active constituents.

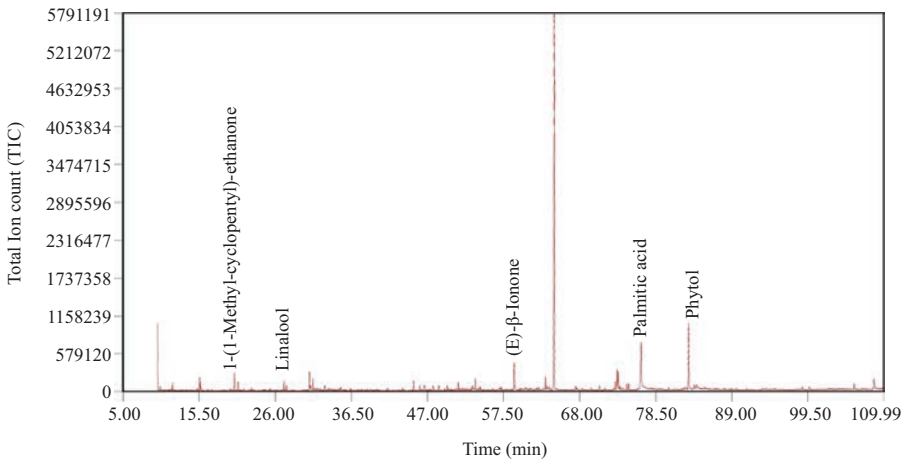

Fig. 1: Gas chromatogram of *Plumeria acuminata* leaf essential oil

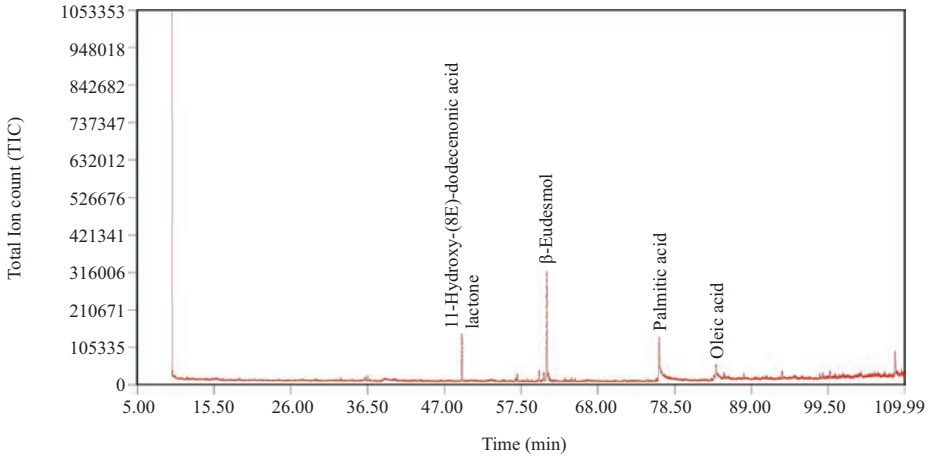

Fig. 2: Gas chromatogram of *Plumeria acuminata* root essential oil

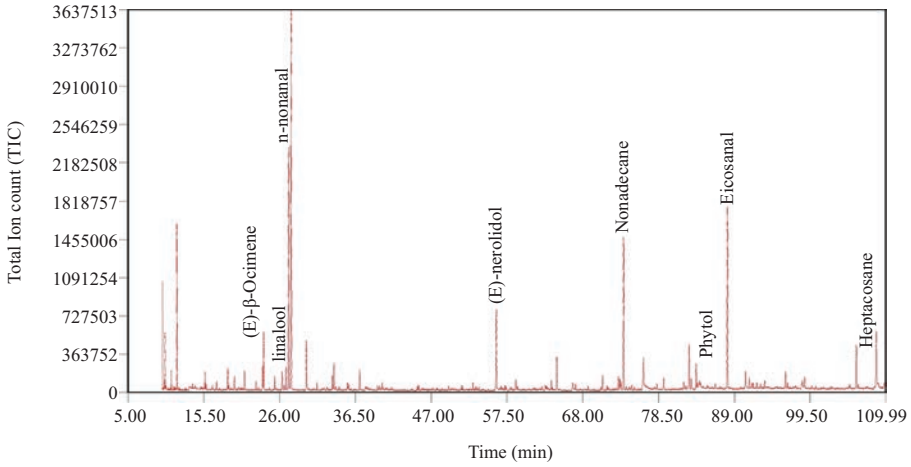

Fig. 3: Gas chromatogram of *Plumeria acuminata* flower essential oil

Table 2: Activity of *Plumeria acuminata* against brine shrimps

Plant/part used	LC_{50} (ppm)	LC limit (ppm)	UC limit (ppm)	CL
Leaves	6.9070	1.1274	14.6368	0.2970
Flowers	23.1982	10.5660	41.7706	0.1514
Root	61.2245	33.8675	104.6401	0.1106

LC_{50}: Concentration with 50% mortality, LC: Lower confidence, UC: Upper confidence and CL: Confidence limit ppm-part per million

Table 3: Antibacterial activity of *Plumeria acuminata* essential oils

		Inhibition zones diameter (mm)						
Sample	Concentration (μL mL^{-1})	*E. coli*	*S. typhi*	*S. aureus*	*L. adecarb*	*M. morganii*	*C. freundii*	*K. pneumoniae*
Leaf essential oil	1	1.7±0.2	na	3.9±0.1	3.9±0.1	6.4±0.1	2.3±0.1	2.0±0.0
	10	4.0±0.1	2.1±0.1	6.0±0.0	6.1±0.1	8.2±0.1	7.9±0.1	4.0±0.1
	100	6.1±0.1	4.1±0.1	8.1±0.1	10.1±0.1	12.0±0.1	12.0±0.0	6.1±0.1
Root essential oil	1	2.3±0.1	na	1.6±0.1	1.9±0.1	3.9±0.1	na	na
	10	6.4±0.1	na	3.9±0.0	3.9±0.1	6.0±0.1	1.9±0.1	na
	100	12.1±0.1	na	5.9±0.1	6.0±0.1	8.1±0.1	4.1±0.1	na
Flower essential oil	1	na	na	na	1.9±0.1	3.8±0.0	3.9±0.0	4.0±0.0
	10	1.8±0.1	1.9±0.0	1.5±0.1	3.8±0.1	5.9±0.1	6.0±0.1	6.0±0.0
	100	4.0±0.0	4.1±0.1	4.0±0.0	5.9±0.1	7.9±0.1	8.1±0.1	8.1±0.1
Gentamycin (10)	9.0±1.4	12.5±0.1	11.5±0.1	8.0±0.0	na	na	na	na
DMSO	na	na	na	na	na	na	na	na

na: No activity, DMSO: Dimethylsulfoxide and Inhibition zones reported as Mean±Standard deviation

Table 4: Antifungal activity of *Plumeria acuminata* essential oils

		Inhibition zones diameter (mm)		
Sample	Concentration (μL mL^{-1})	*C. albicans*	*A. niger*	*F. solani*
Leaf essential oil	1	15.6±0.1	na	na
	10	18.1±0.1	na	4.1±0.1
	100	22.1±0.1	na	7.9±0.1
Root essential oil	1	na	na	na
	10	na	na	1.8±0.1
	100	na	na	4.0±0.1
Flower essential oil	1	8.0±0.1	na	na
	10	10.1±0.1	na	2.1±0.1
	100	12.0±0.0	na	6.0±0.1
Ketoconazole (200 μg)		21.0±1.4	10.5±0.1	10.3±0.4
DMSO		na	na	na

na: No activity, DMSO: Dimethylsulfoxide and Inhibition zones reported as Mean±Standard deviation

Table 5: Free-radical scavenging activity of *Plumeria acuminata* essential oils

Tested samples	Leaf oil	Flower oil	Root oil	α-Tocopherol	BHA
IC_{50} (μg mL^{-1})	695.00	441.09	581.58	81.58	45.11

Antimicrobial activity: The account of the antibacterial and antifungal activities of both essential oils, standard drugs and the solvent (DMSO) used are given in Table 3 and 4, respectively. Their inhibitory activities were reported as inhibition zone diameter. The essential oils exhibited concentration-dependent activity while the solvent do not affect the tested microbes. The antibacterial activity of the oils at 100 μL mL^{-1} against *E. coli*, *S. typhi*, *S. aureus* and *L. adecarboxylata* was lower than that of gentamycin except *M. morganii*, *C. freundii* and *K. pneumoniae* which were resistant to the reference compounds but susceptible to the oils. Only the leaf essential oils demonstrated better antifungal effects than ketoconazole at 100 μL mL^{-1}.

Free-radical scavenging activity: The free radical-scavenging activity of the leaves, flowers and root essential oils of *P. acuminata* compared with the reference compounds α-tocopherol and butylated hydroxyanisole is given in Table 5. The free-radical scavenging assay measures the ability of the essential oils to donate proton or electron to free radicals and this is characterized by IC_{50}. The lower the IC_{50} the higher the scavenging activity and the higher the antioxidant property of the samples. The median Inhibitor Concentrations (IC_{50}) for DPPH radical-scavenging activities were 695, 441 and 582 μg mL^{-1}, respectively, for the leaf, floral and root essential oils, respectively and were much less active than the positive controls α-tocopherol (IC_{50} = 81.6 μg mL^{-1}) or BHA (IC_{50} = 45.1 μg mL^{-1}).

DISCUSSION

A total of 26 components were identified in the leaf oil, 85 in the flower and 12 in the root essential oil representing 70.0, 99.8 and 97.3% of the total oil content, respectively. The leaf oil consisted more of terpenes (46.5%) than the non-terpenes (23.5%). The major constituents were linalool (16.1%), (*E*)-β-ionone (6.1%) and phytol (6.8%). The flower essential oil on the other hand had more non-terpene (72.7%) than terpenes (26.5%) components. The main components were nonanal (20.5%), eicosanal (10.6%), linalool (12.6%) and nonadecane (8.6%). The root essential oil, like the flower oil, had a good proportion of terpene, principally oxygenated sesquiterpenes and an appreciable amount of fatty acid. The most abundant components in the root volatile oil were β-eudesmol (43.0%), palmitic acid (27.6%) and 11-hydroxy-(8*E*)-dodecenoic acid lactone (14.9%). Palmitic acid is a useful ingredient in food, cosmetics, soaps and industrial mould release agents' production[15]. Most of the terpenes and non-terpene compounds identified as major or minor constituents in the *P. acuminata* oils are used in perfumery. Examples are nonanal, linalool, geraniol and citronellol.

In the previous study from Nigeria, the chemotype detected in appreciable quantity were linalool (13.2%) and nonanal (9.6%) in the leaf oil, limonene (9.1%), caryophyllene oxide (7.9%) and (*E*, *E*)-α-farnesene (6.6%) in the flower oil[6]. Most of these compounds were present in small quantities in the present study.

Information from other geographical locations on the essential oils from *Plumeria* species was a little different from the chemotype reported from Nigeria. Some of the main components reported were absent and those present were in small quantity[5,16,17]. Phenylethyl benzoate, methyl phenylacetate, benzyl salicylate, and benzyl benzoate found as prominent constituents in Egyptian, Malaysian and Indian flower oils were not detected in our study. Factors like geographical area, maturity, seasonal variation and extraction procedure might have contributed to the variation in the chemical profiles of these oils.

The oils were potent against the brine shrimps with LC_{50} less than 100 ppm[18]. The leaf oil was most toxic (LC_{50} = 6.9070 ppm) followed by the flower (23.1982 ppm) and then the root oil (61.2245 ppm). The toxicity of the leaf oil could likely be due to the synergistic effects of some of its constituents, which likely are absent in other oil samples. The leaf volatile oil may possess antitumor activity. Negative control DMSO does not affect the brine shrimps.

All the essential oils demonstrated good antimicrobial activities with the leaf oil displaying the highest activity. Comparing the antibacterial activities of the oils with the standard drug, gentamycin showed no activity against *M. morganii*, *C. freundii* and *K. pneumoniae* but the microbes were inhibited by all the oil samples except for the root oil which showed no activity against *K. pneumoniae*. In addition, *S. typhi* was also resistant to only the root oil. The leaf essential oil also showed a higher inhibition zone (22.1±0.1 mm) at 100 μg mL^{-1} against *C. albicans* when compared to that of ketoconazole (21.0±0.0 mm) at 200 μg. Only *F. solani* was susceptible to the root oil. Extracts of the plant have been previously reported as good antimicrobial agents[2,7].

Linalool, β-eudesmol, phytol and palmitic acid which were major constituents in the oils and have shown good antimicrobial activities from the previous works[19-21], might have improved the antimicrobial potentials exhibited by the oils. Synergistic effects of minor constituents like caryophyllene oxide, limonene and α-terpineol can likewise contribute to the oils' activities.

The essential oils exhibited lower activity with IC_{50} values higher than that of reference compounds. The higher the IC_{50} values the lower the scavenging activity. Although linalool, α-terpineol, geraniol, thymol, limonene, α-pinene, phytol and caryophyllene, which have a good record of free radical-scavenging activities in the literature[22] were found in appreciable quantity in the oils, it is expected of the oils to have high ability to scavenge free-radicals. Therefore, antagonistic effects of other constituents could likely have contributed to the low activity displayed by the essential oils[23]. Crude extracts from the plant demonstrated good free-radical scavenging activity[2,7].

CONCLUSION

The essential oils hydrodistilled from the leaf, flower and root of *Plumeria acuminata* showed commendable antimicrobial activities and toxicity against brine shrimp. The results from this study suggest that the essential oils of *P. acuminata*, especially the leaf oil, might be an alternative source of antimicrobial and antitumor agents. However, further investigations to determine the minimum inhibitory concentrations and the toxicity of the oils against human cell lines will be carried out.

SIGNIFICANCE STATEMENT

This study discovered new antibiotics of plant origin that could be beneficial to the pharmaceutical and cosmetic industries.

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