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Antibacterial and phytochemicals status of *Detarium microcarpum* (guill and perr) stem bark

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ABSTRACT: The study aimed to provide confirmatory status on traditional usage of *Detarium microcapum* against urinary and intestinal infections and serve as an alternative in fighting against multi-drug resistance. Hence, the determination of chemical constituents of stem bark using GC-MS because of its sensitivity and high resolving power in identifying compounds, and test the antibacterial potential of methanol crude extract on some bacteria associated with the diseases claimed to be treated by traditional healers. Bacterial isolates were collected, and zone of inhibition diameter (ZID), minimum inhibition concentration (MIC), minimum bactericidal concentration (MBC) and minimum bacteriostatic concentration (MBS) were conducted to determine antibacterial activity. At the same time, gas chromatography coupled with mass spectrometry (GC-MS) was used to identify the phytoconstituents. The mean ZID (mm) ranged from 17.3±21 to 20.0±35 mm and showed a statistical significance difference at P< 0.05 for *S. aureus*, *E. coli*, and *K. pneumonia*. The MIC ranged from 11.67±7.6 to 4.17±1.4 mg/L, MBS 16.67±5.8 to 5.00±00, MBC 20.0±00 to 5.0±00, in order of increasing concentrations. Inspection of the MBC/MIC ratio indicated a bactericidal effect across isolates. From the GC-MS analysis, twenty-six phytochemical constituents were identified, primarily fatty acids or their esters. Chemical constituents' from *D. microcapum* methanol crude extract were identified. The antibacterial activity of some compounds directly correlates with the bioactivity of the same isolated and tested elsewhere, from a different source, thus its potential and therefore supporting the claim for its traditional usage. The research recommends in-depth research toward identifying specific active compounds for public health solutions.

1. INTRODUCTION

Chemo-therapeutants of non-phytotoxic properties, biodegradable and systemic application are mostly found from plant sources (Chamanlal & Verma, 2006). Such knowledge of these constituents is essential in searching for novel therapeutic agents and the authentification of traditional medicinal claims (Sriram et al., 2011). The increasing prevalence of resistance to standard antibiotic therapy among common pathogens is quite challenging, which is rapidly becoming a major health problem throughout the world (Khan et al., 2006). Infections caused by *S. aureus*, particularly multidrug-resistant strains and coagulase negative *Staphylococcus, Enterococcus*, and *Pneumococci*, were previously challenging to treat (Gale et al., 1980).

S. aureus, a leading cause of skin and soft tissue infections. *S. aureus* causes various illnesses, including boils, bacterial meningitis, and wound infections, and is frequently involved in nosocomial infections. It is known for its multi-drug resistance, particularly the presence of the mecA gene, which allows it to develop resistance to most -lactam medicines, including methicillin, and is hence known as methicillin-resistant *S. aureus* (MRSA) (Hiramatsu et al., 2014). *E. coli* is a typical flora of the human digestive system, but it may become pathogenic when ingested orally, causing diseases ranging from urinary tract infections to gastrointestinal tract infections, bacteremia, and newborn meningitis. *K. pneumonia* is a gram-negative bacilli, a facultative anaerobe that ferments lactose and is widely found in the colon. It has also been linked to skin and soft-tissue infections.*P. aeruginosa* is implicated in nosocomial infections, respiratory tract infection (RTI), UTI, GIT, dermatitis, soft tissue infections and infection in patients with cancer, HIV-AIDS and severe burn. Responses to treatment are compromised by inherent multi-drug resistance acquired by the bacteria. It is resistant to β-lactam and aminoglycoside and negatively induces the expression of outer membrane proteins, up-regulation of efflux pumps and can

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cause mutation in topoisomerases (Matar, 2018). *S. typhi* is also a gram-negative bacilli, a facultative anaerobe that infects birds, animals and humans. It is a causative agent of typhoid fever. *Salmonella enterica* serotype Typhi species is very diverse. The disease burden caused by *S. typhi* was estimated by WHO in 2000 to be approximately 22 million illnesses and 220,000 deaths worldwide (Crump et al., 2004).

These have created the need to discover new compounds endowed with antibacterial property, possibly, acting through mechanisms of action that are distinct from those of the well-known classes of antibacterial agents to which many clinically relevant pathogens are now resistant (Abhishek et al., 2010).

Gas chromatography's combination of speed, sensitivity, and high resolving power makes it an excellent technique for separating complex samples. Furthermore, it is simple to connect to spectrometry methods such as mass spectrometry (MS) to identify unknown compounds directly. Gas chromatography can quantitatively determine chemical compounds present at very low concentrations by using the flame ionization and electron capture detectors (both of which have very high sensitivities). It provides both universal and specific information on individual components in a sample and information on the identity of those components. The use of GC-MS is based on the realization that the numerous compounds in a plant can be grouped into only a few classes (alkaloids, flavonoids, phenolics, terpenoids and tannins of the classification system) and that each member of these classes can be characterized by a limited number of ions in the mass spectra. It proves to be a valuable method for the analysis of polar and non-polar compounds and volatile essential oil, fatty acids in the areas of food, beverage, flavor, and fragrance analysis, forensic and criminal cases, pesticides detection, in medical and pharmaceutical applications as well (Jayapriya & Gricilda, 2015; Sermakkani & Thangapandian, 2012).

Hence, thorough validations would provide alternative remedies for easily accessible and affordable to local individuals. It is against this background. The research wishes to determine the antimicrobial activity of *Detarium microcarpum* of stem bark and identify compounds that could be responsible for antimicrobial activity.

The genus Detarium (Fabaceae, subfamily Caesalpiniaceae) of the Detarieae tribe has initially been found in Africa, with three species, namely *D. microcarpum* Guill. (Perr), D. senegalense J. F. Gmelin and D. macrocarpum Harms (Peter et al., 2012). The ecological distribution of the species is different but morphologically looks similar (Akah et al., 2012). In various parts of Nigeria, the roots, bark, leaves and seeds of *D. microcarpum* are used to treat various diseases (P. Abreu & Relva, 2002; Burkill, 1995). Infusion or decoction of plant parts is used to treat urinary tract infections, haemorrhoids, malaria, leprosy, arthritis, digestive problems, pain relief, bronchitis and pneumonia (P. Abreu & Relva, 2002). A decoction of the leaves is used as a postpartum and hemostatic drug in some parts of the country and to treat seizures in some situations (Okwu & Uchegbu, 2009). In Kashere, the bark, leaves and roots are prepared to treat ailments such as skin infections, diarrhea and menstrual pain, rheumatism, urogenital infections and haemorrhoids. Considering the claims made by herbalists, the methanol crude extract of *D. microcarpum* was tested on isolates of bacteria associated with the claimed disease. This is to determine a single compound or group of plant compounds, using the GC-MS method to its advantage in identifying specific phytochemicals associated with current traditional claims and use as herbal medicines.

2. MATERIALS AND METHODS

2.1. Collection and Extraction

Two hundred and fifty grams of *D. microcarpum* stem bark were collected from Kashere and its environs, Gombe. The sample collected (voucher number FUKH 056) was identified by Professor A. M. Gani of the Department of Biological Sciences, Federal University of Kashere. The sample was air-dried at room temperature of 25 ^{o}C for ten days, and 150 g was blended into powder in a kitchen blender (Murugan et al., 2020). 100 g of the powder was flooded with 1 L of 95% ethanol for two weeks at room temperature. The filtrate after that was concentrated using a rotary evaporator (Yusha'u et al., 2014).

2.2. Test Organisms

E. coli, S. aureus, *S. typhi* and *K. pneumonia*, *P. aeruginosa* were obtained from the stock culture of the Department of Microbiology Laboratory, Bayero University Kano Nigeria and confirmed using cultural and biochemical characterization and maintained in a nutrient broth (NB) medium in a refrigerator. The inoculation density was standardized using a corresponding cell concentration of 1.0×10^{8} CFU/mL equivalent to 5 % McFarland standard (Ramalivhana et al., 2014).

2.3. Antibacterial Assay

After the two-fold serial dilution method, broth dilution and disk diffusion techniques were used. Kirby-Bauer test (agar disk diffusion) was employed to give quantitative insight into the level of inhibition exhibited by the plant extract on each isolate. Six millimeters in diameter discs were primed using No. 1 Whatman Filter Paper. Each disc was sterilized in a hot air oven at 160 ^{o}C for 2 hrs after which it was impregnated with 0.2 mL of respective extracts (320, 160, 80, 40, 20, 10, 5, 2.5, 1.3, and 0.63 w/v) concentrations, using dimethyl sulfoxide (DMSO) as solvent (Delnavazi et al., 2014). MH agar plates seeded with test organisms were inoculated with impregnated discs of the ten varying concentrations and incubated at 37 °C for 18 h; then, one inhibition (ZI) was observed and recorded. Zones were compared with that of some standard antibiotics, as sensitive (S) intermediate (I) and resistant (R) as follows; Ampicillin 10 μg (S = $\geq$17, I = 14 – 16, R= $\leq$13) and Cotrimoxazole 1.25/23.75 μg (S= $\geq$16, I= 11-15, R= $\leq$10); for *S. aureus*, *K. pneumonia*, *E. coli*, *S. typhi* and Gentamicin 10 μg (S= $\geq$15, I= 13-14, R= $\leq$12) for *P. aeruginosa* following CLSI (2011) recommendation.

2.4. Minimum inhibitory concentration (MIC)

One millilitre of prepared concentrations of the extract was added to a 9 mL broth of MH after which1 mL of the test organism (standardized) was also added. Inoculated samples were then incubated at 37 °C for 18- 24 h, and observed visual increase in turbidity of the medium in the test tubes. The visual observation was made by comparing the inoculated and uninoculated tubes (Andrews, 2001).

2.5. Minimum Bactericidal/Bactriostatic Concentrations (MBC/MBS)

To determine MBC, the isolates in the medium, identified as MIC, were sub-cultured on fresh NA (nutrient agar) plates and incubated for 18-24 h at 37 ^{O}C for possible bacterial growth. A 99.9 % reduction in the colony count from the initial inoculum was recorded as minimum bactericidal concentration, while samples demonstrating growth were considered minimum bacteriostatic concentration (Saleem et al., 2015).

2.6. Preparation (GC-MS) and Identification of Components

The sample was extracted with methanol and analyzed using GC-MS (GC-MS-QP2010 Plus Shimadzu, Japan). The data were obtained on an Elite-1(100% Dimethylpolysiloxane) column (30 0.25 mm 1 μmdf). Helium (99.999 %) was used as the carrier gas with a 1 mL/min flow rate in the split mode (10:1). An aliquot of 2 μl of the sample's methanol solution was injected into the column at 250 °C through the injector. The linear velocity of 46.3 cm/sec, total 6.2mL/min and purge flow of 3.0 mL/min were set. GC oven temperature started at 80.0 °C and held for 1 min and raised to 200 °C at the rate of 10 °C/min. With 4 min holding, the temperature was allowed up to 280 °C withholding 5 min. Ion source temperature 230 °C, interface 250 °C a solvent Cut Time of 2.50 min. The mass spectrum of compounds in samples was obtained by electron ionization at 70 eV, and the detector was operated in ACQ scan mode with a start of 40 mz and end at 600 mz. A scan start time of 3.00 s and end time of 28.00 min with a speed of 1250 was maintained (Devender & Ramakrishna, 2017).

2.7. Identification of components

The molecular structure, molecular mass and estimated fragment were used to identify the substances. The National Institute of Standards and Technology's database was used to interpret the mass spectrum GC-MS (NIST05s.LIB). Name, molecular weight, and structure of the phytocompounds determined with NIST library Version 2005.

2.8. Statistical analysis

Data were analyzed with IBM SPSS statistic 23; means were expressed in standard deviation (SD) of three replicates; t-test and analysis of variance (ANOVA) have been used, followed by Tukey's multiple comparisons tests; differences were considered significant ($p< 0.05$).

3. RESULTS

3.1. Z ones of inhibition

Antibacterial action of *D. microcarpum* stem bark was measured against S. aureus, *P. aeruginosa*, *E. coli*, *S. typhi,* and *K. pneumonia* and compared to standard Ampicillin 10 g/disc, Cotrimoxazole 1.25/23.75 g/disc, and Gentamicin 10 g/disc following CLSI (2011) recommendations. The concentrations measured ranged from 0.63 mg/Disc to 320 mg/Disc. The mean active concentration of the extract ranged between $\geq$ 106.7 $\pm$ 46.2 mg/disc to 293.3 $\pm$ 115.5 mg/disc among the bacteria tested. Furthermore, the actual ZID values were 20.0 $\pm$ 00 mm for *E. coli*, *P. aeruginosa* 20.0 $\pm$ 3.5 mm, and *K. pneumoniae* had 17.3 $\pm$ 6.1 mm. Both were statistically significant. The mean ZID of *S. typhi* was 17.3 $\pm$ 3.1 mm, and S. aureus 17.3 $\pm$ 2.1 mm showed a significant statistical difference($p< 0.05$), as shown in Table 1.

There was a statistical difference in the active mean concentration between different bacteria. For example, the active mean concentration difference between *E. coli* and *S. aureus* was ($p= 0.0002$), *K. pneumonia* and *S. aureus* was ($p = 0.0002$), *S. typhi* and S. aureus were ($p= 0.0014$), *P. aeruginosa* and E. coli ($p= 0.0014$), *P. aeruginosa* and *K. pneumonia* was ($p= 0.0014$) and *S. typhi* and *P. aeruginosa* ($p= 0.0109$).

Table 1
Effect of *D. microcarpum* methanol crude extract on selected bacterial isolates.

Bacteria	Concentration (mg/Disc)	Sig ($p < 0.05$)	ZID (mm)	Sig ($p < 0.05$)
S. aureus	106.7 $\pm$ 46.2	0.057	17.7 $\pm$ 2.1	0.005
E. coli	293.3 $\pm$ 115.5	0.048	20.0 $\pm$ 00	0
K. pnuemoniae	293.3 $\pm$ 115.5	0.048	17.3 $\pm$ 6.1	0.039
P. aeruginosa	133.3 $\pm$ 46.2	0.038	20.0 $\pm$ 3.5	0.010
S. typhi	266.6 $\pm$ 161.7	0.104	17.3 $\pm$ 3.1	0 .010

ZID = Zones of inhibition diameter; Values are given as Mean $\pm$ standard deviation

3.2. MIC, MBC and MBS

The mean MIC of the methanol crude extract of *D. microcarpum*, which can inhibit the visible growth, was determined. The MIC among the tested bacteria ranged from 4.2 $\pm$ 1.4 mg/mL to 11.7 $\pm$ 7.6 mg/mL. There was no statistical significance difference in the mean MIC observed for S. aureus (11.7 $\pm$ 7.6 mg/mL), *K. pneumonia* (6.7 $\pm$ 2.9 mg/mL), *P. aeruginosa* (6.7 $\pm$ 2.9 mg/mL) and *E. coli* (5.0 $\pm$ 00 mg/mL) at $p < 0.05$, except for *S. typhi* (4.2 $\pm$ 1.4 mg/mL) whose mean MIC showed significance difference ($p < 0.038$) as presented in Table 2. The results of the MBC ranged between 5 $\pm$ 00 to 20 $\pm$ 00 mg/L and showed no statistically significant difference across the tested bacteria. MBS was only significant on *S. aureus* 16.7 $\pm$ 5.8 mg/L ($p = 0.038$), and the results ranged from 5 $\pm$ 00 to 16.7 $\pm$ 5.8 mg/L. Overall assessment of MBC/MIC ratio among the tested bacteria revealed less than $\leq$ 1:4 ratio effect of the *D. microcarpum* stem bark extract. A ratio of 1:1.82 for *S. aureus*, 1:2 for *E. coli*, 1: 2.98, *K. pneumoniae* and 1:0.75 for

P. aeruginosa and 1:1.2 for *S. typhi* was observed as contained in the Table 2.

Table 2
Antibacterial activity of methanolic extract of *D. microcarpum* stem bark on some bacterial pathogens

Bacteria	MIC (mg/L)	Sig (P< 0.05)	MBC (mg/L)	Sig (P< 0.05)	MBS (mg/L)	Sig (P< 0.05)
S. aureus	11.67±7.6	0.118	20.0±00	0	16.67±5.8	0.038
E. coli	5.00±00	0	10.0±00	0	11.67±7.6	0.118
K. pneumoniae	6.67±2.9	0.057	20.0±00	0	10.00±00	0
P. aeruginosa	6.67±2.9	0.057	5.0±00	0	11.67±7.6	0.118
S. typhi	4.17±1.4	0.038	5.0±00	0	5.00±00	0

Values are given as Mean ± standard deviation

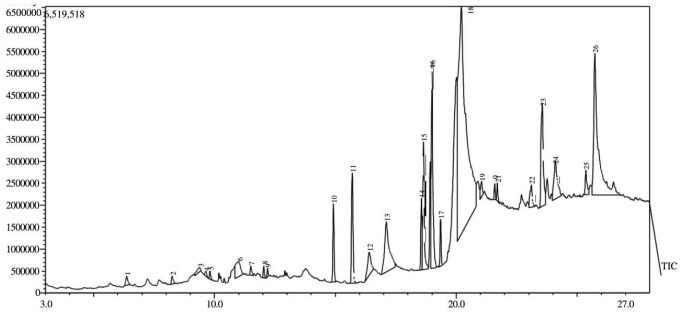

Figure 1. GC-MS chromatogram of the *D. microcarpum* stem bark extract

3.3. Phytocompounds identification

During GC-MS analysis of the *D. microcarpum* stem bark extract, 26 phytochemical components (Figure 1) were discovered. The compounds were identified using the peak, area, retention time (RT), and molecular formula (WM), as indicated in Table 3. The first chemical found was (-)-L-Dibenzoyl-tartaric (RT = 6.37 min, MW = 358, RI = 2883), while the last compound detected was Lineoleoyl chloride (RT = 25.73 min, MW = 298, RI = 2130). At 20.21 RT, Oleic acid ($C_{18}H_34O_2$) has a maximum volume of 40.37 percent, whereas 1, 3-Dimethyl-2,4-dinitrobenzene ($C_8H_8N_2O_4$) has a minimum volume of 0.2 percent. Others found include Oleoyl chloride ($C_{18}H_33ClO$) RT = 18.99 and 7.69 percent volume, 13-Octadecenal (Z) ($C_{18}H_34O$) RT = 23.57 and 5.6 percent volume, and Methyl 11-octadecenoate ($C_{19}H_{36}O_2$) RT = 18.65 min, 4.42 percent volume, and 21 others in decreasing order.

4. DISCUSSION

Detarium microcarpum species were reported to have shown considerable healing effect when the prepared decoction of either the stem barks or roots is administered as widely claimed by traditional healers (Mogana et al., 2020). The stem bark extract showed potent (bactericidal) inhibition at varying concentrations in this work. The differences observed in the effect of the active concentration on respective bacteria could be relatively explained by the number and type of fatty acids (Knapp & Melly, 1986; Mcgaw et al., 2002a) contained in the extract, as observed in Table 3.

Similarly, it was observed, long and medium chains of unsaturated fatty acids formed more than 50% of the fatty acid identified in this plant and were reported to be more active against gram-positive(*S. aureus* and *P. aeruginosa*) bacteria (Nguyen et al., 2017). Methyl esters of Arachidic acid, Dodecanoic acid 4-methyl- methyl, n-Hexadecanoic acid and stearic acid, Z-7-Tetradecenal, Glycerol 1,2-dipalmitate, Sucrose, Lineoleoyl chloride and Oleic acid are the most abundant compounds in this plant. Some investigators have also reported the abundance of these compounds in *D. microcarpum* and other plants and their bioactive properties. For example, Ololade et al. (2016) reported the significant inhibitory effects of essential oils extracted from *Annona muricata* (palmitic acid, oleic acid, cis- 9-hexadecanal, cis-9-

Table 3
Phytocompounds identified from the methanol crude extract of *D. microcarpum* stem bark

Peak	R/T	Area %	M/W	Compound Name	Chemical formula	RI
1	6.37	0.43	358	(-)-L-Dibenzoyl-tartaric acid	$C_{18}H_{14}O_8$	2883
2	8.25	0.33	150	D-Verbenone	$C_{10}H_{14}O$	1119
3	9.40	0.41	110	Resorcinol	$C_6H_6O_2$	1122
4	9.64	0.18	176	3,5-Dodecadiyne, 2-methyl-	$C_{13}H_20$	1284
5	9.82	0.22	186	1-Dodecanol	$C_{12}H_{26}O$	1457
6	11.02	1.74	342	Sucrose	$C_{12}H_{22}O_{11}$	3139
7	11.51	0.32	256	Benzene-1,2-dicarboxylic acid, monobenzyl ester	$C_{15}H_{12}O_4$	2203
8	12.04	0.31	194	5,7-Dodecadiyn-1,12-diol	$C_{12}H_{18}O_2$	1734
9	12.20	0.2	196	1,3-Dimethyl-2,4-dinitrobenzene	$C_8H_8N_2O_4$	1698
10	14.92	2.18	326	Arachidic acid methyl ester	$C_{21}H_{42}O_2$	2276
11	15.70	4.09	228	Dodecanoic acid, 4-methyl-, methyl ester	$C_{14}H_{28}O_2$	1516
12	16.40	1.81	256	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	1968
13	17.11	5.46	172	Decanoic acid	$C_{10}H_{20}O_2$	1372
14	18.56	2.03	322	11,14-Eicosadienoic acid, methyl ester	$C_{21}H_{38}O_2$	2292
15	18.65	4.42	296	Methyl 11-octadecenoate	$C_{19}H_{36}O_2$	2085
16	18.99	7.69	300	Oleoyl chloride	$C_{18}H_{33}ClO$	2131
17	19.35	1.19	298	Stearic acid, methyl ester	$C_{19}H_{38}O_2$	2077
18	20.21	40.37	282	Oleic Acid	$C_{18}H_{34}O_2$	2175
19	21.06	0.64	266	Olealdehyde	$C_{18}H_{34}O$	2007
20	21.59	0.38	330	Glycerol 1-monopalmitate	$C_{19}H_{38}O_4$	2482
21	21.70	0.42	330	Glycerol 1-palmitate	$C_{19}H_{38}O_4$	2482
22	23.11	1.17	238	11-Hexadecenal, (Z)-	$C_{16}H_{30}O$	1808
23	23.57	5.6	266	13-Octadecenal, (Z)	$C_{18}H_{34}O$	2007
24	24.08	3.09	568	Glycerol 1,2-dipalmitate	$C_{35}H_{68}O_5$	4019
25	25.36	1.03	210	Z-7-Tetradecenal	$C_{14}H_{26}O$	1609
26	25.73	14.29	298	Lineoleoyl chloride	$C_{18}H_{31}ClO$	2130

R/T = Retention time, RI=Retention index, % = percentage, M/W = Molecular weight

octadecanal, pentadecanoic acid) against six multidrug-resistant bacteria, including *S. typhimurium*, *P. aeruginosa*, *E. coli* and *S. aureus*.

Oleic acid, Oleoyl chloride and palmitic acid and their derivatives here identified, and other fatty acids esters from the stem bark extract can modify bacterial cell membrane, thereby obstructing oxygen intake and transport of amino acids necessary for cell growth and repairs (Freese et al., 1973). Oleic and Linoleic acids were reported to inhibit *S. aureus* but not active against *K. pneumoniae*, *E. coli*, and *P. aeruginosa* (Dilika et al., 2000). Thus, the gram-positive bacteria tested bactericidal activity could be partly due to some of these fatty acids (Mustapha & Runner, 2016; Raval et al., 2016) seen in Table 3. A synergistic action was also reported of activity of these identified compounds on the tested bacteria in bioactivity between oleic and linoleic acids and between linolenic acid and monoglyceride on inhibition of *S. aureus* (Dilika et al., 2000) as further evidence to support the potentials of long-chain fatty acid remained the possible factor for inhibition of bacteria (gram-positive) tested. However, susceptibility of gram's negative (*E. coli, K. pneumonia* and *S. typhi*) bacteria could not be explained in this context because they are affected by short-chain fatty ($\leq$ 6 Carbon atoms) (Mcgaw et al., 2002a)that were not observed. The susceptibility of bacteria (gram-negative) was influenced by pH and concentration of short-chain fatty acids of a plant (; McGaw et al., 2002a) (Freese et al., 1973; Mcgaw et al., 2002b). Lineoleoyl chloride and 4-Dodecanol were earlier described as potent antimicrobial agents (Jenecius & Mohan, 2014; Saravanan et al., 2013), while n-hexadecanoic acid has nematicide and pesticide effects (Jananie et al., 2011).

Although high numbers of active bioactive compounds were identified, no individual compound was isolated and tested in this work and therefore could not conclude the specific reason for the activity. Nevertheless, to corroborate current findings with previous ones, many other researchers have previously reported the antibacterial activity of *Detarium* against common bacterial pathogens but did not identify the possible phyto-compounds involved (P.M. Abreu et al., 1998; Okwu & Uchegbu, 2009; Sowemimo et al., 2011). Similarly, the susceptibility of these common pathogens toD.senegalense in addition to methanolic extract of the stem back of *D. microcarpum* species has also been reported by Okwu and Uchegbu (2009) and Olugbuyiro et al. (2009).

5. CONCLUSION

This study has provided data on the chemical composition of *D. microcapum*, which is traditionally used to treat urogenital infections, haemorrhoids, intestinal tract infections and pains relievers, and the antibacterial activity of methanol crude extract of stem bark. To date, there are very few reports on the determination of chemical constituents from methanol crude extract of *D. microcapum* stem bark using chromatographic techniques. Here, the bioactivity and presence of some compounds GC-MS identified are reported. The antibacterial activity of the identified compounds directly correlates with the bioactivity of the same compound isolated elsewhere, giving a clear insight on the potential of the plant and supporting the claim for its traditional usage. This report provides for quality control of the herbal use of the plant-based on the concept of phyto-equivalence of herbs, validation of the natural herbal medicine and could be used for new drug formulation in combating the threat of drug resistance by pharmaceutical industries. However, the research recommends further and exhaustive research to ascertain actual active constituents as possible drug candidate.

CONFLICTS OF INTEREST

All authors declare that there is no conflict of interest.

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AUTHOR CONTRIBUTIONS

MD, KOS, TMA - Research concept and design; MD - Collection and/or assembly of data; MD, KOS, TMA - Data analysis and interpretation; MD, KOS - Writing the article, MD, TMA - Critical revision of the article; MDTMA - Final approval of the article.

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