JOPAT Vol 20(2) 640– 655, July – Dec. 2021 Edition.

ISSN2636 – 5448 https://dx.doi.org/10.4314/jopat.v20i2.2

Phytochemical Evaluation and GC-MS Profiling of the Dichloromethane and Ethanol extracts of *Ocimum gratissimum* L. and *Lasianthera africana*. BEAUV

Haruna Baba[*1, 2], Adesegun J. Kashimawo[3], Adrienne C. Ibe[1]

1. Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Madonna University, Elele, Nigeria.
2. Department of Pharmaceutical and Medicinal Chemistry, Faculty of Pharmacy, University of Calabar, Calabar, Nigeria.
3. Department of Pharmaceutical and Medicinal Chemistry, Faculty of Pharmacy, Niger Delta University, Wilberforce Island, Bayelsa state, Nigeria.

ABSTRACT

The study was designed to investigate the bioactive constituents in the dichloromethane (DCM) and ethanol (ETOH) extracts of *Ocimum gratissimum* and *Lasianthera africana* which are commonly used in the treatment of diverse kinds of ailments especially in the southern part of Nigeria. Qualitative phytochemical screening was carried out on the extracts of the plants following documented procedures and gas chromatography-mass spectrometry (GC-MS) was also conducted comparing the mass spectra of the identified compounds with those of the National Institute of Standards and Technology database library. The preliminary phytochemical screening revealed the presence of secondary metabolites such as saponin, tannins, alkaloids, flavonoids and steroids in the extracts of both plants. The GC-MS analysis revealed the presence of both aliphatic and aromatic hydrocarbons as well as the terpenes and fatty acids in the DCM and ETOH extracts of *O. gratissimum* and *L. africana*. Some of these compounds are phenol; phytol and phytol acetate; squalene; caryophyllene; carmigrene; vitamin E; hexanedecanoic acid; 9,Octadecanoic acid (Z)-2-hydroxy-1-(hydroxymethyl)ethyl ester; 9,12,15-octadecatrienoic acid; 1-heptatriacotanol; tert-butylhydroquinone ; methyl (Z)-5,11,14,17-eicosatetraenoic acid; benzene; decahydro-4a-methyl-1 methyl-7-(1-methyl ethenenyl)-naphthalene. The presence of these multifunctional bioactive compounds in *O. gratissimum* and *L. africana* explains the diverse usage of these plants in ethnomedicine.

Key words: Phytochemical evaluation, GC-MS profiling, *Ocimum gratissimum*, *Lasianthera africana*, dichloromethane extract, ethanol extract.

***Corresponding author's email address: harunababa@unical.edu.ng, babharun1@gmail.com.**

Phone: +234-703-440-2435

INTRODUCTION

The use of medicinal plants to manage and treat different ailments predates modern history. Herbal medicine or alternative medicine has enjoyed renewed interest in recent times due to the rising cost of orthodox medicine and its accessibility to rural dwellers. It has been estimated that more than 80% of the world population rely on medicinal herbs for their primary health care needs [1]. Moreover, several medicines in clinical use today are directly derived from exploration of medicinal plants [2, 4, 5]. As such, medicinal plants would continue to play a bigger role in disease management even in distant future. Medicinal plants are present in all parts of the globe. However, the tropical climates are known to be home to vast majority of them [3]. The pharmacological properties of medicinal plants have been linked to the abundant presence of different types of secondary metabolites such as the terpenoids, alkaloids, flavonoids, steroids etc. [4, 5]. In the southern part of Nigeria, some herbs with known medicinal properties are used on routine basis for the preparation of soups and spices for their health benefits [3]. *Ocimum gratissimum* and *Lasianthera africana* are two of such herbs.

O. gratissimum, L. family; Lamiaceae, is used extensively in many cultures for several purposes. The essential oil of this plant is often used to treat different kinds of infectious diseases [6]. Its effectiveness has been linked to the presence of copious amount of eugenol [7-9]. The Igbos in south-east Nigeria uses the plant to manage baby's cord [10]. In Brazil, the leaves decoction is used as a mild sedative for children [11]. The leaf extracts of *O. gratissimum* have shown activity against different causative agents of diarrhoea, including *Shigellae* [12, 13]. Some chemical compounds previously isolated from *O. gratissimum* include beta selinene, bisabololine, alpha humelene, ocimene, oleanolic acid, terpinolene, farnesene [14 - 17].

L. africana, BEAUV a glabrous medicinal plant of the family, Icacinaceae reaches to the height of 61-136 cm [18]. In the southern part of Nigeria where it is used as one of the top six vegetables in the preparation of soup, it is called by different local names. The Ibibios and Efiks of Akwa Ibom and Cross River states call it "Editan" [18]. The plant, in addition to its culinary uses has been reported to be used in the treatment of various ailments including high fever, diarrhoea and candidiasis [19]. The following secondary metabolites were reported in the extracts of *L. africana* viz; alkaloids, terpenes, saponins, tannins, flavonoids, anthraquinones, and cardiac glycosides [20]

The workhorse of contemporary plant metabolite profiling is no doubt gas chromatography coupled to mass spectrometry (GC-MS) [21]. Though information abounds as to the usefulness of the aforementioned plants, their chemical compositions have not been exhaustively reported. Therefore, the preliminary phytochemical screening and GC-MS analyses of the dichloromethane and

ethanol extract of these two important medicinal plants were carried out to profile the chemical constituents in the plants that are responsible for their medicinal properties.

MATERIALS AND METHODS

Chemicals

The solvents (dichloromethane and ethanol) used for the extraction were purchased from QauliChem (India). All other reagents used were of analytical standard.

Plant materials

The leaves of *O. gratissimum* and *L. africana* were obtained from Elele, Rivers state and Uyo, Akwa Ibom state respectively in the month of September, 2019. They were authenticated at the Department of Pharmacognosy, Faculty of Pharmacy, Madonna University. The leaves were then washed, dried and pulverized. The voucher specimen of *O. gratissimum* was prepared and deposited at the Forest Research Institute of Nigeria, Ibadan; with number FHI 113333.

Extraction

The powdered leaves material; 150g of each of the plant was first exhaustively extracted with dichloromethane and subsequently with ethanol by cold maceration procedure. The extracts were filtered using Whatman number 1 filter paper. The resulting filtrate was evaporated to dryness with the aid of rotary evaporator at room temperature. The extracts were stored in a desiccator for further use.

Phytochemical screening

The test was carried out based on procedures outlined by Harbourne, [22] and Sofowora [23]. Preliminary phytochemical screening was performed on the dichloromethane and ethanol extracts for tannins, flavonoids, anthraquinones, saponins, terpenoids, steroids, and alkaloids.

GC-MS analyses

GC- MS analysis was conducted using GC/MS- QP 2010SE SHIMADZU JAPAN with injector temperature of 250˚C and carrier gas pressure of 144.4 kPa. The column length was 30 m with a diameter of 0.25 mm and the flow rate of 38.7ml/min. The temperature program was used as follows: initial temperature of 60˚C (hold time: 2min) programmed at a rate of 12˚C/min to a final temperature of 300˚C (hold time: 2min). The samples were dissolved in dichloromethane and 1 µL volumes were injected into the machine. Nitrogen was used as carrier gas at a flow of 3ml /min, split ratio 10:1. The eluates were automatically passed into a mass spectrometer with a detector voltage set at 1.38 kV and sampling rate of 0.5 sec. The mass spectra was recorded over 45 – 700 amu and revealed the Total Ion Current chromatograms. The temperatures of injector and transfer line were kept at 250˚C and that of the ion source at 230˚C.

RESULTS

Phytochemical Screening

The results of the phytochemical screening of the dichloromethane and ethanol extracts of *O. gratissimum* and *L. africana* are shown in table 1. It revealed the presence of carbohydrate, saponin, tannins, alkaloids, flavonoids and steroids. Anthraquinone and protein were absent in the extracts of the two plants. Mayer's reagent revealed absence of alkaloid in the extracts of the two plants. The compounds revealed by the GC-MS are shown in table 2-5.

Table 1: Phytochemical screening of the dichloromethane and ethanol extracts of *Ocimum gratissimum* and *Lasianthera Africana*

Phytochemical Constituents of Dichloromethane extract	Type of Test	*Ocimum gratissimum*		*Lasianthera africana*	
		Dichloro-methane extract	Ethanol Extract	Dichloro-methane extract	Ethanol Extract
Carbohydrate	Molisch's	+	+	+	+
Saponin	Frothing	+	+	+	+
	Emulsion	+	+	-	+
Tannins	Ferric chloride	+	+	+	+
	Lead acetate	-	+	+	+
Alkaloids	Mayer's	-	-	-	-
	Dragendorff's	+	+	+	+
	Wagner's	-	+	+	+
	Picric	+	+	+	+
Protein	Million's	-	-	-	-
Flavonoid	Ammonium	-	+	+	+
Steroids	Salkwoski's	+	+	+	+
Anthraquinone	Borntrager's	-	-	-	-

(+ present; - absent)

Table 2: Chemical constituents of dichloromethane extract of *O. gratissimum*.

Peak No	% Area	Retention Time	Compounds	Biological activity
15	7.94	10.975	Benzene	The bio-oxidation gives carcinogenic by-products. It has no useful biological activity.
22	6.85	12.538	Caryophyllene	Anti-inflammatory, antibiotic, antioxidant, anticarcinogenic and local anaesthetic activities
29	18.99	13.312	Decahydro-4a-methyl-1 methyl-7-(1-methyl ethenenyl)-naphthalene	Responsible for the biosynthesis of lovastatin
30	5.86	13.364	Chamigrene	Antibacterial activity
33	6.09	13.927	Hydroquinone tert-Butyl	Prevents oxidative injury
34	2.61	14.298	Methyl (Z)-5,11,14,17-eicosatetraenoic acid	Relax smooth muscle
49	3.90	18.912	Phytol	anxiolytic, metabolism-modulating, cytotoxic, antioxidant, autophagy- and apoptosis-inducing, anti-nociceptive, anti-inflammatory, immune-modulating, and antimicrobial effects
55	3.82	19.950	Phytol acetate	Same as phytol
68	3.27	25.085	Squalene	Reduces free radical oxidative damage to the skin

Table 3: Chemical constituents of Ethanol extract of *O.gratissimum*.

Peak No	%Area	Retention time	Compound	Biological activity
4	23.75	10.977	2,3,5,6-Tetramethyl phenol	Antimicrobial, anti-inflammatory, antioxidant
35	12.45	18.920	Phytol	anxiolytic, metabolism-modulating, cytotoxic, antioxidant.
36	3.92	19.268	5alpha-Pregn-16-en-20-one, 12.beta.-hydroxy-, acetate	Precursor/intermediate in the synthesis of most steroid hormones.
39	2.98	19.943	Phytol acetate	Same as phytol
45	3.42	22.806	Cholestan-7-one, cyclic 1,2-ethanediyl acetal, (5-alpha)	A key intermediate in the synthesis of the steroidal moiety of oogoniols.
48	3.34	24.142	1-Heptatriacotanol	Antimicrobial, anticonvulsant, antidepressant, anti-inflammatory,
49	6.56	24.375	9-Octadecenoic acid (Z)-, 2-hydroxy-1-(hydroxymethyl)ethyl ester	Antibacterial activity.

Table 4: Chemical constituents of Dichloromethane extract of *L. africana*.

Peak No	% Area	Retention time	Compound	
1	4.32	10.977	2-Methyl-5-(1-methylethyl)-phenol	Precursor in the biosynthesis of diverse polyphenolic compounds.
16	2.37	16.396	3,7,11,15-Tetramethyl-2-hexadecane-1-ol	No beneficial biological activity recorded.
20	7.27	17.161	Hexadecanoic acid methylester	Moderate antimicrobial activity
21	5.51	17.531	n-Hexadecanoic acid	Anti-inflammatory, anticancer, and antioxidant.
24	7.45	18.807	9,12,15-Octadecatrienoic acid methylester	Antibacterial and anticandidal Antiinflammatory, hypocholesterolemic, cancer preventive.
25	2.66	18.911	2-Hexadecen-1-ol, 3,7,11,15-tetramethyl	Food additive
27	7.53	19.290	9,12, Octadecadienoic acid	Used in the synthesis of prostaglandin and cell membrane.
28	4.34	19.499	9,12,15-Octadecatrienoic acid, 2,3-dihydroxy propyl ester	Inhibition of 5-lipoxygenase.
34	6.57	22.804	Vitamin E	Antioxidant, gene expression, cell signaling.
35	8.48	24.244	9,Octadecanoic acid (Z)-2-hydroxy-1-(hydroxymethyl)ether ester	Antimicrobial activity.
36	4.16	24.931	9,12,15-Octadecatrienoic acid,	Antimicrobial activity.
37	11.73	25.085	Squalene	Reduces free radical oxidative damage to the skin
38	3.22	25.483	1-Octadecanesulphonyl chloride	Not reported

Table 5: Chemical constituents of Ethanol Extract of *L. africana*.

Peak No	%Area	Retention time	Compound	Activity
7	3.37	16.396	Phytol acetate.	Same as phytol
12	3.39	17.534	n-Hexadecanoic acid.	Anti-inflammatory, anticancer, and antioxidant.
13	5.09	17.747	Hexadecanoic acid ethyl ester.	Moderate antimicrobial activity
16	18.99	18.918	Phytol.	anxiolytic, metabolism-modulating, cytotoxic, antioxidant.
19	12.06	19.502	9,12,15-Octadecatrienoic acid, ethyl ester.	Inhibition of 5-lipoxygenase.
23	3.13	22.451	Z,Z-3,13-Octadecedien-1-ol	cytotoxic , , antifungal , and analgesic
25	6.99	22.804	Vitamin E.	Antioxidant, gene expression, cell signaling.
28	8.75	24.244	9-Octadecanoic acid (Z)-2-hydroxyl-1-(hydroxymethyl)ethyl ester.	Has been reported to inhibit the proliferative effect in keloid fibroblasts
30	4.66	24.931	2,6,10,14,18-Pentamethyl-2,6,10,14,18-eicosapentaene	Antibacterial, antifungal, antitumor.
31	3.94	25.082	Squalene.	Reduces free radical oxidative damage to the skin

LIST OF TITLES FOR FIGURES

1. GC-MS Spectrum of the dichloromethane extract of *Ocimum gratissimum*

2. GC-MS Spectrum of the ethanol extract of *Ocimum gratissimum*

3. GC-MS spectrum of the dichloromethane extract of *Lasianthera africana*

4. GC-MS spectrum of the ethanol extract of *Lasianthera africana*

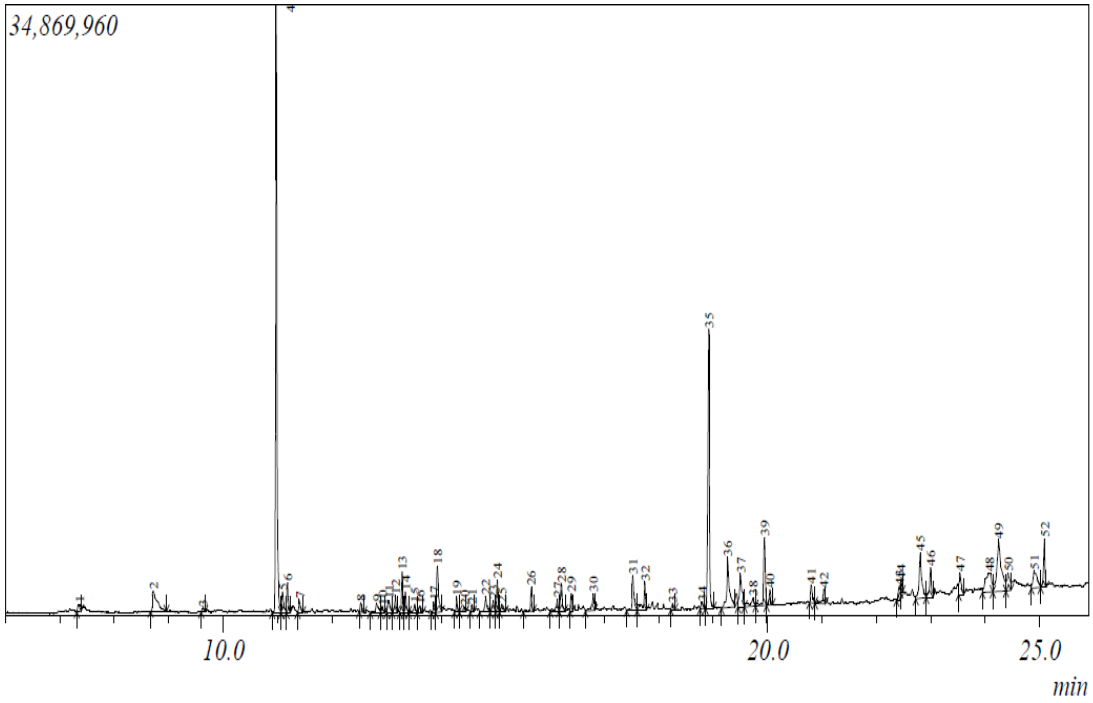

Figure 1: GC-MS Spectrum of the dichloromethane extract of *Ocimum gratissimum*

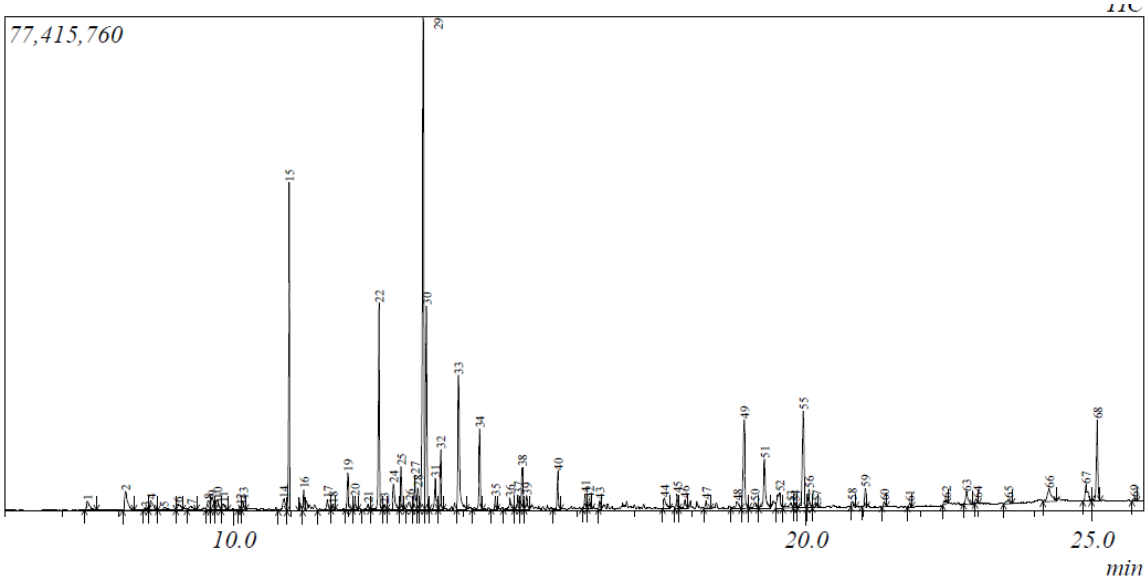

Figure 2: GC-MS Spectrum of the ethanol extract of *Ocimum gratissimum*

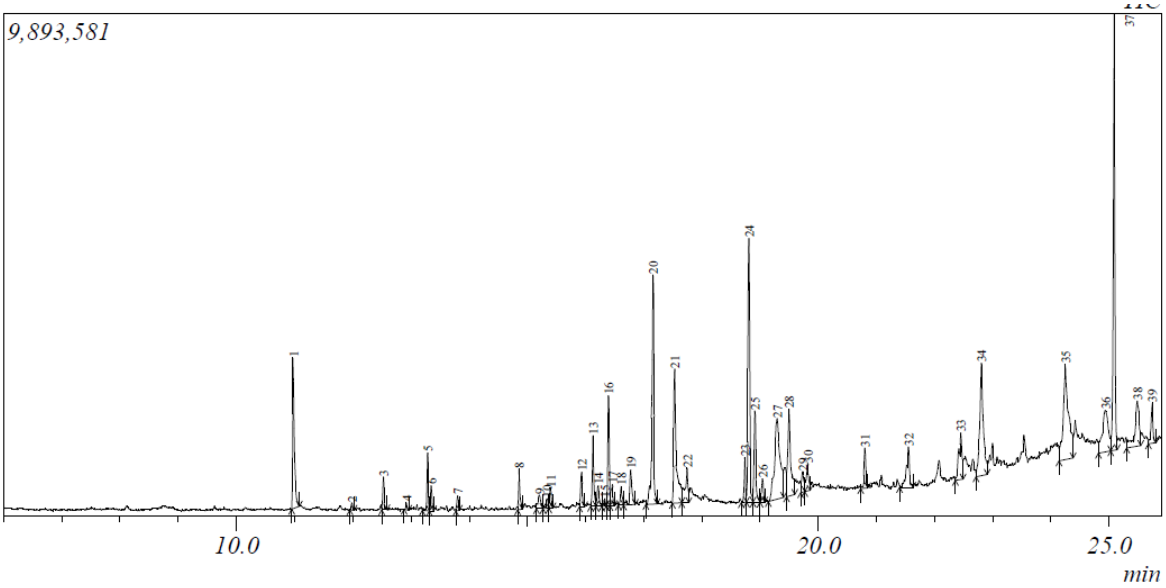

Figure 3: GC-MS spectrum of the dichloromethane extract of *Lasianthera africana*

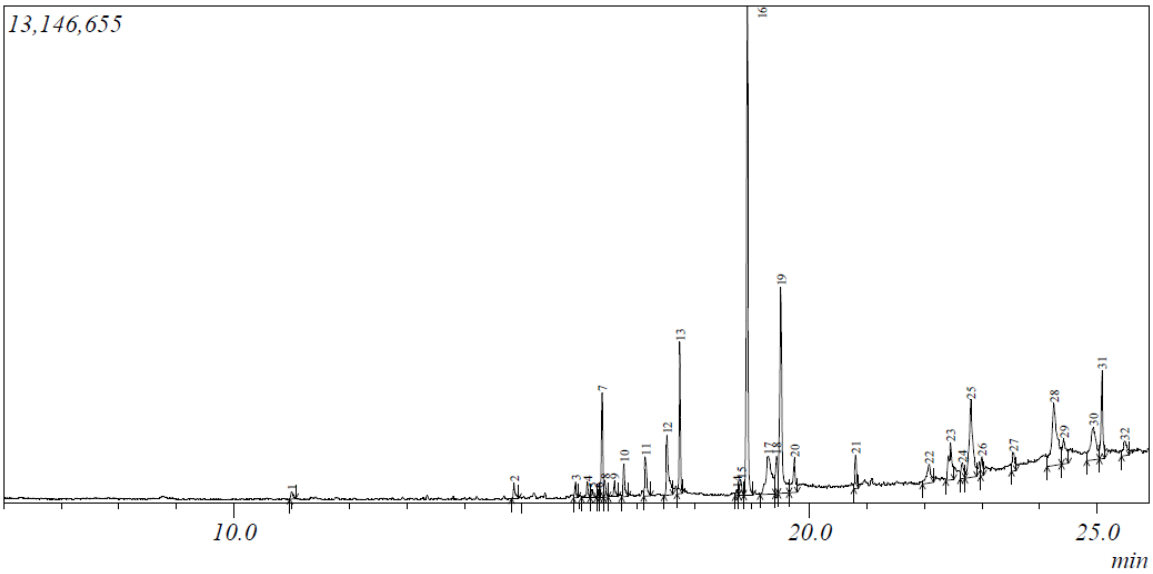

Figure 4: GC-MS spectrum of the ethanol extract of *Lasianthera Africana*

DISCUSSION

The phytochemical screening of these two important herbs revealed the presence of carbohydrate, saponin, tannins, alkaloids, flavonoids and steroids. Plant secondary metabolites are known to possess various bioactivities; which are responsible for their usefulness as medicinal plants. The presence of alkaloids confers on the plant the following activities; antimicrobial, antifungal, anthelminthics, antidiarrheal, anti-inflammatory, antihypertensive, antimalarial, antidepressant, anaesthetic and amoebicide [24-26]. Flavonoids on the other hand are known for their antioxidant activity, hypolipidemic, antimicrobial, antiviral, tumor inhibitory, vasoprotective, anti-thrombotic and anti- allergic activity [27]. Plants containing saponin could be useful as immune boosters, anti-inflammatory, cholesterol lowering, anti-cancer and as antidiarrheal [28].Tannins are responsible for anti-microbial, anti-inflammatory and anti-diarrheal properties among others [29]. Steroids are useful as cardiotonic, insecticidal and antimicrobial agents [30]. The aforementioned properties of the secondary metabolites found in the extracts of *O. gratissimum* and *L. africana* could be responsible for the wide applicability of these two medicinal herbs. The GC-MS analysis further unveiled some important chemical constituents in *O. gratissimum* and *L. africana*. Prominent among them is phytol; a diterpine alcohol and its acetate which are reported to have many important biological functions such as anxiolytic, metabolism-modulating, cytotoxic, antioxidant, autophagy- and apoptosis-inducing, anti-nociceptive, anti-inflammatory, immune-modulating, and antimicrobial effects [32,35]. Caryophyllene, a natural bicyclic sesquiterpene detected in the dichloromethane extract of *O. gratissimum* is known for anti-inflammatory, antibiotic, antioxidant, anticarcinogenic and local anaesthetic activities [31]. Other terpenes in the same extract are chamigrene and squalene. While chamigrene exhibits certain level of antimicrobial activity, squalene reduces free radical oxidative damage to the skin [33, 36]. Fatty acid was equally detected by the GC-MS analysis. One of such is eicosatetraenoic acid; which is known to have smooth muscle relaxing property [34]. It is interesting to note that the terpenes which were found in the dichloromethane extract were not detected in the ethanol extract of the plant. This would serve as a useful guide in plant extraction when certain hydrocarbons are to be avoided. Less polar solvent such as dichloromethane could be used to remove the unwanted hydrocarbons. The major constituents of the ethanol extract detected by the GC-MS analysis are: phenols, diterpene alcohol (phytol), fatty acids and steroids. Phenols are known to have some useful biological activities including; antimicrobial, anti-inflammatory and antioxidant property [37, 38]. 1-Heptatriacotanol ($C_{37}H_{76}O$) which was also found in the ethanol extract of *O. gratissimum* is known to exhibit the following

activities; antimicrobial, anticonvulsant, antidepressant, anti-inflammatory, analgesic, antiplatelet, antimalarial, anticancer, antifungal, antitubercular, antiviral and cardio protective [39].

The GC-MS analysis of the dichloromethane extract of *L. africana* contains some vital compounds such as 2-Methyl-5-(1-methylethyl)-phenol , 3,7,11,15-Tetramethyl-2-hexadecane-1-ol, Hexadecanoic acid methylester , n-Hexadecanoic acid, 9,12,15-Octadecatrienoic acid methylester, 2-Hexadecen-1-ol, 3,7,11,15-tetramethyl, 9,12, Octadecadienoic acid, 9,12,15-Octadecatrienoic acid, 2,3-dihydroxy propyl ester, Vitamin E, 9,Octadecanoic acid (Z)-2-hydroxy-1-(hydroxymethyl) ether ester, 9,12,15-Octadecatrienoic acid, squalene, 1-Octadecanesulphonyl chloride. The phenols and fatty acids are known to prevent neurodegeneration, and also function as food additives [40-44]. This could explain the use of the plants in preparation of soups and spices [3]. Vitamin E found in both the dichloromethane and ethanol extracts is a known antioxidant [45]; and the usefulness of the vitamin cannot be over emphasized. In addition to these, the ethanol extract of *L. africana* contains some aliphatic alcohols fatty acids and branched chain hydrocarbons which are indicated for various important functions in the body [46-48]

CONCLUSION

The presence of the diverse multifunctional compounds in *O. gratissimum* and *L. africana* could explain the wide spread use of these two medicinal plants to treat and manage various kinds of health complaints in so many rural and even urban communities in Nigeria.

ACKNOWLEDGEMENT

The assistance of Mr. Chris Nwadilibe, the technologist in the Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Madonna University is acknowledged by the authors.

REFERENCES

1. Johanna, K., Harald, F., Elfriede P., and Michael H.(2005). Historical and modern plant uses-the example of the Ch'orti'Maya and Ladinos in Eastern Guatemala. *J. Pharm. Pharmacol*., 57:1127-1152.

2. Rates, S. K.(2001). *Plants as a Source of drugs. Toxicon., 39: 603-613*

3. Baba, H., Vwioko, O., and Timikare J. (2018). Free radical scavenging activity of two edible vegetables from the Niger delta region of Nigeria. GSC Biological and Pharmaceutical Sciences. 05(02): 069–073

4. Ameenah, G. F. (2006). Medicinal plants: Traditions of yesterday and drugs of tomorrow. Molecular Aspects of Medicine, 27: 1-93.

5. Kong, J. M., Goh, N. K., Chia, L., and Chia, T. F. (2003). Recent advances in traditional plant drugs and orchids. *Acta Pharmacol Sin*., 24(1): 7-21.

6. Adebolu, T. T., and Oladimeji, S. A. (2005). Antimicrobial activity of leaf extracts of *Ocimum gratissimum* on selected diarrhoea causing bacteria in southwestern *Nigeria. Afr J Biotech.* 4: 682-4

7. Matasyoh, L. G., Matasyoh, J. C., Wachira, F. N., Kinyua, M. G., Thairu, M. A. W., and Mukiama, T. K. (2008). Antimicrobial activity of essential oils of *Ocimum Gratissimum* from different populations of Kenya. *Afr J Tradit Complementary Altern Med.* 5: 187-93.

8. Ahonkhai, I., Ba, A., Edogun, O., and Mu, U. (2009). Antimicrobial activities of the volatile oils of *Ocimum bacilicum* L. and *Ocimum gratissimum* (Lamiaceae) against some aerobic dental isolates. *Pak J Pharm Sci*. 22: 405-9.

9. Amengialue, O.O., Edobor, O., and Egharevba, A. P. (2013). Antibacterial activity of extracts of *Ocimum gratissimum* on bacteria associated with diarrhea. *Bayero J Pure Appl Sci*. 6: 143-5.

10. Okechukwu, O. I, and Tahir, F. (2008). Phytochemical and antimicrobial screening of the leaf extract of *Ocimum gratissimum* L. *J Econ Taxon Bot.* 32: 171-7.

11. Di Stasi, L. C., Oliveira, G. P., Carvalhaes, M. A., Queiroz-Junior, M., Tien, O. S., Kakinami, S. H., and Reis, M. S.(2002). Medicinal plants popularly used in the Brazilian Tropical Atlantic Forest. Fitoterapia. 73(1): 69-91

12. Nweze, E. I., and Eze, E. E. (2009). Justification for the useof *Ocimum gratissimum* L in herbal medicine and its interaction with disc antibiotics. Complementary and Alternative Medicine. 9(37):1472.

13. Oboh, F. O. J., Madsodje, H. I., and Enabulele, S. A. (2009). Nutritional and antimicrobial properties of *Ocimum gratissimum* leaves. Journal of Biological Sciences. 9(4):377-380.

14. Pandey, A. K., and Chowdhury, A. R. (2000). Composition of the essential oil of *Ocimum gratissimum* grown in Madhya Pradesh. *J Med Aromat Plant Sci*. 22(23): 26-8.

15. Matasyoh, L. G., Josphat, C. M., Francis, N. W., Miriam, G. K., Anne, W. T, M., and Titus, K. M. (2007). Chemical composition and antimicrobial activity of the essential oil of *Ocimum gratissimum* L. growing in Eastern Kenya. *Afr J Biotech*. 6: 760-5.

16. Jirovetz, L., Buchbauer, G., Ngassoum, M. B., Ngamo, L.T., and Adjoudji, O.(2005). Combined investigation of the chemical composition of essential oils of *Ocimum gratissimum* and *Xylopia aethiopica* from Cameroon and their insecticidal activities against stored maize pest *Sitophilus zeamais*. *Ernähr*, 29: 55-60.

17. Njoku, C. J., Zeng, L., Asuzu, I. U., Oberlies, N. H., and Mclaughlin, J. L. (1997).

Oleanolic acid, a bioactive component of the leaves of *Ocimum gratissimum* (lamiaceae). Int *J Pharmacognosy*. 35: 134-7.

18. Hutchison, J. and Dalziel, J. M. (1973). Flora of West Tropical Africa (2nd ed.) Crown Agents, London. 638.

19. Ebana, R. U. B., Essien, A. l and Ekpa, O. D. (1995). Nutritional and potential medicinal values of the leaves of *Lasianthera africana* (BEAUV). Global journal of pure science. 1:1-8.

20.Bassey, M. E, Etuk, E. U. I, Ubom, R. and Obop I. E. (2006). Chemotaxonomic study of *Lasianthera africana* (lcacinaceae) in Akwa Ibom State of Nigeria. Nigeria Journal of Botany. 19, 99-102

21. Kadhim, M. J, Sosa, A. A., and Hameed I. H. (2016). Evaluation of anti-bacterial activity and bioactive chemical analysis of Ocimum basilicum using Fourier transform infrared (FT-IR) and gas chromatography-mass spectrometry (GC-MS) techniques. International Journal of Pharmacognosy and Phytochemical Research. 8(6): 127-146.

22. Harborne J. B. (1973). Phytochemical methods, A Guide to Modern Technique of Plant analysis, Chapman and Hall Ltd., London New York. 33-38

23. Sofowora, A. (1993). Screening plants for bioactive agents. Medicinal plants and traditional medicine in Africa, spectrum books Ltd, Ibadan. 134

24. Sofowora, A. (1993). Medicinal Plants and Traditional Medicine in Africa. 2nd ed. Spectrum Books Ltd. Ibadan, Nigerian. 289.

25. Okwu, D. E,and Okwu, M. E. (2004). Chemical composition of *Spondiasmombin* Linn. Plants parts. *J Sustain Agric Environ*., 6:140-7.

26. Gurib-Fakim, A. (2006). Medicinal plants: traditions of yesterday and drugs of tomorrow. *Mol Aspects Med*., 27:1-93.

27. Trease, E., and Evans, W. C. (2002). Phenols and phenolic glycosides. In: Textbook of Pharmacology 15th ed. London: Elsevier Science Ltd. p. 214-52.

28. Mandeau, A., Debitus, C., Ariès, M. F, and David, B. (2005). Isolation and absolute configuration of new bioactive marine steroids from *Euryspongia* n.sp. Steroids.,70:873-8.

29. Rivière, C., Hong, V. N, Pieters, L., Dejaegher, B., Heyden, Y. V, Van, M. C, *et al.* (2009). Polyphenols isolated from antiradical extracts of *Mallotus metcalfianus*. *Phytochemistry*;70:86-94.

30. Okwu, D. E. (2004). Phytochemicals and vitamin content of indigeneous spices of South Eastern Nigeria. *J Sustain Agric Environ.* 6:30-4.

31. Bettarini, F., Borgonovi, G. E, Fiorani, T., Gagliardi, I., Caprioli, V., Massardo, P *et*

al. (1993). Antiparasitic Compounds from East African Plants: Isolation and Biological Activity of Anonaine, Matricarianol, Canthin-6-One and Caryophyllene Oxide. *Int J Trop Insect Sci* **14,** 93–99.

32. Islam, M. T., Ali, E. S., Uddin, S. J., Shaw S., Islam, M. A., Ahmed, M. I et al. (2018). Phytol: A review of biomedical activities. *Food Chem Toxicol*.121:82-94.

33. Mulyaningsih. S., Youns, M., El-Readi, M. Z., Ashour, M. L., Nibret, E., Sporer, F., Herrmann, F., Reichling, J. Wink, M.(2010). Biological activity of the essential oil of *Kadsura longipedunculata* (Schisandraceae) and its major components. Journal of Pharmacy and Pharmacology. 62: 1037-1044

34. Asakawa, Y., and Ludwiczuk, A. (2017). Chemical constituents of bryophytes: structures and biological activity. Journal of natural products. 81(3):641-60.

35. Nafis, A., Kasrati, A., Jamali, C. A., Custódio, L., Vitalini, S., Iriti, M., and Hassani, L. A. (2020).Comparative Study of the in Vitro Antimicrobial and Synergistic Effect of Essential Oils from Laurus nobilis L. and Prunus armeniaca L. from Morocco with Antimicrobial Drugs: New Approach for Health Promoting Products. Antibiotics. 9(4):140.

36. Oliveira, D. M, Furtado, F. B., Gomes, A. A., Belut, B. R., Nascimento, E.A., Morais, S.A., Martins, C.H., Santos, V. C., da Silva, C. V., Teixeira, T. L., and Cunha, L. C. (2020). Chemical Constituents and Antileishmanial and Antibacterial Activities of Essential Oils from *Scheelea phalerata*. ACS omega.5(3):1363-70.

37. Teixeira, B., Marques, A., Ramos, C., Serrano, C., Matos, O., Neng, N. R., Nogueira, J. M., Saraiva, J. A., and Nunes, M. L,. (2013). Chemical composition and bioactivity of different oregano (*Origanum vulgare*) extracts and essential oil. Journal of the Science of Food and Agriculture.93(11):2707-14.

38. Lee, K. S., and Lee, K. Y. (2010). Biological activity of phenol compound from a cactus Cheonnyuncho (*Opuntia humifusa*) in Korea. Journal of the Korean Society of Food Science and Nutrition.;39(8):1132-6.

39. Abdulhafiz, F., Mohammed, A., Kayat, F., Bhaskar, M., Hamzah, Z., Podapati, S. K, and Reddy, L. V. (2020). Xanthine Oxidase Inhibitory Activity, Chemical Composition, Antioxidant Properties and GC-MS Analysis of Keladi Candik (Alocasia longiloba Miq). Molecules. 25(11):2658.

40. Al Otibi, F., and Rizwana, H. (2019). Chemical composition, FTIR studies, morphological alterations, and antifungal activity of leaf extracts of Artemisia sieberi from Saudi Arabia. International journal of agriculture and biology. 21(6):1241-8.

41. Abbas R. K. (2019). Chemical Constituents of The Goat Margarine and Antibacterial Activity Against Bacterial Pathogens in Sudan. *J Pure Appl Microbiol*. 13(1):225-32.

42. Fatema, S., Ubale, M., Farooqui, M., and Arif, P. M. (2019). Analysis of biological activity and gas chromatography-mass spectrometry study of conventional extraction of Vitex negundo Linn. leaves. *Asian J Pharm Clin Res*. 12(1):289-92.

43.Kumari, A., Guleria, I., Pathania, D., and Verma, R. (2020). Phytochemicals analysis and antibacterial activity of leaf extracts of Populus ciliata Wall. ex Royle. Medicinal Plants-International Journal of Phytomedicines and Related Industries. 12(1):120-9.

44. Youssif, K., Elshamy, A., Rabeh, M., Gabr, N., and Haggag, E. (2019). A Phytochemical and Biological Review on Plants of The family Aizoaceae. Journal of Advanced Pharmacy Research. 3(4):158-81.

45. Miyazawa, T., Burdeos, G. C, Itaya, M., Nakagawa, K., and Miyazawa, T. (2019). Vitamin E: regulatory redox interactions. Iubmb Life.;71(4):430-41.

46. Prabhu, V., Poonkodi, K., Mini, R., Devi, K.V., and Priya, M. K., (2018). GC-MS analysis of bioactive compounds present in the petroleum ether, chloroform and methanol extract of ixora coccinea's flower and in-vitro cytotoxic activity of methanolic extract against DLA and EAC cancer cell lines. *Int. J. Res. Anal. Rev*. 5:801-7.

47. Mehra, M., Pasricha, V., and Gupta, R. K. (2015). Estimation of nutritional, phytochemical and antioxidant activity of seeds of musk melon (Cucumis melo) and water melon (Citrullus lanatus) and nutritional analysis of their respective oils. Journal of Pharmacognosy and Phytochemistry.;3(6):98-102.

48. Powder-George, Y. L., and Mohammed, F. K. (2018). GC-MS analysis of the bioactive phytoconstituents of various organic crude extracts from the seed kernels of Manilkara bidentata (balata) collected in Trinidad, WI. Natural Product Research. 32(3):358-61.