GC-MS Analysis of the Rauwolfia vomitoria Ethanol Extracts

I. I. Asoro¹*, O. A. T. Ebuehi¹ and M. N. Igwo-Ezikpe¹

¹Department of Biochemistry, College of Medicine, University of Lagos, P.M.B. 12003, Lagos, Nigeria.

Authors’ contributions

This work was carried out in collaboration between all authors. Author IIA designed the study and prepared the manuscript performed the statistical analysis. Author OATE substantively revised visualised and approved of the work and author MNIIE managed literature, reviewed, edited the work and references.

Article Information

DOI: 10.9734/EJMP/2021/v32i630398

Received 14 March 2021
Accepted 19 May 2021
Published 07 August 2021

ABSTRACT

Bioactive compounds are the frontline potent agents in both nutraceuticals and pharmaceutical industries. The bioactive compounds are gaining much importance for their ability in enhancing resistance to various diseases and to improve the health of people both by traditional and modern ways of administrations. R. vomitoria is one of the medicinal plants used traditionally to manage hypertension, diabetes and mental disorder. This present study sought to characterize the bioactive components of R. vomitoria leaf and root ethanol extracts using Gas-Chromatography-Mass Spectrophotometry (GC-MS). The results of the GC-MS analysis provide different peaks indicating the presence of 22 phytochemicals in the plant leaf and 16 phytochemicals in the root. The major bioactive compounds in the leaf were squalene (18.69%), phytol (16.47%), n-hexadecanoic acid (15.68%), 7-tetradecenal, (Z) (12.90%), 9,12,15-octadecatrienoic acid, ethyl ester, (Z,Z) (9.56%) and others, while the roots contain; cis-vaccenic acid (32.13%), n-hexadecanoic acid (15.41%), (E)-9-octadecenoic acid ethyl ester (9.83), cyclohexanecarbonitrile 1-(4-chlorophenyl (9.45%), 8H-azeceno[5,4-b] indol-8-one, 5-ethylidene (7.66%) and other minor compounds. Pharmacological
activities of these compounds indicated that the compounds present in the leaf of the plant can be used as a crude drug which could be developed into a novel drug. Some of these compounds have antimicrobial, antioxidant, hepatoprotective, hypocholesterolemic as well as cancer preventive activities amongst others. The findings suggest that there is an indication that both *R. vomitoria* leaves and roots contain potent bioactive compounds that may be linked to its beneficial effects on health, with the leaf taking the lead. It is therefore recommended as a plant of phytopharmaceutical significance.

Keywords: Rauwolfia vomitoria; leaf; root; bioactive compounds; phytochemicals.

1. INTRODUCTION

In many countries worldwide medicinal plants remain the dominant form of medicine for the treatment and prevention of a wide range of diseases. Medicinal plants used as alternative drugs are indicative of the vital role that plants play in many developing countries, and are also sources of novel plant-derived constituents that could be leads for treatment of malaria and other diseases [1]. Plants have the capacity of synthesizing the organic compounds and are called as secondary metabolites, they have unique and complex structures. The secondary metabolites are used in the treating of chronic as well as infectious diseases [2]. One of the plants of medicinal value from the humid tropics is *Rauwolfia*, a tropical shrub with white or greenish flowers. The plant *Rauwolfia vomitoria* belongs to the family Apocynaceae. *Rauwolfia vomitoria* is called serpent wood, serpent snake root and swizzle stick, as well as, “asofeyeje” in Yoruba, “ira” in Igbo, “wadda” in Hausa, “akata” in Bini and “utoenyin” in Efik as vernacular names. It is mostly found in the forest of the southern part of Nigeria [3]. Research showed that herbal preparations of alkaloid extract of *R. vomitoria* have been used in traditional folk medicine in Africa as antihypertensive [4]. *Rauwolfia vomitoria* is used for treating nervous conditions [5] and can also act as antioxidant and anti-inflammatory [6], antiglycemic [7], anticonvulsant [8]. Administration of ethanolic leaf and root bark extracts of *Rauwolfia vomitoria* on the 7th through 14th day of gestation may be cardiotoxic on the fetal heart of the developing rats and the extract of the root bark has more teratogenic potentials than the leaf extract [9]. The root bark extract of *R. vomitoria*, has great potential in the management of psychotic disorders [8]. Methanolic extract can be used as antimalaria [10].

Aqueous extract of *Rauwolfia vomitoria* can be used to treat typhoid, and jaundice [11] while, *Rauwolfia vomitoria* with or without vitamin E improved the immunity and enhances the hematological indices [12]. Aqueous methanolic extract of *Rauwolfia vomitoria* leaves are used also as antisickling agents [13].

There has been an increasing interest on natural product research especially on medicinal plants which seem to have restorative properties [14]. Bioactive compounds are used in pharmaceutical industries as potents agents in treatment of many diseases. Bioactive compounds are gaining importance for the treatment of many diseases in recent days [15]. Identification and evaluation of these active compounds otherwise known as phytochemicals of uncommonly used plants could help provide information that would be useful in the development of a new drug [16] or in the production of a nutraceuticals. Phytochemicals are naturally occurring chemical compounds found in medicinal plants, leaves, vegetables and roots. They possess variety of protective properties against various diseases. The phytoconstituents from most medicinal plants for example flavonoids are considered as supplemental interventions for health subsistence and disease management. The biological activity of flavonoids in neurodegenerative disorders, inflammation, cancer and cardiovascular diseases involves the regulation of cell growth and production, enzyme activity and the accent of cellular signaling cascades [17].

Over the last few decades, use of herbal drugs has been emphasized due to their easy availability, therapeutic potential, least side effects and minimum cost. At present nearly 80% of the world populations rely on plant based drugs for their health care need [18]. GC-MS is the best technique to identify the bioactive constituents of long chain hydrocarbons, alcohols, acids, esters, alkaloids, steroids, amino and nitro compounds etc. Hence, Gas chromatography (GC) and Mass spectroscopy (MS) associated with particular detection techniques have become a sophisticated means for analysis of various compounds [19]. The combination of the separation technique (GC)
with the best identification technique (MS) makes GC-MS an ideal technique for qualitative and quantitative analyses for volatile and semi-volatile compounds. This study therefore, aims at utilizing a rapid method, Gas Chromatography-Mass Spectrometry (GC-MS) technique, for quantitative determination of bioactive compounds in Rauwolfia vomitoria Afzel leaf and root extracts.

2. MATERIALS AND METHODS

2.1 Materials

All chemicals and reagents used were of analytical grade.

2.1.1 Plant collection and identification

The leaf and root of R. vomitoria were collected from Lambo Lasunwom village, Ikorodu, Lagos State, Nigeria in April, 2015. The plant was identified and authenticated by Prof J.D. Olowokudejo, Department of Botany, University of Lagos. A voucher specimen was deposited in the University herbarium with reference number LUH 6213.

2.1.2 Preparation of leaf and root extract of R. vomitoria

R. vomitoria leaves were washed with distilled water to free them of dust and sand. The cleaned leaves were air dried at room temperature (28 ± 2.0°C) until dry and ground to a powdery form. Roots were cleaned and cut into tiny pieces. The roots were left to dry and then ground to a coarse powdery form with Christy-Norris Laboratory Hammer Mill and kept in an air tight container until needed for use.

2.2 Extraction by Maceration

600g of the dried ground leaf and root were then extracted separately with 5L of 70- 95% ethanol for 24h. Upon complete extraction, the solvents were completely evaporated using a rotary evaporator and the concentrates were dried in a Plus 11 Gallenkamp oven at 45-50°C. Extracts were refrigerated at 4°C until needed.

2.3 Determination of Bioactive Constituents and their Structural Composition in R. vomitoria

GC-MS analysis was carried out on a GC-MS (Model: QP2010 PLUS Shimadzu, Japan) comprising AOC-20i auto sampler and chromatograph interfaced to a mass spectrophotometer (GC-MS). The instrument was equipped with a VF 5 ms fused silica capillary column of 30 m length, 0.25 mm diameter and 0.25 μm film thickness. The temperature employed were; column oven temperature 80°C, injection Temp 250°C at a pressure of 108.0 kPa, with total flow and column flow of 6.20 and 1.58 mL min⁻¹, respectively. The linear velocity was 46.3 cm sec⁻¹, and a purge flow of 3.0 mL min⁻¹. The GC program ion source and interface temperature were 200.00 and 250.00°C respectively with solvent cut time of 2.50 min. The MS program starting time was 3.00 min which ended at 30.00 min. with event time of 0.50 sec, scan speed of 1666 μL sec⁻¹, scan range 40 – 800 u and an injection volume of 1 μL of the plant extract (split ratio 10:1). The total running time of GC-MS was 30 min. The relative percentage of the extract was expressed as percentage with peak area normalization. Interpretation on mass spectrum GC-MS was conducted using the database of National Institute standard and technology (NIST) having more than 62,000 patterns [20]. The spectrum of the known compounds stored in the NIST library. The name, molecular weight, and structure of components of test materials were ascertained.

3. RESULTS AND DISCUSSION

The mass spectrum of unknown component was compared with the spectrum of the known component stored in the National Institute Standard and Technology (NIST). Interpretation of mass spectrum of GC-MS was done using database of National Institute Standard and Technology (NIST). Major components were identified with authentic standards and recorded from computerized libraries. The compound name, probability, molecular formula, molecular weight, peak area and biological activity of the test materials were ascertained. GC-MS analysis revealed the presence of 22 compounds in R. vomitoria leaf extract and 16 compounds in R. vomitoria root extract. The results of the GC-MS analysis of the leaf extract of R. vomitoria are listed in Fig. 1. The list of constituents is given in Table 1. The results of the GC-MS analysis of the root of R. vomitoria are listed in Fig. 3. The list of constituents is given in Table 2.

Five major components were identified and characterized to be seen in R. vomitoria leaf extract (Fig. 2). Likewise, five components were identified and characterized as the major bioactive compounds (Fig. 4). The mass spectrometer analyzes the compounds eluted at different times to identify the nature and structure.
of the compounds. The large compound fragments into small compounds giving rise to appearance of peaks at different m/z ratios. These mass spectra are fingerprint of that compound which can be identified from the data library.

The compounds in the leaf and root extracts of *Rauwolfia vomitoria* used in this study are, diterpene, titerpene, fatty acids, their ethyl esters, organic hydrocarbons. Others include compounds whose biological activities is yet unknown. The identified major compounds possess some important biological potential for future drug development. However, isolation and characterization of individual phytochemical constituents may proceed to discover the novel drugs and their pharmacological activities. Numerous pharmacological active compounds (tryptophan, serotonin, melatonin) have an indole nucleus. A number of compounds bearing the indole moiety have been described to own affinity toward different serotonin receptors [31]

Neurotransmitters like serotonin have structure similar to indole alkaloids and this has led to the prediction of neurological activity of indoles [43]. n-Hexadecanoic acid with undecanoic acid found in *Rauwolfia vomitoria* has also being reported by [44] in a GC-MS metabolite profiling of the methanol stem bark extract of T. pachysiphon (Apocyanacae) to be the most predominant metabolites with n-Hexadecanoic acid (27.49%), Oleic acid (14.60%) and Octadecanoic acid (6.38%). n-Hexadecanoic acid with undecanoic acid have been reported to be an acidifier, acidulant, increase aromatic acid decarboxylase activity, inhibitor of uric acid production and arachidonic acid [45]. Acidifiers are chemicals that reduce the pH of the body and are needed for food digestion in patients suffering from achlorhydria [45]. These phytocompounds will be beneficial since it increases gastric acid when ingested. Phytol, one of the major compounds detected in this experiment seems to possess antimicrobial activity. Also the interaction between other major and minor components could contribute to the antimicrobial properties. Phytol is a diterpene with antimicrobial properties, significantly against many bacterial strains [46]. Phytol has been reported to have activities such as antimicrobial, anti-cancer, anti-inflammatory, anti-diuretic, immune-stimulatory and anti-diabetic activities [47]. Phytol is a key acyclic diterpene alcohol that is a precursor for vitamin E and K1. It is used along with simple or corn syrup as a hardener in candies. It was found to possess as well as preventive and therapeutic results against arthritis [48].

Similarly, phytol and squalene also showed the various biological activities as reported for *Coldenia procumbens* Linn [2]. Research work also revealed GC-MS profiling of some other Apocyanaceae family namely Gongronema latifolium, Vincetoxicum rossicum and Marsdenia edulis species revealed biologically functional compounds with therapeutic properties including linoleic acid, phytol, neophytadiene, n-hexadecanoic acid, squalene, transfarnesol, 5-pentadecen-7-ynie, and mercaptoacetic acid [49]. Squalene, another constituent identified in GCMS is a natural triterpene known to decrease immobility time in FST [50]. [51] identified squalene have the property of antioxidant. Squalene is a hydrocarbon and a triterpene and possesses chemopreventive activity against colon carcinogenesis. It also has the property of antioxidant [52] and possesses chemopreventive activity against colon carcinogenesis [53]. Thus, it is possible that these major compounds identified in the plant from GC-MS are responsible for the antidepressant-like, antimicrobial, antioxidant, hepatoprotective, hypcholesterolemic as well as cancer preventive activities amongst others.

![Fig. 1. GC-MS chromatogram of the bioactive constituents in Rauwolfia vomitoria Leaves](image-url)
Spectra of compounds identified by GC-MS

Squalene

Phytol

n-Hexadecanoic acid

7-Tetradecanal, (Z)-

9,12,15-Octadecatrienoic acid, ethyl ester, (Z,Z)-

**Fig. 2.** Mass spectrum of major compounds of *Rauwolfia vomitoria* root extract

**Fig. 3.** GC-MS chromatogram of the bioactive constituents in *Rauwolfia vomitoria* Root
Table 1. List of compounds identified at various retention times from leaves of *Rauwolfia vomitoria* by GC-MS.

| Peak No | Component | Retention time | MW | Area % | Nature of Compound | Activity |
|---------|-----------|----------------|-----|--------|-------------------|---------|
| 1       | Undecanoic acid | 26.332         | 186 | 1.02   | Fatty acid       | Acidifier, urinary acidulant |
| 2       | 3-O-Methyl-d-glucose | 26.718         | 194 | 1.68   | Sugar moiety     |         |
| 3       | 1-Acetyl-2,2,6,6-tetramethyl-4-acetyloxy-piperidyn | 26.945         | 241 | 1.76   | Antimicrobial     | [21] |
| 4       | Hexadecanal | 27.520         | 240 | 1.45   | Alkanal           | Antimicrobial, Antioxidant [22] |
| 5       | 2-Pentadecanone, 6,10,14-trimethyl | 27.624         | 268 | 0.22   | Diterpenoids     | Antimicrobial |
| 6       | 3,7,11,15-Tetramethyl-2-hexadecen-1-ol | 27.935         | 296 | 0.25   | Terpene alcohol  | Antimicrobial |
| 7       | 3,7,11,15-Tetramethyl-2-hexadecen-1-ol | 28.241         | 296 | 0.47   | Terpene alcohol  | Antimicrobial |
| 8       | n-Hexadecanoic acid | 28.641         | 256 | 1.85   | Fatty acid       | Antibacterial and antifungal [23] |
| 9       | Hexadecanoic acid, ethyl ester | 29.083         | 284 | 0.49   | Fatty acid ester | Anti-inflammatory [24] |
| 10      | n-Hexadecanoic acid | 29.859         | 256 | 15.68  | See above        |         |
| 11      | Hexadecanoic acid, ethyl ester | 30.077         | 284 | 3.98   | Fatty acid       | Antimicrobial, Antiandrogenic, Flavor, Hemolytic |
| 12      | Phytol | 31.377         | 296 | 2.44   | Diterpene        | Antimicrobial, Anti-inflammatory, Anticancer [25] |
| 13      | Phytol | 31.992         | 296 | 16.47  | Diterpene        | Anticancer Antioxidant, Antiinflammatory, Diuretic. See above |
| 14      | 7-Tetadecenal, (Z) | 32.531         | 210 | 12.90  | Alkanal           | Larvicidal and repellent activity [26] |
| 15      | 9,12,15-Octadecatrienoic acid, ethyl ester, (Z,Z)- | 32.760         | 306 | 9.56   | Linolenic acid ester | Anticancer, Antimicrobial, Antioxidant and Hypocholesterolemic [27] |
| 16      | Octadecanoic acid, ethyl ester | 33.119         | 312 | 3.38   | Fatty acid ester | Antifungal, Antimicrobial [28] Anti-cancer [29] |
| 17      | Hexacosane | 34.596         | 366 | 0.48   | See above        |         |
| 18      | 9-Octadecenamide, (Z)- | 35.605         | 281 | 3.88   | Amide             | Anti-inflammatory activity, antibacterial activity and antioxidant [30] |
| 19      | 9-Octadecenamide, (Z)- | 35.946         | 281 | 1.64   | Amide             | See above |
Asoro et al.; EJMP, 32(6): 34-45, 2021; Article no. EJMP.68096

| Peak No | Component | Retention time | MW | Area % | Nature of Compound | Activity |
|---------|-----------|----------------|-----|--------|--------------------|----------|
| 20      | Squalene  | 41.333         | 410 | 18.69  | Triterpene         | Antibacterial, Antioxidant, Antitumor, Cancer preventive, Immunostimulant, chemo preventive, Lipoxigenase-inhibitor, Pesticide [31], [2] |
| 21      | 2-methylhexacosane | 42.030         | 380 | 1.13   | Branched alkane    | Antibacterial |
| 22      | Oxirane, 2,2-dimethyl-3-(3,7,12,16,20-pentane | 42.517         | 426 | 0.55   |                    |           |

Table 2. List of compounds identified at various retention times from root of *Rauwolfia vomitoria* by GC-MS

| Peak No | Component | Retention time | MW | Area % | Nature of Compound | Activity |
|---------|-----------|----------------|-----|--------|--------------------|----------|
| 1       | Benzoic acid, 3,4,5-trimethoxy | 26.775         | 212 | 0.58   | Aromatic compound  | Food preservative, antifungal [32] |
| 2       | n-Hexadecanoic acid | 29.728         | 256 | 15.41  | Fatty acid         | Antioxidant, Anti-inflammatory, Hypo-cholesterolemic |
| 3       | Hexadecanoic acid, ethyl ester | 30.051         | 284 | 3.09   | Palmitic acid ester | Anti-inflammatory, Anticancer, Hepatoprotective, Anti-arthritic, Anti-coronary [33] |
| 4       | cis-Vaccenic acid | 32.488         | 282 | 32.13  | Omega-7 fatty acid | Anti-cancer [34] |
| 5       | 9,12-Octadecadienoic acid (Z,Z)-6 32.717 | 32.631         | 280 | 2.84   | Polysaturated fatty acid | Anti-oxidant [34] |
| 6       | (E)-9-Octadecanoic acid ethyl ester | 32.717         | 310 | 9.83   | Fatty acid ester   | Neurotransmitter regulator [35] |
| 7       | Octadecanoic acid, ethyl ester | 33.098         | 312 | 2.13   | Fatty acid ester   | Antineuroinflammation [36] |
| 8       | 9-Tricosene | 34.451         | 322 | 1.50   | Pheromone          | Pesticide [37] |
| 9       | 8,11,14-Eicosatrienic acid | 34.702         | 306 | 1.02   | Unsaturated fatty acid | NF |
| 10      | 1-Heneicosanol | 37.185         | 312 | 1.55   | Fatty alcohol      | Anti-tuberculosis [37] |
| 11      | Cyclohexanecarbonitrile 1-(4-chlorophenyl | 40.189         | 307 | 9.45   | Cyclic Hydrocarbon |           |
| 12      | Spiro[androst-5-ene-17, 1’ Cyclobutan] 2’ one | 40.611         | 350 | 1.48   | Ketone compound    | Anti-bacterial [38] |
| 13      | 8H-Azeceno[5,4-b] indol-8-one, 5-ethylidene | 40.876         | 326 | 7.66   | Aromatic heterocyclic organic | Antidepressant properties [39] |
| 14      | Squalene | 41.282         | 410 | 2.90   | Triterpene        | Neurotransmission [40], Anti-tumor [40,41] |
| 15      | Hepta-fluorobutyrlic acid, n-tetradecyl ester | 41.611         | 289 | 4.01   | Organofluorine | NF |
| 16      | Ethyl-iso-allocholate | 41.677         | 334 | 4.43   | Steroid           | Anti-microbial [42] |
Spectra of compounds identified by GC-MS

Spectra of compounds identified by GC-MS

Spectra of compounds identified by GC-MS

Spectra of compounds identified by GC-MS

Fig. 4. Mass spectrum of major compounds of *Rauwolfia vomitoria* root extract

4. CONCLUSION

This study clearly shows that GC-MS is a powerful technique enabling fast separation and characterization of bioactive metabolites. The high sensitivity of this technique helps in characterization of active compounds in *R. vomitoria* that can be used as drugs. The findings suggest that there is an indication that *R. vomitoria* leaves and roots contain an array of
bioactive compounds that may be linked to its beneficial effects on health. Therefore it is recommended as a plant of phytopharmaceutical importance.

CONSENT

It’s not applicable.

ETHICAL APPROVAL

It’s not applicable.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Mboowa G. Genetics of Sub-Saharan African Human Population: Implications for HIV/AIDS, Tuberculosis and Malaria. Int. J. Evol. Biol. 2014;1–2. Available: https://doi.org/10.1155/2014/108291
2. Kesava RB, Usha Rani G. GC-MS analysis of volatile components in petroleum ether extracts of Coldenia procumbens Linn. International Journal of Pharmacy and Biological Sciences. 2016;7(2):241-245.
3. Ehiagbonare EJ. Regeneration of Rauwolfia vomitoria. Afr J Biotech. 2004;6(8):979-981. Available: http://www.academicjournals.org/AJB
4. Lobay D. Rauwolfia in the Treatment of Hypertension. Integrative medicine (Encinitas, Calif.). 2015;14(3):40–46.
5. Ezekwesili-Ofili JO, Okaka ANC. Herbal Medicines in African Traditional Medicine; 2019. DOI: 10.5772/intechopen.80348
6. Youmbie DDB, Dzeufiet DPD, Nkwengoua ZE, Zingue S, Mezui C, Bibi FAO, Tankeu NF, Pieme CA, Dimo T. Anti-Inflammatory andAntioxidant Effects of the Stem Bark Aqueous Extract of Rauwolfia vomitoria (Apocynaceae) In Female Wistar Rats. European Journal of Pharmaceutical and Medical Research. 2015;2(7):64-73.
7. Campbell-Tofte JI, Molgaard P, Josefsen K. Randomized and double-blinded pilot clinical study of the safety and anti-diabetic efficacy of the Rauwolfia-Citrus tea, as used in Nigerian traditional medicine. J. Ethnopharmacol. 2011;(133):402-411.
8. Bisong SA, Brown RE, Osim EE. Comparative extrapyramidal effects of Rauwolfia vomitoria, chlorpromazine and reserpine in mice. Journal Nat Med. 2013; 67:107-112. Available: https://doi.org/10.1007/s11418-012-0657-8
9. Eluwa MA, Udoaffah MT, Vulley MB, Ekanem TB, Akpantah AO, Asuquo OA, Ekong MB. Comparative study of teratogenic potentials of crude ethanolic root bark and leaf extract of Rauwolfia vomitoria (apocynaceae) on the fetal heart. North American Journal of Medical Sciences. 2010;2(12):592–595. Available: https://doi.org/10.4297/najms.2010.2592.
10. Tlhapi DB, Ramaite IDI, Van Ree T, Anokwuru CP, Orazio T-S, Hoppe HC. Isolation, chemical profile and antimalarial activities of bioactive compounds from Rauwolfia caffra Sond. Molecules. 2019; 24:39. Available: https://doi.org/10.3390/molecules24010039.
11. Aquaisua A, Mbadugha C, Bassey E, Ekong M, Ekanem T, Akpanabiatu M. Effects of Rauwolfia vomitoria on the cerebellar histology, body and brain weights of albino wistar rats. Journal of Experimental and Clinical Anatomy, Rauwolfia vomitoria. 2017;16(1):41.
12. Isaiah AM, Olawale O, Effiong EE, Idongesit NJ, Fidelis UA, Friday UU. Vitamin E supplementation with Rauwolfia vomitoria root bark extract improves hematological indices. North American Journal of Medical Sciences. 2012; 4(2):86–89. Available: https://doi.org/10.4103/1947-2714.93383
13. Tavs A Abere, Ogechi KO, Freddy OA, Gerald IE. Anticsickling and Toxicological Evaluation of the Leaves of Rauwolfia vomitoria Afzel (Apocynaceae). Journal of Science and Practice of Pharmacy. 2014; 1(1):11-15.
14. Eswani N, Abdkudus K, Nazre M, Awang-Noor AG. Medicinal plant diversity and vegetation analysis of logged over bill forest of Tekai Tembeling Forest Reserve, Jerantut, Pahang. Journal of Agricultural Science. 2010;2:189-210. Available: www.ccsenet.org/jas
15. Nandagopalan V, Johnson MG Doss. A GC-MS analysis of bioactive compounds of the methanol extract of *Hibiscus tiliaceus* Linn. Asian Journal Plant Sciences Research. 2015;5(3):6-10. Semantic Scholar. Corpus ID: 37652867. Available: https://www.semanticscholar.org/paper/GC-MS-analysis-of-bioactive-components-of-the-ofagopolan-Gritto/b5b6d5ea576b6016bc85ace2269ba5a33d886731

16. Achikanu CE, Eze-Steven PE, Ude CM, Ugwuokolie OC. Determination of the vitamin and mineral composition of common leafy vegetables in South-Eastern, Nigeria. Int. J. Curr. Microbiol. App. Sci. 2013;2(11):347-353. Available: www.ijcmas.com

17. Darvesh AS, Carroll RT, Bishayee A, Darvesh AS, Carroll RT, Bishayee A, Gritto/b5b6d5ea576b6016bc85ace2269ba5a33d886731. Dietary polyphenols as potential therapeutic agents. Expert Rev. Nutr. 2010;10:729-745. Available: https://doi.org/10.1586/ern.10.42

18. Sermakkani M, Thangapandian V. GC-MS analysis of Cassia icalica leaf methanol extract, Asian Journal of Pharmaceutical and Clinical Research. 2012;5(2):90-94.

19. Vinodh KS, Natarajan A, Devi K, Senthilkumar B. Chemical composition of aqueous leaf extract of Murraya Koenigii. Int J Pharm Biol Archiv. 2013;4:493-7.

20. Chidambaram V, Niraimathi, L, Sudha V, Lavanya, R, Vadivel V, Brindha P. Spectrophotometric, HPTLC and GC-MS studies on selected spice extracts. Int J Pharm Pharm Sci. 2015;7:184-90.

21. Li C, Hou J, Huang Z, Zhao T, Xiao L, Gao G, Hamoode C, Dong A, Assessment of 2,2,6,6-tetramethyl-4-piperidinol-based amine N-halamine-labeled silica nanoparticles as potent antibiotics for deactivating bacteria, Colloids Surf B. Biointerfaces. 2015;126:106-14. Available: https://doi.org/10.1016/j.colsurfb.2014.11.051

22. Neda MD, Biljana B, Marina S, Natasa S. Anti-microbial and antioxidant activities of *Melissa officinalis* Linn. (Lamiaceae) essential oil. Journal of Agricultural and Food Chemistry. 2004;52:2485-2489. Available: https://doi.org/10.1021/jf030698a

23. Chandrasekaran M, Senthilkumar A, Venkatesalu V. Antibacterial and antifungal efficacy of fatty acid methyl esters from leaves of Sesuvium portulacastrum L. Eur. Rev. Med. Pharmcol. Sci. 2011;15:775–780. PMID: 21780546

24. Aparna V, Dileep KV, Mandal PK, Karthe P, Sadasivan C, Haridas M. Anti-Inflammatory Property of n-Hexadecanoic Acid: Structural Evidence and Kinetic Assessment. Chemical Biology & Drug Design. 2012;80(3):434-439. DOI: 10.1111/j.1747-0285.2012.01418.x

25. Gnanavel V, Mary AS. GC-MS analysis of petroleum ether and ethanol leaf extracts from *Abrus precatorius* Linn. International Journal of Pharmacy and Biological Sciences. 2013;4(3):37-44.

26. Sivakumar R, Jebanesan A, Govindarajan M, Rajasekar P. Larvicidal and repellent activity of tetradecanoic acid against *Aedes aegypti* (Linn.) and Culex quinquefasciatus (Say.) (Diptera: Culicidae). Asian Pac. J. Trop. Med. 2011;4:706-710. Available: https://doi.org/10.4103/0974-7645(11)00178-8

27. Praveen PK, Kumaravel S, Lalitha C. Screening of antioxidant activity, total phenolics and GC-MS study of *Vitex negundo*. African Journal of Biochemistry Research. 2010;4(7):191-195. Available: http://www.academicjournals.org/AJBR

28. Gehan MA, Hanan AE, Hassan AHI, Okbah MA. Marine natural products and their potential applications as antiinfective agents. World Sciences Journal. 2009;7(7):872-880. Corpus ID: 14930735

29. Yu FR, Lian XZ, Guo HY, Mc Guire PM, Li RD, Wang R, Yu FH. Isolation and characterization of methyl esters and derivatives from Euphorbia kansui (Euphorbiaceae) and their inhibitory effects on the human SGC-7901 cells. J. Pharm. Pharmc. Sci. 2005;8:528–535.

30. Ezhilan BP, Neelamegam R. GC-MS analysis of phytocomponents in the ethanol extract of *Polygonum chinense* L. Pharmaco -gnosy Research. 2012;4(1):11-14. Available: https://doi.org/10.4103/0974-8490.91028

31. Agnei RA, Mohan VR. GC-MS analysis of bioactive compounds presents in the whole plant of *Andrographis echoides* (Linn.) Nees (Acanthaceae). European Journal of Biomedical Pharmaceutical Sciences. 2014;1(3):443-452.
32. Krátký M, Vinšová J. Antifungal activity of salicylanilides and their esters with 4-(Trifluoromethyl) benzoic Acid. Molecules. 2012;17(8):9426–9442. DOI: 10.3390/molecules17089426.

33. Adnan M, Nazim Uddin Chy M, Mostafa Kamal A, Azad MOK, Paul A, Uddin SB, Barlow JW, Faruque MO, Park CH, Cho DH. Investigation of the biological activities and characterization of bioactive constituents of Ophiirrhiza rugosa var. prostrata (D.Don) and Mondal Leaves through In Vivo, In Vitro, and In Silico Approaches. Molecules. 2019;24:1367. Available:https://doi.org/10.3390/molecules24071367

34. Kehkashan K, Sadiqa F, Aqeel A, Nida F, Muhammad N, Munawwer R, Shaheen F. GC-MS profile of antimicrobial and antioxidant fractions from Cordia-rothii roots, Pharmaceutical Biology. 2016; 54(11):2597-2605. DOI: 10.3109/13880209.2016.1172320

35. Ohta K, Miyamoto H, Yaguchi T, Nagai K, Yamamoto S, Nomura T, Nishizaki T. Stearic acid facilitates hippocampal neurotransmission by enhancing nicotinic ACh receptor responses via a PKC pathway. Molecular Brain Research. 2003; 119(1):83–89. DOI: 10.1016/S0921-8388(03)00470-9.

36. Hanley ME, Dunn DW, Abolins SR, Goulson D. Evaluation of (Z)-9-tricosene baited targets for control of the housefly (Musca domestica) in outdoor situations. Journal of Applied Entomology. 2004; 128(7):478–482. DOI: 10.1111/j.1439-0418.2004.00876.x

37. Nganso DYO, Soh D, Ndogo EO, Mala OMTG, Nyasse B. Fatty Alcohols Isolated from Prosopis africana and Evaluation of Antibacterial and Antituberculosis Activities. Journal of Diseases and Medicinal Plants. 2018;4(5):128-132. DOI: 10.11648/j.jdmp.20180405.12

38. Okeke IN, Babalola CP, Byarugaba DK, Djimde A, Osoniyi OR. Broadening Participation in the Sciences within and from Africa: Purpose, Challenges, and Prospects. CBE—Life Sciences Education. 2017;16(2):2. DOI: 10.1187/cbe.15-12-0265

39. Kochanowska-Karamyan AJ, Hamann MT. Marine indole alkaloids: potential new drug leads for the control of depression and anxiety. Chem. Rev. 2010;110:4489–4497. Available:https://doi.org/10.1021/cr900211p

40. Sasaki K, Othman MB, Ferdousi F, Yoshida M, Watanabe M, Tominaga K, Isoda H. Modulation of the neurotransmitter systems through the anti-inflammatory and antidepressant-like effects of squalene from Aurantiocyrtium sp. PLOS ONE. 2019;14(6):e0218923. DOI: 10.1371/journal.pone.0218923.

41. Amarowicz R. Squalene: A natural antioxidant?. European Journal of Lipid Science and Technology. 2009;111(5): 411–412. DOI: 10.1002/ejlt.200900102

42. Muthulakshmi A, Jothibai MR, Mohan VR. GC-MS analysis of bioactive components of Feronia elephantum Correa (Rutaceae). J. Appl. Pharmaceut. Sci. 2012;2(2): 69 –74.

43. Hazrulrizawati AH, Aizi NMR, Mashitah MY. Indole Alkaloids from Plants as Potential Leads for Antidepressant Drugs: A mini review. Front Pharmacol. 2017;8:96.

44. Uwumarongie HO, Onwukaeme DN, Igbe I. GC-MS metabolite profiling, antinoceceptive and antipyretic activities of methanol stem bark extract of Tabernaemontana pachysiphon Stapf. (Apocynaceae). Journal of Science and Practice of Pharmacy. 2018;5(1).

45. Nwakudu ON, Madubuike AJ, Achi NK. Preliminary evaluation of phytochemicals in Iresine herbistii ethanol leaf extract using gas chromatography-mass spectrophotometry analysis. J. Environ. Life Sci. 2017;2:21-28.

46. Bharathy V, Maria Sumathy B, Uthayakumari F. Determination of phytocomponents by GC-MS in leaves of Jatropha gossypifolia L. Science Research Reporter. 2012;2(3):286-290.

47. Raman BV, Sameul LA, Pardha SM, Narashimha RB, Naga VKA, Sudhakar M, Radhakrishnan TM. Antibacterial, antioxidant activity and gas chromatography–mass spectrometry analysis of Eupatorium odoratum. Asian Journal of Pharmaceutical and Clinical Research. 2012;5(2):99-106.

48. Ogunlesi M, Okiei W, Osibole AE. Analysis of the essential oil from the dried leaves of Euphorbia hirta Linn. (Euphorbiaceae), a potential medication for asthma. Afr J Biotechnol. 2009;8(24):7042-50.
49. Willie P, Uyoh EA, Aikpokpodion PO. Gas Chromatography-Mass Spectrometry (GC-MS) assay of Bio-active compounds and phytochemical analyses in three species of Apocynaceae. Pharmacognosy Journal. 2021;13(2):383-392.

50. Yildiz A, Forkey JN, McKinney SA, Ha T, Goldman YE, Selvin PR. Myosin V walks hand-over-hand: single fluorophore imaging with 1.5 nm localization. Science. 2002;300(5628):2061-2065. DOI: 10.1126/science.1084398

51. Kala SMJ, Balasubramanian T, Tresina S, Mohan VR. GC-MS determination of bioactive components of Eugenia singampattiana Bedd. Int. J. Chem. Tech. Res. 2011;3:1534-1537. Available:https://link.springer.com/articles/casredirect/1%3ACAS%3A528%3ADC%252BC3MXhtVyksrO

52. Lalitharani S, Mohan VR, Regini GS, Kalidass C. GC-MS analysis of ethanolic extract of Pothos scandens L. Leaf. J Herb Med Toxicol. 2009;3:159-60.

53. Rajeswari G, Muruga M, Mohan VR. GC-MS analysis of bioactive compounds of Hugonia mystax L. Bark (Linaceae). J Pharm Biomed Sci. 2013;29(29):818-24.

© 2021 Asoro et al.; This is an Open Access article distributed under the terms of the Creative Commons Attribution License (http://creativecommons.org/licenses/by/4.0), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Peer-review history:
The peer review history for this paper can be accessed here: https://www.sdiarticle4.com/review-history/68096