Phytochemical Screening, GC-MS Analysis and Antioxidant Activity of Three Medicinal Plants From Nigeria

Ezekwe Ahamefula S.¹*, Ordu Kenneth S.² and Oruamabo Raphael S.³

¹Department of Medical Biochemistry, Faculty of Basic Medical Sciences, Rivers State University, Nkpolu Oroworokwo, Port Harcourt, Nigeria.

²Department of Human Anatomy, Faculty of Basic Medical Sciences, Rivers State University, Nkpolu Oroworokwo, Port Harcourt, Nigeria.

³Department of Paediatrics, College of Medical Sciences, Rivers State University, Nkpolu Oroworokwo, Port Harcourt, Nigeria.

Authors’ contributions
This work was carried out in collaboration among all authors. Authors EAS and OKS designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors EAS and ORS managed the analyses of the study. Authors EAS, OKS and ORS managed the literature searches. All authors read and approved the final manuscript.

Article Information
DOI: 10.9734/AJACR/2020/v6i330161

Editor(s):
(1) Dr. Angélica Machi Lazarin, State University of Maringá, Brazil.

Reviewers:
(1) Gayatri Sanjeevkumar Kamble, ABMSP’s Anantrao Pawar College of Engineering and Research, Savitribai Phule Pune University, India.
(2) Bhagwan Singh Chandravanshi, Addis Ababa University, Ethiopia.

Complete Peer review History: http://www.sdiarticle4.com/review-history/59399

Received 20 May 2020
Accepted 27 July 2020
Published 07 August 2020

Original Research Article

ABSTRACT
Phytochemical screening, GC-MS analysis and antioxidant activity of some medicinal plants were investigated using standard methods. Leaves of Gongronema latifolium, Petrocarpus mildbraedii, and Piper guineense plants were prepared properly and used in the entire study. Results of phytochemical screening for the plants revealed the presence of tannins, saponins, alkaloids, phenolic compounds, reducing sugars amongst others at different concentrations. Results of GC-MS analysis for G.latifolium showed a total of thirty-three compounds of which Cyclohexano, 2-l(methlaminomethyl)-trans had the highest retention time, squalene had the highest molecular weight and phytol had the highest peak area. GC-MS result for P.mildbraedii revealed a total of twenty-three of which Acatamide,2,2,2-trichloro- had the highest retention time, Erucic acid had the...
highest molecular weight whereas n-Hexadecanonic acid the highest peak area. Only ten compound were observed in *P. guineense* of which Ethyl (3-hydroxyphenyl) carbamate had the highest retention time, Benzamidine, N-(4,5-dichloro-1,3-benzothiazol-2yl)-3,5-dimethoxy- had the highest molecular weight and Benzene, 1,2,3-trimethoxy-5-(2-propenyl)- has the highest peak area in *P. guineense*. The antioxidant activity of the plant extracts as assessed, followed the order *P. guineense* > *G. latifolium* > *P. mildbraedii* in comparative with ascorbic acid used as the control. The observed compounds become very important when the usefulness of these plants as medicinal plants are considered. They could as well be behind the antioxidant property of the plants in this study. This study has revealed the phytochemical screening, GC-MS analysis and antioxidant activity of some medicinal plants.

**Keywords:** Antioxidants activity; GC-MS analysis; phytochemicals; medicinal plants.

1. **INTRODUCTION**

Plants have contributed greatly to the benefit of mankind on this planet Earth [1-12]. The numerous species of plants are easily identified when either their flowers, leaves or fruits are available [13]. It has been reported that plants such as trees and shrubs abound in Nigeria [14-17]. Generally, plants are versatile materials having a wide range of applications [2,5,9,12,13,18-22]. A wide range of them have been implicated against fungi as antifungal, bacteria as antibacterial, and inflammation as anti-inflammatory agents [5,10,23-25]. Studies have shown that parts of some plants such as leaves, roots, barks, and seeds could serve as analgesic, active antipyretic laxative, amongst others [7,19,22,24-30]. Apart from serving as food materials [8,20,22,26,31-36], it has been noted that different parts of plants are appreciated for their texture, flavor, chemical and nutritional properties [18-20]. The chemical and nutritional properties of plants have been noted by different authors as being behind their therapeutic applications [2-7,9-11,14-19,23-30]. Okwu [37] noted that the chemical constituents in plants are known as phytochemicals. Duru et al. [34] and Duru et al. [38] reported that both phytochemicals and nutrients such as vitamins, minerals, proteins and amino acids; and carbohydrates found in plants are biological active in nature and are effective against disease causing pathogens. According to Morebise and Fafuns [23]; Sofowora [25]; Okiigbo and Mmeka [27]; Duru et al. [38], Duru et al. [39]; and Amadi et al. [40] plants with biologically active constituents are termed medicinal plants.

*Gongronema latifolium* Benth, *Petrocarpus mildbraedii* Harms and *Piper guineense* Schum. & Thonn are amongst the plants terms as medicinal plants with wide range of applications. *Gongronema latifolium*, known commonly as amaranth globe, is a glaborous plant that grows in tropical Africa. It is a climber with characteristics greenish yellow flowers [41]. It can inhabit secondary forest, deciduous forest, rainforest or mangrove forest. It can be propagated by seed, stem cutting or softwood [16,41-42]. The flower grows around July and August. Aside Nigeria, the plants can also be found in found in Senegal, Chad, and DR Congo [16,42-44]. In Nigeria, the Igbos, Efik/Ibibios call *G. latifolium* “utasi”, the Yorubas call it “arokeke” or “madumaro”. According to Edim et al. [43], the Akan-asantes in Ghana, the Serers in Senegal and the Kissis in Sierra leone call it “kurutu nsgurogya”, “gasub” and “ndondo-polole” respectively. *G. latifolium* have sweetly bitter in taste. It is used as in soups, salads and sauces as spice [45]. It is also used in the local brewing of beer. The stem is sometimes used as chewing stick [41-43]. The plant has wide application in folkric medicine against abnormal blood glucose levels, diarrhoea, tussive, etc [16,41-42]. It has bioactive constituents that are effective against disease causing microorganisms and certain diseases [46-48]. *P. mildbraedii* grows up to 15-25 m tall. It has fast growth tendency and cappices well. *P. mildbraedii* has superficial root system and most of them are in the top 30 cm of the soil. It has an intermittent pattern of leaf flushes. Flushes appear in the dry season when other leafy vegetables are scarce. It is a lowland green or semi-deciduous forest. It belongs to Papilionaceae family. It is known as “uruhe”, “madoobiyaá” and “ọ́ha” by Edo, Hausa and Igbo tribes of Nigeria respectively [17,49]. *P. mildbraedii* has green stify leaves, which are used for soup preparation in Nigeria. Extracts from *P. mildbraedii* are used in the treatment of headaches, pains, fever, convulsions, respiratory disorders, and as antimicrobial agents [17,50-52]. Its wood is also applied in wood construction [50,53]. *Piper guineense* respectively known as “uziza” and “iyere” by the Igbo and Yorubas
tribes of Nigeria, is an evergreen climbing shrub that produces woody stem of about 4-20 meters tall. The plant bears the capacity to support itself on other plants with the help of its adventitious roots, which are produced with stems [17, 52-53]. The plant and its pepperish, fruit product are common spices in Africa [52-54]. It could be harvested as wild or semi-cultivated. It is also cultivated for use both as a spice and also as a medicine [52, 54]. The fruit of P. guineense both fresh and dried are used as spice in flavouring soups, rice and other foods [52, 54]. The fruits and leaves are used fresh and dried as components of medicinal preparation [52, 54]. The roots are chewed and the juice swallowed as an aphrodisiac. The root is also used as chewing stick for cleaning of teeth [17, 52-54].

The production of reactive oxygen species (ROS) has been associated with normal cellular metabolism [55], and their excessive production result in progressive damage as well as ultimately cell death. Various stresses leading to excessive production of reactive oxygen species have been recognized. According to Sharma et al. [55], a cell is said to be in state of oxidative stress when the level of ROS exceeds the defense mechanisms. The defense mechanisms include group of complex antioxidative system comprising of nonenzymatic such as the major cellular redox buffers ascorbate (AsA) and glutathione (γ-glutamyl-cysteinyl-glycine, GSH) as well as tocopherol, carotenoids, and phenolic compounds; and enzymatic components such as superoxide dismutase (SOD), catalase (CAT), guaiacol peroxidase (GPX), enzymes of ascorbate-glutathione (AsA-GSH) cycle ascorbate peroxidase (APX), monodehydroascorbate reductase (MDHAR), dehydroascorbate reductase (DHAR), and glutathione reductase (GR) [56-57]. They scavenge and as well combat the activities of ROS and prevent them from being excessive as to pose a threat to cells by causing peroxidation of lipids, oxidation of proteins, damage to nucleic acids, enzyme inhibition, activation of programmed cell death (PCD) pathway and ultimately leading to death of the cells [55]. The active oxygen species (AOS) such as singlet oxygen, super oxide radical anion, hydrogen peroxide associated with oxidative stress has been linked to play cardinal role as free and non-free radicals in the etiology of several diseases like arthritis, cancer, atherosclerosis, diabetes, cardio- and cerebrovascular diseases, tumors, epilepsy, mutagenesis, carcinogenesis, arteriosclerosis, Alzheimer’s disease, tissue injury [58-60].

Due to the linkage of certain disease conditions with oxidative stress and coupled with the facts that natural compounds found in plants or their synthetic forms are the basis of modern pharmacopeia [25, 61-62], there is a renewed interest on medicinal plants with antioxidants activity. The present study investigated the phytochemical constituents of G.latifolium, P. guineense and P.mildbraedii and P.guineense with advanced method of GC-MS and their antioxidant activity.

2. MATERIALS AND METHODS

2.1 Sample Collection and Identification

The samples of G. latifolium, P.mildbraedii, and P.guineense used were collected from Imo State University school farm and were identified in the Department of Plant Science and Biotechnology of the same institution by Botanist in the Department. The leaves of interest were collected, properly cleaned, shade dried and coarsely powdered for further usage.

2.2 Extracts Preparation

Ten grams (10 g) of each sample was extracted by maceration in 50 ml of water for 3 days with frequent agitation at a speed of 280 rpm at 28°C in dark. Between extractions, the samples were centrifuged for 10 min with 2000 rpm. The combined supernatants were collected, filtered through Whatman No. 1 filter paper and concentrated in vacuum. They were kept in a vacuum desiccator for complete removal of solvent. The yield extracts were thus used for some of phytochemical screening, GC-MS analysis and assessment of antioxidant activity.

2.3 Qualitative Phytochemical Determinations

2.3.1 Test for tannins

To 1 mL each of the plant extract, equal volume of bromine water was added. The formation of a greenish to red precipitate was taken as the presence of tannins,

2.3.2 Test for saponins

1 mL of each extract was boiled with 5 mL of distilled water for 5 min. and decanted while hot.
4 mL of distilled water was added to 1 mL each of the filtrate before they were shaken vigorously for observation of stable froth on standing.

2.3.3 Test for flavonoids

0.5 g of each selected plant extract were added in a test tube and 10 ml of distill water, 5 ml of dilute ammonia solution were added to a portion of the aqueous filtrate of each plant extract followed by addition of 1 ml concentrated H_2SO_4. Indication of yellow color shows the presence of flavonoid in each extract.

2.3.4 Test for alkaloids

1 mL each of the plant extract was shaken with 5 mL of 2% HCl on a steam bath and then filtered. To 1 mL each of the filtrate, Wagner’s reagent (iodine in potassium –iodide solution) was added and reddish brown precipitates was observed for positive result.

2.3.5 Test for steroids

The amount of 0.5 g each of the extract was dissolved in 10 mL anhydrous chloroform and filtered. The filtrate was divided into two equal portions for the following tests. The first portion of the solution above was mixed with one mL of acetic anhydride followed by the addition of 1 mL of concentrated sulphuric acid down the side of the test tube to form a layer underneath. The test tube was observed for green colouration as indicative of steroids.

2.3.6 Test for terpenoids

1 g of selected plant samples was taken in a test tube, then poured 10 mL of methanol in it, shaken well and filtered to take 5 ml extract of plant sample. Then 2 mL of chloroform were mixed in extract of selected plant samples and 3 mL of sulphuric acid were added in selected sample extracts. Formation of reddish brown color indicates the presence of terpenoids in the selected plants.

2.3.7 Cardiac glycosides

1 mL each of the plant extract was dissolved in in 2 mL of chloroform in a test tube. 1 mL conc.H_2SO_4 was carefully added to each of the test tubes through their sides and they were observed for a red or reddish brown colouration at the interphase, which indicates positive result.

2.3.8 Test for phlobatannins

One percent aqueous hydrochloric acid was added was added to each of the plant extract in a test tube (about 2 mL), and then boiled with the help of Hot plate stirrer. Formation of red coloured precipitate confirmed a positive result.

2.3.9 Test for phenolic compounds

To 2 mL of each plant extract, 1% FeCl_3 was added and observation was made for blue, violet, purple, green or red-brown colour.

2.3.10 Test for proteins

Five drops of 1% hydrated copper sulphate was added to 2 mL each of the sample extract in a test tubes. Two mL of 40% NaOH was also added, and the test tubes were shaken vigorously to mix the contents and presence of purple colouration indicated the presence of proteins.

2.3.11 Test for reducing sugars

One mL of ethanol was mixed with 2 mL each of the plant extract, after which 1 mL each of Fehling solution A and B were added to the test tubes. The test tubes were heated to boiling while observation was made for presence of reddish brown colouration which indicates positive results.

2.3.12 Test for anthraquinones

One gram of the ground plant was placed in a dry test tube and 20 mL of chloroform was added. This was heated in steam bath for 5 min. The extract was filtered while hot and allowed to cool. To the filtrate was added with an equal volume of 10% ammonia solution. This was shaken and the upper aqueous layer was observed for bright pink colouration, which for the presence of anthraquinones. This was repeated with all the plant samples.

2.4 GC-MS Analysis of the Extracts

GC-MS analysis of the aqueous extracts was carried out using AOC-20i auto sampler and gas chromatograph interface to a mass spectrometer (GC-MS) instrument. Employing the following conditions; column Elite-1 fused silica capillary column (30 mm×0.25 mm ID×1μM df, composed of 100% Dimethyl poly siloxane), operating in electron impact mode at 70 eV; helium (99.999%) was used as carrier gas at a constant flow of 1 ml/ min, and an injection volume of
3.2.5 Determination of Antioxidant Activity

Phytochemicals are chemicals that protect cells and various macromolecules from oxidative processes and environmental hazards. However, it was in recent times that it was clearly observed that phytochemicals play important roles in human health, when they are appropriately taken in significant concentrations [65-66]. Phytochemicals become important when their roles or functions are considered in humans [40]. The concentration of tannins was high in G. latifolium while P. guineense and P. mildbraedii had moderate concentrations of tannins. According to Saxena et al. [64] and Serrano et al. [67], many health benefits have been recognised for the intake of tannins. Epidemiological associations have linked tannins to decreased frequency of chronic diseases [64]. Tannin containing plant extracts are used as astringents, as diuretics, against diarrhoea, stomach and duodenal tumours. Tannins are also potent antiseptic, antioxidant, anti-inflammatory and haemostatic pharmaceuticals [68-69]. Saponins are group of secondary metabolites known for their stable foam or foaming property in aqueous solutions [70]. Extensive research studies have revealed the membrane-permeabilising, immunostimulant, hypocholesterolaemic and anticarcinogenic properties of saponins. Saponins have also been found to be effective against protozoans, molluscs, fungi and viruses [64]. Saponin concentration was high in G. latifolium while P. guineense in the present study but was moderate in P. mildbraedii. Flavonoids are ubiquitous in nature and are known to have numerous derivatives [64]. They protect carbohydrates, proteins, lipids and DNA macromolecules from oxidative processes and protect the biological systems from harmful effects of such oxidative processes [64]. Flavonoids were moderately present in G. latifolium and P. guineense, but absent in P. mildbraedii in this study. Alkaloids offer significant protection and ensure survival of plants. They exhibit antihypertensive, antiarrhythmic, antimalarial, and anticancer effect [64]. Some alkaloids have stimulant property as caffeine and nicotine, morphine are used as the analgesic and quinine as the antimalarial drug [71]. Alkaloid was high in G. latifolium, but absent in P. mildbraedii. Steroids were only observed in P. mildbraedii. The roles of steroids in the formation if steroid hormones have long been noted. Terpenoids have anticarcinogenic, antimalarial, anti-ulcer, hepaticidal, antimicrobial or diuretic activity. It is on record that drugs have been manufactured from some terpenoid derivatives [72-73]. Cardiac glycosides at low concentrations are highly beneficial to the heart [70]. Terpenoids and cardiac glycosides were moderately observed in

2.5 Determination of Antioxidant Activity

2.5.1 DPPH (1, 1-Dipheny I-2-picrylhydrazyl) radical scavenging assay

The free radical scavenging activity was measured by DPPH assay method. Four mg of DPPH (0.1 mM) was dissolved in 100 mL of distill water to obtain working solution. One mL of each extract was mixed separately with 2.0 mL of 0.1 mM DPPH followed by 30 min incubation in dark. The reduction of the DPPH free radical was measured by taking the absorbance at 517 nm [63]. Colour of DPPH was reduced from purple to yellow. The antioxidant activity of each extracts was evaluated by calculating the inhibition % of free radical formation using the formula:

\[
\% \text{ inhibition} = \left[\frac{(A-A_1)}{A}\right] \times 100; \quad A= \text{ absorbance of the blank (DPPH)}; \quad A_1= \text{ absorbance of the extract (DPPH+ extract)} 
\]

3. RESULTS AND DISCUSSION

The phytochemical screening of G. latifolium, P. mildbraedii and P. guineense represented in Table 1 shows the presence of tannins, saponins, flavonoids, alkaloids, steroids, terpenoids, cardiac glycosides, phlobactanins, phenolic compounds, proteins, reducing sugars and anthoquinones at different concentrations in the plants. Saxena et al. [64] noted that phytochemicals are chemicals that protect cells in plants against pathogenic attack and environmental hazards. However, it was in
P. guineense and P. mildbraedii in the present study. The anti-inflammatory, analgesic and wound healing properties of phlobatannins have been reported [70,74]. Phlobatannins were moderately concentrated in G. latifolium and P. mildbraedii. Phenolic compounds have been implicated against increased bire secretion, reduction of lipid levels and blood cholesterol. They also exhibit antimicrobial activity against some strains of bacteria. G. latifolium had high concentration of phenolic compounds while P. mildbraedii and P. guineense had moderate concentrations of the compounds. Proteins are reported sources of amino acids, which invariably repairs and build worn-out tissues. Proteins occurred moderately in G. latifolium but were highly concentrated in P. mildbraedii and P. guineense. Reducing sugars and anthroquinones occurred moderately in P. mildbraedii and P. guineense while G. latifolium had high concentration of anthroquinones.

Result of GC-MS analysis of G. latifolium showing retention time, molecular formula, molecular weight and peak area as presented on Table 2 revealed the presence of 33 constituents showing their retention time in minutes, molecular formula, molecular weight and peak areas in percentage. Among the compounds observed in G. latifolium were 4-Aminobutanoic acid, Ethylene oxide, Methyltetradecanoate, M’N-Diacetylenediamine, Methy1 8-methyleneoctanoate, Urea, butyl, Teichloroacetic, undec-10-enylester, 8-none-2-one, 3,8-Dioxatricyclo (5.1.0.0(2,4) octane, 4-ethyl, Undecanoic acid, 10-methyl, methylester, 7-Hexadecanoic acid, methyl ester (Z), Hexadecanoic, methyl ester, 1-Tridecanamine, n-Hexadecanoic acid, Cyclo pentanemethanamine 2-amino, Acetaldehyde, 9,12-Octadecanoic acid, methyl ester, 15-Octadecanoic acid, methyl ester, Phytol, Methyl sterate, Cis-vaccenic acid, Di-Phenylephrine, 9-Octadecanoic acid (Z), methyl ester, 9-Hexadecanoic acid, 9-Oxabicyclo[6.1.0]nonane, cis, 13-Docosenic acid, methyl ester, Erucic acid, 6-Nitroudec-5-ene, Cis-9-Hexadecanoic acid, Squalene, 6-Nitronudec-5-ene, 2-Methylaminomethyl-1,3-dioxolane, and Cyclohexano, 2-l(methylaminomethyl)-trans. Cyclohexano, 2-l(methylaminomethyl)-trans had the highest retention time, squalene had the highest molecular weight while phytol had the highest peak area in G. latifolium as observed in the present study.

Result of GC-MS analysis of P. mildbraedii showing retention time, molecular formula, molecular weight and peak area as presented on Table 3 revealed the presence of 23 constituents showing their retention time in minutes, molecular formula, molecular weight and peak areas in percentage. Among the compounds observed in P. mildbraedii were Ethylene, 2,4 (1H, 3H)-Pyrimidinedione, dihydro-5-hydroxy, N-(3-Methy1butyl) acetamide, Bicyclo[3,1,1]heptane-2,6,6-trimethyl-[1R-(1α, 2α, 5α], 1-Heptadecanamine, 2-Butene ozonide, Cyclopentanol, 2-(aminomethyl)-trans, Hexadecanoic acid, methyl ester, 1H-Azaine, octahydro, n-Hexadecanoic acid, n-Hexadecanoic acid, Heptane, 1-Heptadecanamine,N,N’-Bis (3-aminopropl)ethylenediamine, Cis-Vaccenic acid, Octadecanoic acid, Acetamide 2,2,2-trichloro-, Cis-11-Eicosenoic acid, Di(pent-4-4nyl)amine, Cyanoacetyurea, Erucic acid, 3-fluoroamphetamine, Benzoethanamine,4-fluoro, and Acatamde,2,2,2-trichloro-. Acatamde,2,2,2-trichloro- had the highest retention time, Erucic acid had the highest molecular weight whereas n-Hexadecanoic acid the highest peak area in P. mildbraedii.

**Table 1. Phytochemical screening of G. latifolium, P. mildbraedii, and P. guineense**

| Phytochemical       | G. latifolium | P. mildbraedii | P. guineense |
|---------------------|---------------|---------------|-------------|
| Tannins             | ++            | +             | +           |
| Saponins            | ++            | +             | ++          |
| Flavonoids          | +             | -             | +           |
| Alkaloids           | +             | -             | ++          |
| Steroids            | -             | +             | -           |
| Terpenoids          | -             | +             | +           |
| Cardiac glycosides  | -             | +             | +           |
| Phlobatannins       | +             | +             | -           |
| Phenolic compounds  | ++            | +             | +           |
| Proteins            | +             | ++            | +           |
| Reducing sugars     | -             | +             | +           |
| Anthroquinones      | ++            | +             | +           |

++: present in high concentration; +: moderate concentration; -: absent
| S/N | RT   | Name of compound                                           | Molecular formula | Molecular weight | Peak area % |
|-----|------|-----------------------------------------------------------|-------------------|------------------|-------------|
| 1   | 2.493| 4-Aminobutanoic acid                                      | C₆H₁₂NO₂          | 103              | 1.37        |
| 2   | 4.312| Ethylene oxide                                            | C₆H₁₂N₂O          | 44               | 0.74        |
| 3   | 8.227| Methyltetradecanoate                                      | C₁₂H₂₄NO₂         | 242              | 2.79        |
| 4   | 8.548| M’N-Diacetylenediamine                                    | C₁₂H₁₆N₂O₂        | 144              | 1.51        |
| 5   | 8.741| Methy l 8-methyldecanoate                                 | C₁₂H₂₆O₃          | 200              | 1.00        |
| 6   | 8.944| Urea, butyl                                              | C₆H₁₄N₂O          | 116              | 0.62        |
| 7   | 9.019| Teichloroacet ic, undec-10- enylester                    | C₁₃H₂₂Cl₂O₂       | 314              | 1.83        |
| 8   | 9.062| 8-non e-2-one                                            | C₈H₁₆O            | 140              | 1.93        |
| 9   | 9.313| 3,8-Diox atricylo (5.1.0.0(2.4) octane, 4-ethyl         | C₁₅H₂₆NO₂         | 138              | 0.77        |
| 10  | 9.308| Undecanoic acid, 10-methyl, methyl est er                 | C₁₃H₂₄O₆          | 214              | 0.66        |
| 11  | 9.489| 7-Hexadecanoic acid, methyl est er (Z)                    | C₁₇H₃₄O₂          | 268              | 2.24        |
| 12  | 9.663| Hexadecanoic, methyl est er                               | C₁₇H₃₄O₂          | 268              | 2.24        |
| 13  | 9.800| 1- Tridecamine                                           | C₁₃H₂₆N           | 194              | 1.20        |
| 14  | 9.944| n-Hexadecanoic acid                                      | C₁₅H₃₂O₂          | 256              | 6.96        |
| 15  | 10.105| Cyclopanemethanamine 2- amin o                           | C₁₈H₃₄N₂          | 114              | 1.51        |
| 16  | 10.286| Acetaldehyde                                            | C₆H₁₂O            | 44               | 0.52        |
| 17  | 10.720| 9,12-Octadecanoic acid, methyl est er                     | C₁₉H₃₄O₂          | 294              | 3.94        |
| 18  | 10.762| 15-Octadecanoic acid, methyl est er                      | C₁₉H₃₆O₂          | 296              | 3.98        |
| 19  | 10.832| Phytol                                                  | C₁₅H₄₀O           | 296              | 17.44       |
| 20  | 10.913| Methyl st er                                             | C₁₉H₃₈O₂          | 298              | 2.62        |
| 21  | 11.051| Cis-vaccenic acid                                         | C₁₈H₃₂O₂          | 282              | 5.07        |
| 22  | 11.051| Di-Phenylephrine                                         | C₁₈H₃₄NO₂         | 167              | 1.02        |
| 23  | 11.950| 9-Octodecanoic acid (Z), methyl est er                    | C₁₉H₃₈O₂          | 296              | 5.09        |
| 24  | 12.217| 9-Hexadecanoic acid                                      | C₁₀H₂₀O           | 254              | 1.72        |
| 25  | 12.913| 9-Oxabicyclo[6.1.0]nonane, cis                          | C₁₀H₂₀O           | 126              | 1.18        |
| 26  | 13.036| 13-Docosonic acid, methyl est er                          | C₁₂H₂₄O₂          | 352              | 4.38        |
| 27  | 13.276| Erucic acid                                              | C₁₂H₂₄O₂          | 338              | 1.34        |
| 28  | 13.966| 6-Nitroudec-5-ene                                        | C₁₁H₁₇NO₂         | 199              | 1.43        |
| 29  | 14.624| Cis-9-Hexadecanoic acid                                  | C₁₅H₃₀O₂          | 254              | 0.65        |
| 30  | 14.624| Squalene                                                | C₁₅H₃₀O₂          | 410              | 0.61        |
| 31  | 15.057| 6-Nitroudec-5-ene                                        | C₁₁H₁₇NO₂         | 199              | 1.43        |
| 32  | 18.047| 2-Methylaminomethyl-1,3-dioxolane                       | C₁₀H₁₁NO₂         | 117              | 0.59        |
| 33  | 19.144| Cyclohexano, 2-(methylaminomethyl)-trans                | C₁₁H₂₁NO₂         | 143              | 1.56        |

RT = Retention time

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Table 3. Result of GC-MS analysis of *P. mildbraedii* showing retention time, molecular formula, molecular weight and peak area

| S/N | RT   | Name of compound                                           | Molecular formula | Molecular weight | Peak area % |
|-----|------|------------------------------------------------------------|-------------------|------------------|-------------|
| 1   | 2.374| Ethylene                                                   | C2H4O             | 44               | 0.79        |
| 2   | 4.681| 2,4 (1H, 3H) Pyrimidinedione, dihydro-5-hydroxy             | C2H6N2O3          | 130              | 1.17        |
| 3   | 8.537| (N-(3-Methylbutyl) acetamide                               | C2H10NO           | 129              | 2.96        |
| 4   | 9.013| Bicyclo[3,1,1]heptane-2,6,6-trimethyl-[1R-(1α, 2α, 5α]-    | C10H12            | 138              | 4.67        |
| 5   | 9.051| 1-Heptadecanamine                                         | C17H32            | 255              | 1.05        |
| 6   | 9.211| 2-Butene ozonide                                          | C2H4O3            | 104              | 1.71        |
| 7   | 9.307| Cyclopentanol, 2-(aminomethyl)-trans-                      | C6H12NO           | 115              | 2.14        |
| 8   | 9.628| Hexadecanoic acid, methyl ester                           | C17H32O2          | 270              | 2.87        |
| 9   | 9.783| 1H-Azoline, octahydr                                       | C6H13N            | 127              | 3.65        |
| 10  | 9.933| n-Hexadecanoic acid                                       | C16H33O2          | 256              | 24.08       |
| 11  | 10.752| Heptane                                                   | C7H14O            | 114              | 1.44        |
| 12  | 10.816| 1-Heptadecanamine                                        | C17H32N           | 255              | 1.58        |
| 13  | 10.912| N,N'-Bis (3-aminopropyl)ethyleneidiamine                  | C12H15N2          | 145              | 0.85        |
| 14  | 11.040| Cis-Vaccenic acid                                         | C18H22O2          | 282              | 18.25       |
| 15  | 11.174| Octadecanoic acid                                         | C18H32O2          | 284              | 10.33       |
| 16  | 11.944| Acetamide 2,2,2-trichloro-                                | C12H22ClNO        | 161              | 0.87        |
| 17  | 12.201| Cis-11-Eicosenoic acid                                    | C20H34O2          | 310              | 7.59        |
| 18  | 12.324| Di(pent-4-ynyl)amine                                     | C16H16N           | 153              | 2.41        |
| 19  | 13.030| Cyanoacetylene                                            | C6H10NO2          | 127              | 1.38        |
| 20  | 13.271| Erucic acid                                               | C22H34O2          | 338              | 7.20        |
| 21  | 15.148| 3-fluoroanphetamine                                       | C9H15FN           | 153              | 1.06        |
| 22  | 19.117| Benzeneethanamine,4-fluoro                                | C9H15FNO2         | 185              | 0.97        |
| 23  | 21.080| Acatamide,2,2,2-trichloro-                                | C12H22ClNO        | 161              | 0.98        |

RT= Retention time

Table 4. Result of GC-MS analysis of *P. guineense* showing retention time, molecular formula, molecular weight and peak area

| S/N | RT   | Name of compound                                           | Molecular formula | Molecular weight | Peak area % |
|-----|------|------------------------------------------------------------|-------------------|------------------|-------------|
| 1   | 5.62 | 4,9-decadienoic acid, 2-nitro-, ethyl ester               | C12H19O4          | 241              | 2.64        |
| 2   | 5.90 | (+)-2-carene, 4-alpha,-isopropenyl                         | C13H30            | 176              | 3.21        |
| 3   | 6.93 | Benzene, 1,2,3-trimethoxy-5-(2-propenyl)-                 | C13H18O3          | 208              | 56.27       |
| 4   | 9.63 | Hexadecanoic acid, methyl ester                           | C17H34O2          | 270              | 3.19        |
| 5   | 9.96 | Hexadecanoic acid                                         | C16H32O2          | 256              | 7.50        |
| 6   | 10.68| Cyclotetradecane                                         | C14H28            | 196              | 6.87        |
| 7   | 10.83| Phytol                                                    | C20H40O           | 296              | 11.27       |
| 8   | 11.05| Cis-vaccenic acid                                         | C18H34O2          | 282              | 4.92        |
| 9   | 14.50| Benzinamide, N-(4,5-dichloro-1,3-benzothiazol-2-yl)-3,5 dimethoxy- | C16H12Cl2N2O35  | 382              | 2.16        |
| 10  | 14.65| Ethyl (3-hydroxyphenyl) carbamate                         | C8H11NO3          | 181              | 1.97        |

RT= Retention time

Result of GC-MS analysis of *P. guineense* showing retention time, molecular formula, molecular weight and peak area as presented on Table 4 revealed the presence of 10 constituents showing their retention time in min., molecular formula, molecular weight and peak areas in percentage. Among the compounds observed in *P. guineense* were 4,9-decadienoic acid, 2-nitro-, ethyl ester, (+)-2-carene, 4-alpha,-isopropenyl, Benzene, 1,2,3-trimethoxy-5-(2-propenyl)-, Hexadecanoic acid, methyl ester, Hexadecanoic acid, Cyclotetradecane, Phytol, Cis-vaccenic acid, Benzinamide, N-(4,5-dichloro-1,3-benzothiazol-2-yl)-3,5- dimethoxy- and Ethyl (3-hydroxyphenyl) carbamate. Ethyl (3-hydroxyphenyl) carbamate had the highest retention time, Benzinamide, N-(4,5-dichloro-1,3-benzothiazol-2-yl)-3,5-dimethoxy- had the highest molecular weight and Benzene, 1,2,3-trimethoxy-5-(2-propenyl)- has the highest peak area in *P. guineense*. 
The observed compounds revealed by GC-MS in the plants and those of phytochemical screening become important when their functions and contributions in nature are considered.

The antioxidant activity of the G.latifolium, P.mildbraedii and P.guineense. as presented in Fig. 1 revealed increased % inhibition of 34%, 43% and 51% respectively for 5 mg/L, 10 mg/L and 15 mg/L concentrations of G.latifolium. % inhibition for P. mildbraedii were observed to be 32.89%, 34.13% and 50.78% respectively for concentrations of 5 mg/L, 10 mg/L and 15 mg/L of P. mildbraedii. The % inhibition activity of 32.8%, 40.8% and 54.27% respectively with concentrations of 5 mg/L, 10 mg/L and 15 mg/L were observed for P.guineense. Generally, the antioxidant activity of the plants increased with increase in concentration of the extracts, and they followed the order P.guineense > G.latifolium > P. mildbraedii in comparative with ascorbic acid used as the control.

4. CONCLUSION

G.latifolium is enriched with enormous compound, followed by P.mildbraedii and then P.guineense. Their antioxidant capacity followed the order P.guineense > G.latifolium > P. mildbraedii. These compounds as revealed by GC-MS analysis could no doubt be behind the observed antioxidant activity of the plants and as well as behind their efficacies as medicinal plants. This study has revealed the phytochemical screening, GC-MS analysis and antioxidant activity of some medicinal plants.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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