

Tropical Journal of Phytochemistry & Pharmaceutical SciencesAvailable online at <https://www.tjpps.org>**Original Research Article****Quantitative Phytochemical Analysis and Antioxidant Evaluation, Fractionation and Structural Elucidation of Oat Meal Extract**Onyeloni O Sunday^{1*}, Uzochukwu C Ikemefuna², Innocentmary Ejiofor²¹Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Delta State University, Abraka, Delta State, Nigeria²Department of Pharmaceutical/Medicinal Chemistry, Faculty of Pharmaceutical Sciences, Nnamdi Azikiwe University, Awka, Nigeria**ABSTRACT**

Avena sativa (Fam. Poaceae) popularly called oat is a cereal crop regarded as a functional food because it has medicinal uses in addition to its food ingredient like carbohydrates, proteins, lipids and fibres. It is reported to have anti-inflammatory, antimicrobial, antioxidant, anti-diabetic, anti-obesity, central nervous effect. The aim of this study is the quantitative phytochemical analysis, antioxidant evaluation, column chromatographic fractionation, TLC purification and isolation of fractions and GC-MS structural determination of isolated compounds. The aim of this study therefore is the bioassay guided extraction, fractionation purification to separate useful constituents and subsequent GC-MS structural elucidation of compounds present. The quantitative determination of the methanol extract was done using UV spectrophotometer for flavonoids, phenolics, alkaloids, tannins and saponins. The antioxidant activity was carried out using DPPH scavenging assay with vitamin E as standard. The Oat meal extract was subjected to column chromatography to separate it into its component fraction. The eluted fractions were further analyzed using TLC. GC-MS analysis of fraction was performed using a Perkin Elmer GC Clarus 800 system. The result shows that methanol extract of Oat meal is a rich source of flavonoids, phenolics, alkaloids, tannins and saponins (198.85, 60.51, 60.35, 20.30 and 60.15% respectively). Oat meal extract also show significant antioxidant activity (68%) which may be attributed to the synergistic effect of phytochemical content. Lipophilic extracts were mainly observed in the chromatographic separation and purification. The GC-MS analysis reviewed the presence of 15 bioactive compounds.

Keywords: Antioxidant, Phytochemical, Elucidation, Chromatography

Received 31 December 2024

Revised 07 January 2025

Accepted 29 April 2025

Published online 01 May 2025

Copyright: © 2025 Onyeloni *et al.* This is an open-access article distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

Introduction

Phytochemicals are essential biomolecules produced by plants through enzyme catalyzed biosynthetic pathways used for its protection against bacteria, fungi, viral infections and consumption by pests¹. They also have therapeutic values because of their contents of polyphenols, flavonoids, isoflavonoids, anthocyanidins, phytoestrogens, terpenoids, carotenoids, limonoids, phytosterols, glucosinolates, alkaloids and fibers². These molecules are found in leaves, barks, roots, stem and flowers of medicinal plants, which form the basis of their use in ethnomedicine and providing a template for the discovery of lead compounds which may have potentials to be used as drugs³.

Generated in the body as a result of environmental pollution and stressful conditions occasion by worsening economic conditions, especially in developing countries are reactive oxygen free radicals⁴. These free radicals have been implicated in the etiology of many degenerating and debilitating diseases like cancer, diabetes, mental health disorders, DNA damage and premature ageing⁵. These free radicals which are generated through oxidative stress can cause tissue damage when accumulated in the body because of their ability to react with electron donors which damage their vital macromolecules such as protein, lipid and DNA. These free radicals include hydrogen peroxide (H₂O₂), superoxide anion (O₂⁻), hydroxyl radical (-OH), nitric oxide (NO), organic hydroperoxide (ROOH)⁶.

Oat (*Avena sativa*) which is a functional food is reported to have anti-inflammatory, anti-diabetic, immune-modulatory, anti-pruritic, antithrombotic, antimicrobial with rich sources of phytochemicals⁷. Oat meal is a potential source of antioxidant which are free radical scavengers which remediate the damaging effect of oxidative stress. The aim of the study is the quantitative phytochemical evaluation and antioxidant determination, fractionation and structural elucidation of Oat meal extract that may enhance the development of lead compounds. For eventual drug discovery compounds from Oat meal, a functional food with both nutritional and medicinal uses will most likely possess little or no toxicity with very high therapeutic index, however, there is need for further investigation of compounds isolated from Oat meal especially the three whose medicinal uses have not been reported but occur in nature. This study provides a template for further work to be

*Corresponding author. Email: onyeloniso@gmail.com

Tel: +2348062725284

Citation: Onyeloni SO, Ikemefuna CU, Ejiofor I. Quantitative Phytochemical Analysis and Antioxidant Evaluation, Fractionation and Structural Elucidation of Oat Meal Extract. Trop J Phytochem Pharm Sci 2025; 4(4) 174 – 180. <http://www.doi.org/10.26538/tjpps/v4i4.5>

Official Journal of Natural Product Research Group, Faculty of Pharmacy, University of Benin, Benin City, Nigeria.

intensified on the potential therapeutic uses of food because of the occurrence of side effects or adverse drug reactions are improbable.

Materials and Methods

Sample Collection

Oat milk (Quaker Oat™) was purchased in Abraka, Delta State, Nigeria on the 30th of September, 2023.

Sample Extraction

About 900 g of Quaker Oat™ was dissolved in 70% methanol and filtered under suction. The residue was further extracted in methanol using stoppered container and allowed to stand for at least 72 hours with intermittent shaking. The extract was then filtered under suction and lyophilized using rotary evaporator and stored at 4°C for further analysis

Qualitative Phytochemical Analysis of Oat Meal Extract

Flavonoids: About 3ml of the Oat meal extract was mixed with few drops of 5M NaOH and an intense yellow color was formed. A few drops of 70% HCl was then added and the yellow color disappeared. The formation and disappearance of deep yellow color indicates the presence of flavonoids⁸.

Phenolics: The presence of phenolic is determined by mixing 2ml of filtered sample and 2 mL of 5% aqueous FeCl₃. The appearance of blue color indicates the presence of phenols⁸.

Alkaloids: The presence of alkaloids in the sample was determined by dissolving 2 mL of each of the filtered sample in dilute hydrochloric acid and filtered. The solution was treated with Dragendorff's reagent (solution of potassium bismuth iodide). The formation of a red precipitate shows the presence of alkaloids⁸.

Tannins: Tannins determination was done according to the method described by Gown Rajkumar *et al.*⁹. 2 mL of the filtered sample of Oat meal was added to 10% alcoholic FeCl₃ in a test tube and mixed thoroughly. The formation of a black/brown blue indicate the presence of tannins.

Saponins: About 2 mL of filtered sample was added to 4 mL of distilled water in a test tube mixed well and shaken vigorously. The presence of foam that persist for 10 minutes shows the presence of saponins⁸.

Quantitative Phytochemical Analysis of Oat Meal Extract

Determination of Flavonoids

The quantitative determination of flavonoids was done according to the method described by Madhu *et al.*¹⁰. Total flavonoids was determined by aluminium chloride method using catechine (flavan-3-ol) as a standard. 1 mL of Oat meal extract was added to 4 mL of water in a volumetric flask and mixed. 0.3 mL of 5% sodium nitrite, 0.3 mL of 10% aluminium chloride was then added after 5 minutes and incubated for 6 minutes at room temperature. 2 mL of 1M sodium hydroxide was then added to the reaction mixture and the volume made up immediately to 10ml with distilled water. The UV absorbance of the reaction mixture was measured at 510nm against a blank. Results were expressed in catechine equivalent (mg catechine/g dried extract).

Determination of Total Phenolics

The total phenolics content of Oat meal was determined using Folin Ciocalteu reagent (FCR), different concentration of the sample (20% w/v, 10% w/v, 5% w/v, 2% w/v, 1% w/v) was prepared and added into 5 different test tubes. 5ml distilled water and 0.5 mL of Folin Ciocalteu were added to each of the test tubes and mixed thoroughly. The test tubes were allowed to stand for 5 minutes at room temperature and a 0.5 mL volume of 10% sodium carbonate was added and the volume made up to 10 mL with distilled water and mixed thoroughly. The different test tubes were incubated at 50°C for 2 hours and absorbance measured at 765nm against distilled water as blank. A calibration curve was then plotted using catechol solution as standard and the total

phenolics of the extract was expressed in terms of milligrams of catechol per gram of dry weight and the standard graph¹⁰.

Determination of Alkaloids

The quantitative determination of alkaloids in Oat meal was done as described by Madhu *et al.*¹⁰. 5 mL of pH 4.7 phosphate buffer was added to 1 mL of sample extract in a test tube. 5 mL BCG was added and the mixture shaken with 4 mL of chloroform. The mixture was added to 100 mL volumetric flask and further diluted with chloroform. The absorbance of the chloroform complex was measured at 470nm against distilled water as blank with atropine used as standard and the assay compared with atropine equivalent.

Determination of Tannins

To 0.5 mL of filtered sample was added 3.75 mL of distilled water and 0.25 mL of Folic Ciocalteu reagent. 0.5 mL of 35% sodium carbonate was added in a 100 mL plastic bottle and shaken thoroughly. The absorbance was measured at 725nm using a spectrophotometer. The blank of the above reagent in distilled water was prepared, different concentration of the sample was prepared and a calibration curve of absorbance against concentration. The tannins content of the sample was measured in mg/mL of tannic acid¹⁰.

Determination of Saponins

The quantitative evaluation of saponins was carried out according to the method described by Madhu *et al.*¹⁰. About 2 g of sample extract was dissolved in 20 mL of 80% methanol in a beaker and 2 mL of vanillin in ethanol was added and mixed well. 2 mL of 75% sulphuric acid was then added mixed well and heated on a water bath at 60°C for 10 minutes. Absorbance was measured at 544nm against a blank with diosgenin used as standard material.

Antioxidant evaluation of oat meal

The radical scavenging activity of Oat meal extract against stable 2,2 diphenyl 2 picryl hydrazyl hydrate (DPPH) was evaluated using the method of Baliyan *et al.*¹¹ but using solution of vitamin E in ethanol as standard. DPPH reacts with an antioxidant which donates hydrogen to the DPPH and subsequently reduce it with change in colour from deep violet to light yellow. 0.1 mL of the extract and vitamin E (200 µg/mL) was mixed with 2.9 mL of 100 µg of freshly prepared methanolic solution. The mixture was then incubated for 30 minutes at 25°C in darkness, after which the decrease in absorbance at 517nm was measured on a UV spectrophotometer (Product Model: 721, Jinan, Shandong, China). The scavenging ability of the extract and vitamin E was calculated as follows;

$$\text{DPPH radical scavenging activity (\%)} = \frac{\text{Abs control} \times \text{Abs sample}}{\text{Abs control}} \times 100 \dots \text{Eqn 1}$$

Where;

Abs control is the absorbance of DPPH radicals and methanol

Abs sample is the absorbance of DPPH radical and sample/standard.

Column Chromatographic Fractionation, Isolation and Purification of Oat Meal Extract

The Oat meal extract was subjected to column chromatography to separate it into its component fractions using a column size of diameter 4.0 cm and length 39.0 cm, using silica gel a highly hygroscopic substance as stationary phase and varying solvent combination of increasing polarity as the mobile phase. Wet packing method was used in packing the glass column and held in a vertical position using a retort stand. The slurry will be prepared by mixing 200 g of silica gel with 300 mL of hexane and then carefully poured down into the column with its tap left open to allow the free flow of solvent into a stabilized conical flask placed on the base of the retort stand. This will allow the solvent to drain unhindered without eroding the silica gel.

The sample was prepared in a mortar by adsorbing 15.0 g of the Oat meal extract to 25.0 g of powdered silica gel in methanol and dried in a hot plate at 40°C with continuous stirring of the adsorbed sample to dryness. The dried sample was allowed to cool and gently spread on top of the column and covered with purified sand to avoid direct contact with eluent. The tap was then opened to allow the eluent to flow at a

controlled rate of 40 drops per minutes. The extract was then eluted with solvent system of increasing polarity in the following ratios: hexane: ethyl acetate 100:0, 95:5, 90:10, 80:20, 70:30, 60:40, 50:50, 40:60, 30:70, 20:80, 10:90, 0:100. The same ratio was applied to ethyl acetate methanol combination. The volume of each solvent system was collected, measured and sprayed uniformly by the sides of the glass and into the column each time. The eluted fractions was then collected in aliquot of 25 mL in 50 mL sterile bottle.

The eluted fractions was further analyzed using TLC with silica gel as stationary phase and ethyl acetate : methanol as mobile phase for further separation the Rf factor was then determined according to the formula described by Gomathi *et al.*,¹² as;

$$R_f = \frac{\text{Distance travelled by the sample from the starting point}}{\text{Distance travelled by solvent from the starting point to the solvent fractions that showed similar}} \dots \text{Eqn 2}$$

TLC mobility and band formation pattern were bulked into a beaker and were subjected to another round of TLC analysis to determine the number of spots. The pooled fractions were concentrated using rotary evaporator at 40°C at 10w pressure then weighed. The oily fractions were then dissolved in 5 ml of n-hexane and stored in a refrigerator for GC-MS spectrophotometric analysis.

Gas Chromatography – Mass spectroscopic Analysis of Oat Meal Fractions

GC-MS analysis of n-hexane fractions was performed using a Perkin Elmer GC Clarus 500 system with a gas chromatograph interfaced with a mass spectrometer. Helium gas was used as a carrier gas at a constant flow rate of 1mL/min and an injection volume of 2 µL was employed. Turbo mass Gold Perkin Elmer was used as mass detector and the software adopted to handle mass spectra and chromatogram was Turbo mass version 5.2. The injection was operated at 250°C and the oven temperature was moderated at 60°C for 15mins and increased gradually increased to 280°C at 3mins. The identification of compounds was based on leedex libraries as well as comparison of their retention indices. The compound were identified after comparison with those in nature using computer library attached to the GC-MS instrument¹².

Results and Discussion

The result of quantitative phytochemical analysis and antioxidant evaluation are presented in Tables 1 and 2 while Table 3 contains results of GC-MS analysis.

Phytochemical analysis

The result of the phytochemical investigation of methanolic extract of Oat meal revealed the presence of flavonoids, phenolics, alkaloids, tannins and saponins. The analysis shows that the quantity of flavonoids, phenolics, saponins and tannins are particularly high in Oat meal, however compared to investigations in several medicinal plants, the quantity of alkaloids in Oat is relatively low from Table 1. The phytochemicals with the highest quantity are flavonoids and phenolics. Flavonoids and phenolics are reported to have antioxidant, antimicrobial and anti-inflammatory properties¹³. Flavonoids are also known to alter the body biochemical reaction and inhibits biomolecules that triggers allergies¹⁰. These phytochemicals may be responsible for anti-inflammatory, antioxidant and antipruritic activities associated with Oat meal.

Tannins commonly found in fruit and cereals are known to have antimicrobial, antioxidant, anti-inflammatory, immunomodulatory effects. Saponins are reported to have antidiabetic effects, they are also useful as expectorant and in treatment of upper respiratory tract infections. Alkaloids which was the least found phytochemical in Oat meal has an analgesic and sedative effect.

Table 1: Qualitative and Quantitative Phytochemical Analysis of Oat Meal Extract

Phytochemicals	Qualitative analysis	Quantitative analysis
Flavonoids	+	198.85
Phenolics	+	60.51
Alkaloids	+	60.36
Tannins	+	20.30
Saponins	+	60.15

Antioxidant

The free radical scavenging activity of the methanolic extract of Oat meal was found to be high when compared to the standard (vitamin E) (Figure 1). The antioxidant properties of Oat meal may be due to their phenolic, polyphenols and saponins content which may possess synergistic effect on its free radical scavenging activity using DPPH which provides information on the reactivity of DPPH which gives a strong absorption band at 517nm. Phenolics and flavonoids produce their antioxidant activities through proton loss electron transfer or inhibition of reactive oxygen species (ROS) producing enzymes.

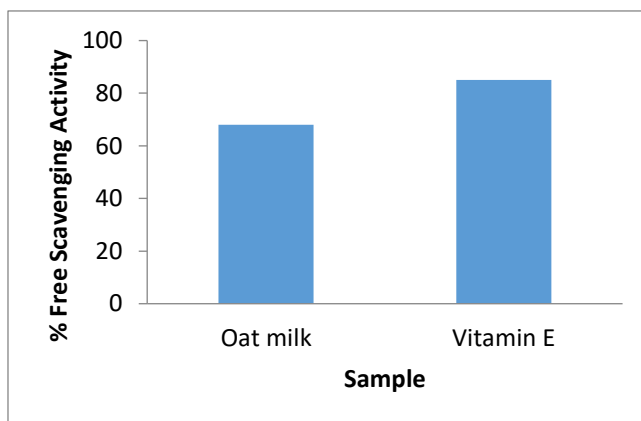


Figure 1: Antioxidant Activity of Oat Meal Extract against Vitamin E (Standard)

Table 2: Antioxidant Evaluation of Oat Meal Methanolic Extract

Sample	Concentration (µg/ml)	% scavenging activity
Oat meal	200	68
Vitamin E	200	85

Column Chromatographic Fractionation, Isolation and Purification of Oat Meal Extract

Bioassay guided fractionation are primarily performed to exclude phytochemicals in plants that may be of no therapeutic benefit. Subsequent purification of these fractions will help in optimizing the isolation and analysis of useful bioactive substance present in plants. Lipophilic fractions were mainly observed in the chromatographic separation and purification of extract of Oat meal.

GC-MS Analysis of Oat Meal Pooled Fractions

GC-MS analysis of n-hexane extract resulted in the identified of 15 useful bioactive compounds (Fig. 2-16), among the identified compounds is Octanamide, N-(2-hydroxyethyl) useful in the management of pain and inflammation, and amphiphilic properties useful as an emulsifier in cosmetic preparations and may also be useful for designing more complex lipid based ingredient that may be useful in creams and ointment.

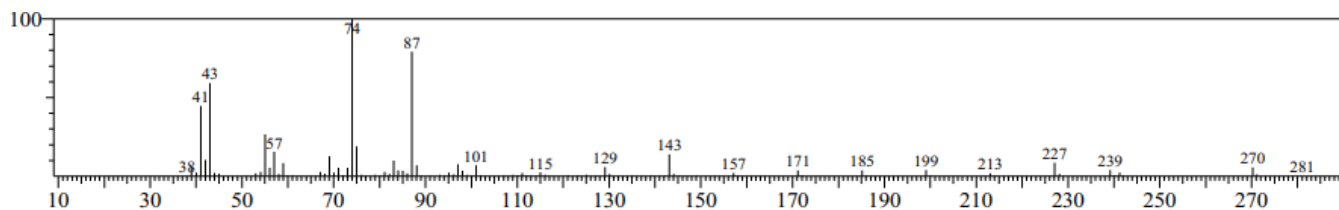


Figure 2: Hexadecanoic acid, methyl ester

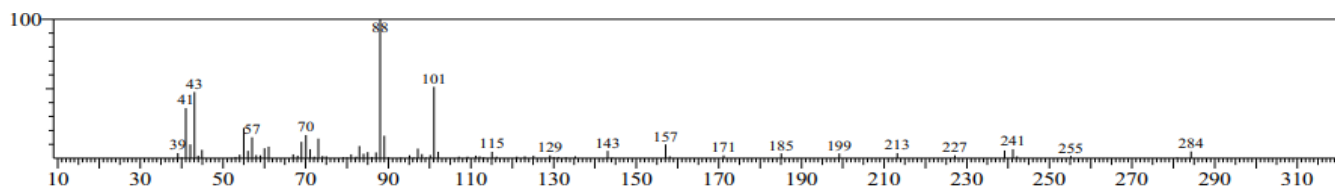


Figure 3: Hexadecanoic acid, ethyl ester

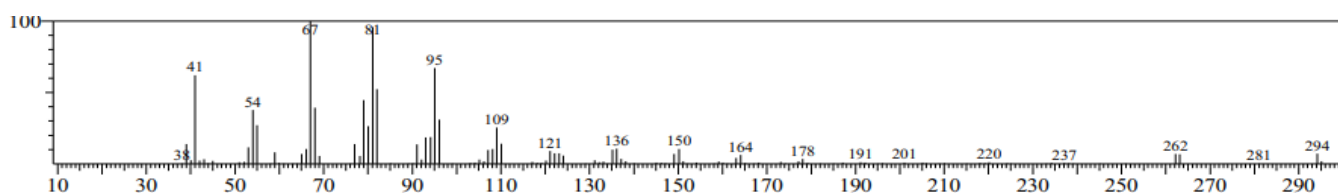


Figure 4: 9,12-Octadecadienoic acid (Z,Z)-, methyl ester

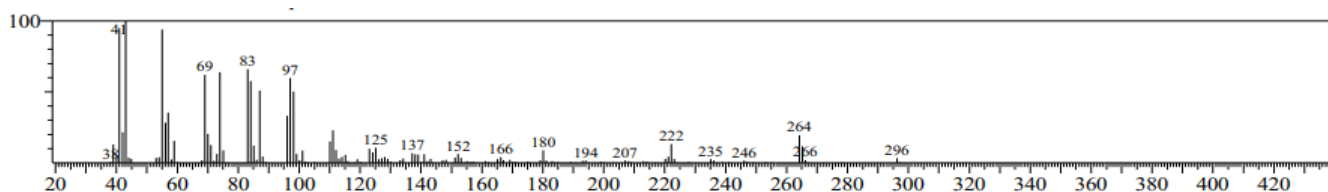


Figure 5: 9-Octadecenoic acid (Z)-, methyl ester

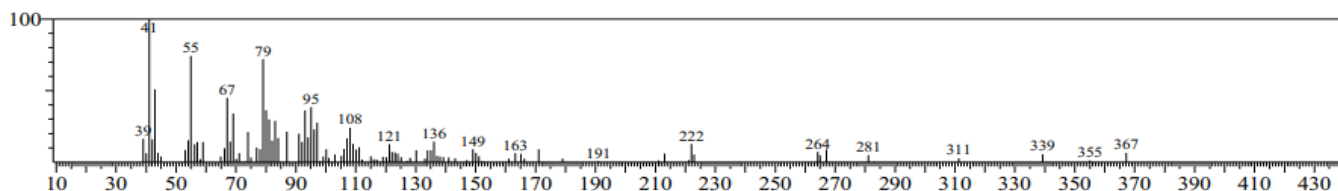


Figure 6: 11,14,17-Eicosatrienoic acid, methyl ester

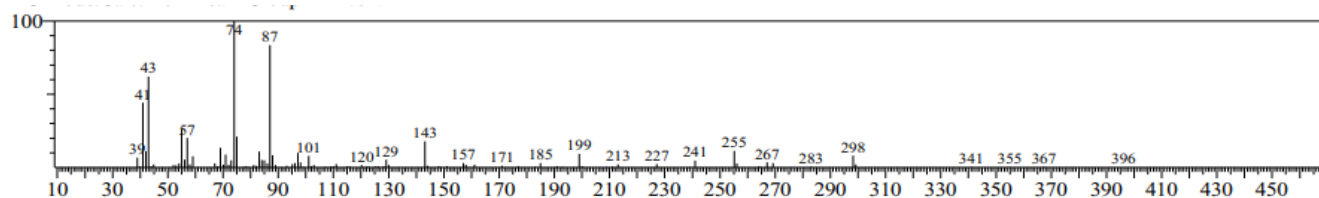


Figure 7: Methyl stearate

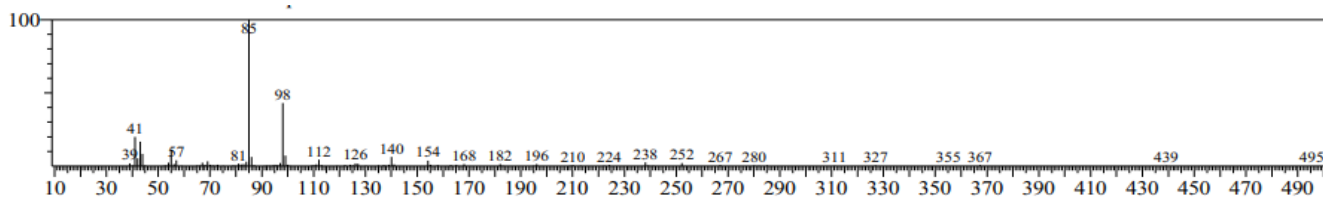


Figure 8: Decanamide, N-(2-hydroxyethyl)-

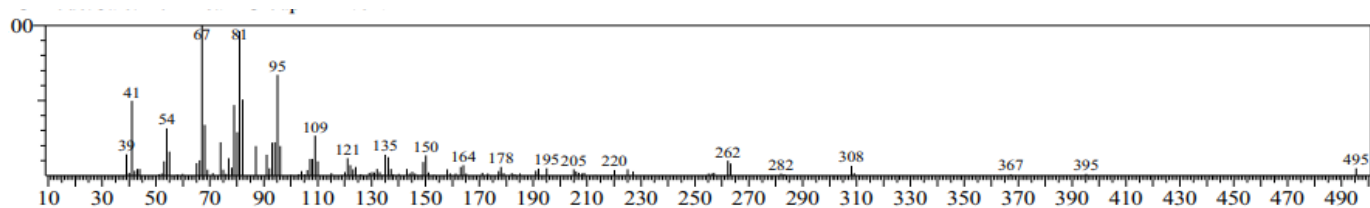


Figure 9: 9,12-Octadecadienoic acid (Z,Z)-, methyl ester

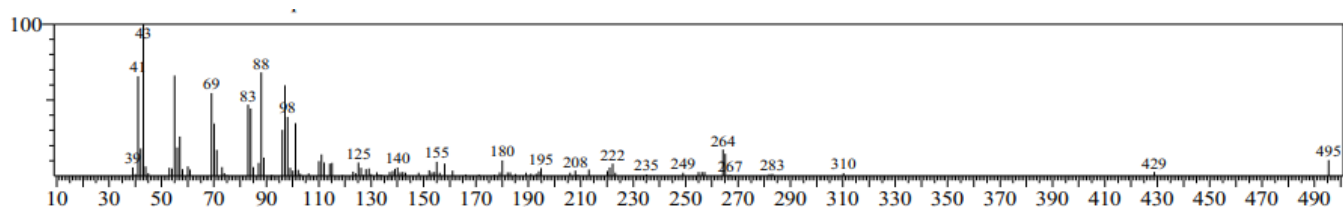


Figure 10: 9-Octadecenoic acid, ethyl ester

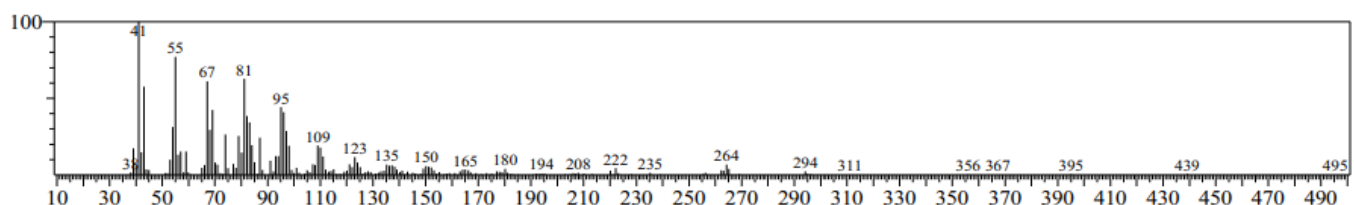


Figure 11: 9,12-Octadecadienoyl chloride, (Z,Z)-

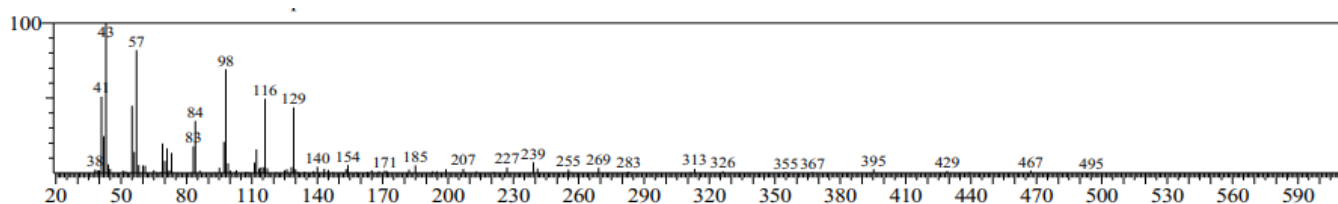


Figure 12: Octadecanoic acid, 2-hydroxy-1,3-propanediyl ester

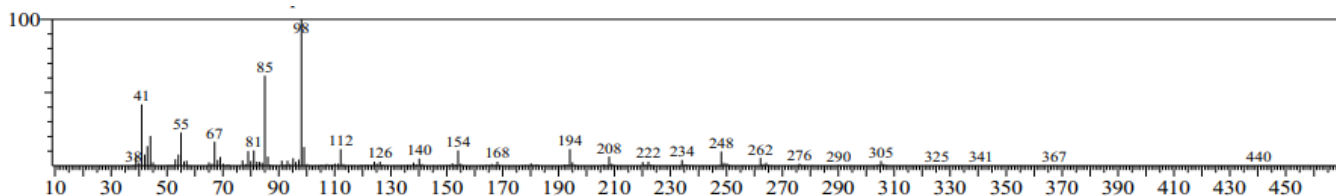


Figure 13: Octanamide, N-(2-hydroxyethyl)-

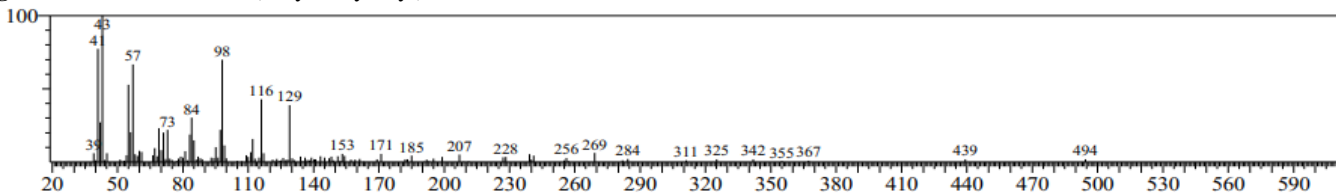


Figure 14: Hexadecanoic acid, 1-[[[(2-aminoethoxy)hydroxyphosphinyl]oxy]methyl]-1,2-ethanediyl ester

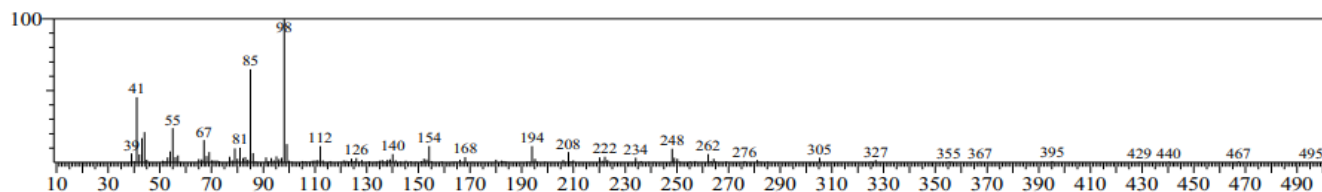


Figure 15: Propan-2-ol, 1-(2-isopropyl-5-methylcyclohexyloxy)-3-(1-piperidyl)-

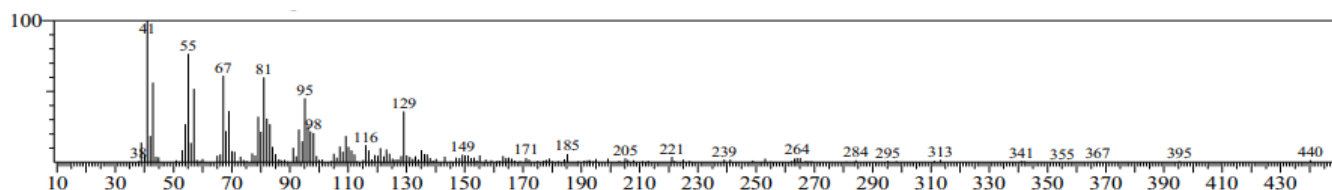


Figure 16: 2-Octylcyclopropene-1-heptano

Decanamide, N-(2-hydroxyethyl) have been shown to have anti-convulsant activities and in the management of neurotoxicity with lower toxicity than vaproate an anti-epileptic agent. Hexadecanoic acid, methyl ester is reported to have antibacterial agent against bacteria such as *Staphylococcus aureus*, *Klebsiella pneumonia*, anti-inflammatory and anti-arthritis properties, cancer preventive and coronary protective activities that may prevent coronary artery disease (CAD). Hexadecanoic acid, ethyl ester have been shown to have antioxidant, antimicrobial, hepatoprotective, anti-arthritis, anti-ulcer, anti-diuretic, neuroprotective and anti-helminthic properties. 9,12-Octadecadienoic acid (Z,Z)-, methyl ester has a variety of therapeutic uses including pain relief, antioxidant, hepatoprotective, spleen strengthen and asthma management and allergic respiratory tract disorder. 9-Octadecenoic acid (Z)-, methyl ester possesses antibacterial and antifungal potential. 11,14,17-Eicosatrienoic acid, methyl ester is a reagent used in the production of drugs and cosmetics including anti-inflammatory drugs, anti-aging products and skin moisturizers. Methyl stearate has neuroprotective properties and in cosmetics as nonionic surfactant, emulsifier and antioxidant. 9-Octadecenoic acid, ethyl ester has been shown to have antibacterial and antifungal properties. 9,12-Octadecadienoyl chloride, (Z,Z) have been shown to have antimicrobial, antifungal and antioxidant properties. Hexadecanoic acid, 1-[(2-aminoethoxy)hydro] has been shown to have antioxidant, antimicrobial, anti-inflammatory and antibacterial properties. Although there seems to be no information on medicinal uses of Octadecanoic acid, 2-hydroxy-1,3-propanediyl, Propan-2-ol, 1-(2-isopropyl-5-methylcyclohexyl) and 2-Octylcyclopropene-1-heptanol but they occur in nature in some medicinal plants with varied medicinal uses such as *Solanum xanthocarpum*¹⁴.

Table 3: GC-MS Analysis of n-hexane Fractions of Oat Meal

S/N	Rt (min)	Name of compound	Molecular formula	Molecular weight (g/mol)	Peak
1.	16.653	Hexadecanoic acid, methyl ester	C ₁₇ H ₃₄ O ₂	270.45	10.92
2.	17.954	Hexadecanoic acid, ethyl ester	C ₁₈ H ₃₆ O ₂	284.477	1.75
3.	20.056	9,12-Octadecadienoic acid (Z,Z)-, methyl ester	C ₁₉ H ₃₄ O ₂	294.472	22.81
4.	20.104	9-Octadecenoic acid (Z)-, methyl ester	C ₁₉ H ₃₆ O ₂	296.487	20.31
5.	20.201	11,14,17-Eicosatrienoic acid, methyl ester	C ₂₁ H ₃₆ O ₂	320.509	1.31
6.	20.481	Methyl stearate	C ₁₉ H ₃₈ O ₂	298.50	0.97
7.	21.017	Decanamide, N-(2-hydroxyethyl)-	C ₁₂ H ₂₅ NO ₂	259.385	1.55
8.	21.238	9,12-Octadecadienoic acid (Z,Z)-, methyl ester	C ₁₉ H ₃₄ O ₂	294.472	5.11

9.	21.270	9-Octadecenoic acid, ethyl ester	C ₂₀ H ₃₈ O ₂	310.514	3.03
10.	23.273	9,12-Octadecadienoyl chloride, (Z,Z)-	C ₁₈ H ₃₁ Cl	298.89	4.41
11.	24.012	Octadecanoic acid, 2-hydroxy-1,3-propanediyl	C ₃₉ H ₇₆ O ₂	625.02	2.06
12.	24.529	Octanamide, N-(2-hydroxyethyl)-	C ₁₀ H ₂₁ NO ₂	187.28	18.62
13.	26.184	Hexadecanoic acid, 1-[(2-aminoethoxy)hydro]	C ₃₅ H ₇₀ NO ₈ P	663.9	0.79
14.	26.663	Propan-2-ol, 1-(2-isopropyl-5-methylcyclohexyl)	C ₁₀ H ₂₀ O	156.26	2.14
15.	28.125	2-Octylcyclopropene-1-heptanol	C ₁₈ H ₃₄ O	266.5	4.21
				100.0	0

Conclusion

Phytochemical screening of the extracts of *K. ivorensis*, *F. paniculata* and *R. beninensis* showed the presence of steroids, flavonoids, saponins, and terpenoids. Significant quantities of tannins, phenols and reducing sugar were found in the extracts of *K. ivorensis* and *F. paniculata*. The GC-MS profiling of the extracts yielded compounds such as octadecanoic, hexadecanoic acid, ethyl oleate and p-mentha-1, 5, 8-triene acid in significant quantities. The extracts of the 3 plants contain secondary metabolites that could be sources of bioactive agents for therapeutic purposes. The information derived from the characterization of plant extracts for their phytochemical composition could be used to screen them for bioactivity.

Conflict of Interest

The authors declare no conflict of interest.

Author's Declaration

The authors hereby declare that the work presented in this article are original and that any liability for claims relating to the content of this article will be borne by them.

Acknowledgments

The authors are grateful to the technical staff in the Department of Pharmaceutical Microbiology and Biotechnology, Faculty of Pharmacy, University of Lagos for their support during this work and Dr Nodza George of the Department of Botany, University of Lagos, who authenticated the plant sample.

References

1. Surampudi V. What are phytochemicals? (And why should you eat more of them?). UCLA Health. Cited December 2024. Retrieved from <https://www.uclahealth.org/news/article/what->

[are-phytochemicals-and-why-should-you-eat-more-them/](#).

Retrieved on 23rd November, 2024

2. Kumar APN, Kumar M, Jose A, Tomer V, Oz E, Proestos C, Zeng M, Elobeid TKS, Oz F. Major Phytochemicals: Recent Advances in Health Benefits and Extraction Method. *Mols*, 2023; 28(2):887.
3. Chow YY, Ting ASY. Influence of fungal infection on plant tissues: FTIR detects compositional changes to plant cell walls. *Fungal Ecol*. 2019; 37: 38–47.
4. Okafor CE, Ijoma IK, Igboamalu CA, Ezebalu CE, Eze CF, Osita-Chikeze JC, Uzor CE, Ekwuekwe AL. Secondary metabolites, spectra characterization, and antioxidant correlation analysis of the polar and nonpolar extracts of *Bryophyllum pinnatum* (Lam) Oken. *BioTechnologia (Pozn)*. 2024; 105(2):121-136.
5. Ijoma IK, Ajiwe EIV, Odinma CS. The organic extracts from the leaves of *Ficus thonningii* Blume, *Jatropha tanjorensis* J.L Ellis and Saroja and *Justicia carnea* Lindley as potential nutraceutical antioxidants and functional foods. *Trends Phyto Res*, 2023; 7(1): 76.85.
6. Nakai K, Tsuruta D. What Are Reactive Oxygen Species, Free Radicals, and Oxidative Stress in Skin Diseases? *Int J Mol Sci*, 2021; 22(19):10799.
7. Onyeloni SO, Etoh AP. Qualitative phytochemical screening, micronutrients and heavy metal evaluation of Oat meal (*Avena sativa*). *Int J Bio*, 2023; 22(4): 103-107.
8. Dhivya R, Manimegalai K. Phytochemical Screening and analysis of active secondary metabolites present in the ethanolic extract of *Calotropis gigantea* leave using GC-MS technique. *World J. Pharm Pharma Sci*. 2016; 5(10): 1510-1523.
9. Rajkumar P, Sundari S, Selvaraj S, Natarajan A, Suganya R, Jayaprakash R, Kasthuri K, Kumaresan S. GC-MS, Phytochemical Analysis and In Silico Approaches of a Medicinal Plant *Acalypha indica*. *J Sci Res*, 2022; 14(2): 671-684.
10. Madhu M, Sailaja V, Satyadev T, Satyanarayana MV. Quantitative phytochemical analysis of selected medicinal plant species by using various organic solvents. *J Pharm Phyto*, 2016; 5(2):25-29.
11. Baliyan S, Mukherjee R, Priyadarshini A, Vibhuti A, Gupta A, Pandey RP, Chang CM. Determination of Antioxidants by DPPH Radical Scavenging Activity and Quantitative Phytochemical Analysis of *Ficus religiosa*. *Molecules*, 2022; 27(4):1326.
12. Gomathi D, Kalaiselvi M, Ravikumar G, Devaki K, Uma C. GC-MS analysis of bioactive compounds from the whole plant ethanolic extract of *Evolvulus alsinoides* (L.) L. *J Food Sci Technol*, 2015; 52(2):1212-7.
13. Zhang Y, Cai P, Cheng G, Zhang Y. A Brief Review of Phenolic Compounds Identified from Plants: Their Extraction, Analysis, and Biological Activity. *Natural Product Communications*, 2022; 17(1).
14. Nithya M, Ragavendran C, Natarajan D. Antibacterial and free radical scavenging activity of a medicinal plant *Solanum xanthocarpum*. *Inter J F Propert*, 2018; 21(1): 313–327.