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[1]Department of Biological Sciences, Faculty of Sciences, National Open University of Nigeria, Jabi Abuja
[2]Department of Chemistry, Shehu Shagari College of Education Sokoto, Sokoto State Nigeria
[3]Department of Biochemistry, College of Medicine, University of Lagos Idi-Araba, Lagos, Nigeria

***Corresponding author's email:**

ualetan@noun.edu.ng

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Phytochemical Profiling of *Euphorbia polycnemoides* Aerial Parts Extracts by GC–MS and Their Pharmacological Relevance

Uduak I. Aletan*[1], Hadiza K. Adamu[2], Olayinka O. Onifade[3], Evans E. Ahams[1], and Janee E. Olubiyi[1]

The Euphorbia genus (family *Euphorbiaceae*) comprises more than 2000 species widely distributed in tropical and subtropical regions, many of which are used in traditional medicine for managing infections, inflammation, and metabolic disorders. However, there is limited phytochemical information on *Euphorbia polycnemoides*. In this study, Gas Chromatography–Mass spectrometry (GC–MS) analysis was conducted to profile the phytochemical constituents of its aerial parts across different solvent extracts. A total of 8 compounds were identified in the ethyl acetate extract, 11 in the ethanol extract, 14 in the methanol extract, and 18 in the aqueous extract. The ethyl acetate extract was dominated by phytol (20.08%) and n-hexadecanoic acid (6%). The ethanol extract exhibited a high content of n-hexadecanoic acid (10.86%) and hexadecanoic acid, ethyl ester (2.76%). The methanol extract showed a high abundance of phytol (10.84%), hexadecanoic acid (4.14%), and hexadecanoic acid, ethyl ester (6.33%). The aqueous extract was found to be rich in terpenes, such as o-cymene (3.08%), L-.alpha.-terpineol (16.36%), and safrole (1.52%) The identified compounds included fatty acids and their esters, phthalates, terpenoids, alcohols, aldehydes, and heterocyclic derivatives. Major constituents such as n-hexadecanoic acid , octadecanoic acid , benzene, 1-methyl-4-(1-methylethenyl)-,phytol, and methyl stearate are known to possess antioxidant, antimicrobial, anti-inflammatory, and anticancer properties. The results provide chemical evidence supporting the ethnomedicinal use of *E. polycnemoides* and suggest its potential as a source of bioactive compounds for therapeutic applications.

Keywords: *Euphorbia polycnemoides*, GC–MS, phytochemicals, bioactive compounds, pharmacological relevance.

1. Introduction

The genus *Euphorbia* (family *Euphorbiaceae*) comprises over 2,000 species worldwide, many of which are well-known for their diverse secondary metabolites, including terpenoids, flavonoids, alkaloids, and fatty acids (Rauf *et al*., 2019). These compounds are responsible for a wide range of biological activities, such as antimicrobial, antioxidant, anti-inflammatory, anticancer, and cytotoxic effects (El-Sherbiny *et al*., 2024). Due to their chemical diversity, species within this genus have been extensively investigated for pharmaceutical and nutraceutical applications.

According to the African Flowering Plants Database (Conservatoire et Jardin Botaniques, n.d.) Euphorbia polycnemoides is an unarmed annual or short-lived perennial herb characterized by branching, erect stems that may reach up to 35 cm in height. It is typically harvested from the wild and utilized in local traditional medicine. In Nigeria, *Euphorbia polycnemoides* (Plate 1) is traditionally utilized in the management of diverse ailments. The crushed leaves, commonly mixed with palm oil, are applied topically to relieve skin eruptions associated with measles, chickenpox, and previously smallpox. When taken orally, preparations of the plant are employed to treat gastrointestinal disorders such as diarrhea and dysentery, while infusions of the dried leaves or whole plant are used as mild laxatives. Additionally, the species is valued in the treatment of respiratory conditions, including cough, sore throat, asthma, and bronchitis. Despite its traditional use, detailed studies elucidating its phytochemical composition and linking it to bioactivities are limited. Understanding the chemical constituents of plant extracts is critical for validating traditional claims,

discovering new bioactive compounds, and guiding further pharmacological research.

Plate 1: *Euphorbia polycnemoides*

Previous studies on *Euphorbia* species have highlighted the importance of solvent polarity in extracting bioactive compounds. Polar solvents such as methanol and water preferentially extract phenolic compounds and glycosides, while semi-polar and non-polar solvents like ethanol and ethyl acetate tend to yield terpenoids and fatty acid derivatives (Chatepa *et al*.,2024). This suggests that a multi-solvent extraction approach can provide a more comprehensive chemical profile and help correlate specific metabolites with pharmacological activities.

The present study aims to perform a GC–MS analysis of ethyl acetate, methanol, aqueous, and ethanol extracts of *Euphorbia polycnemoides* aerial parts. The objectives are to; identify the chemical composition of each extract, quantify the major compounds, and link the identified metabolites to reported antimicrobial and antioxidant activities, including those previously documented for the ethanol and aqueous extracts (Aletan *et al*., 2025). By providing a detailed phytochemical profile and discussing the pharmacological relevance of major compounds, this study seeks to support the ethnomedicinal use of *E. polycnemoides* and identify potential candidates for further pharmacological investigation.

2. Material and Methods

2.1 Sample Collection and Identification

Fresh aerial parts of *Euphorbia polycnemoides* were collected from various sites in Dukai village, Gande, Silame Local Government Area, Sokoto State, Nigeria. Botanical identification of the plant material was carried out at the Botany Unit, Department of Biological Sciences, Faculty of Science, Usmanu Danfodiyo University, Sokoto, Nigeria. A voucher specimen (No. UDUS/ANS/0876) was prepared and deposited in the departmental herbarium for future reference

2.2 Sample Preparation

The collected leaves were thoroughly cleaned to remove dust and extraneous materials, rinsed with deionized water, and shade-dried at ambient temperature to prevent loss of thermolabile constituents. The dried plant material was ground into a fine powder using an electric blender and sieved to achieve uniform particle size. The powdered sample was stored in clean, labelled Ziploc™ polyethylene bags under dry conditions until further use (Aletan & Kwazo, 2019).

2.3 Preparation of Extracts

A total of 250 g of the powdered plant material was separately macerated in various solvents (1:10, w/v), namely ethyl acetate, ethanol, methanol, and distilled water (aqueous), in sequential order of increasing polarity. The mixtures were left to stand for 72 hours at room temperature with intermittent shaking to enhance extraction. Afterward, the extracts were filtered using Whatman No. 1 filter paper, and the filtrates were concentrated to dryness using a rotary evaporator. The dried extracts were stored in airtight containers until analysis

2.4 GC–MS Analysis

The chemical constituents of the extracts were analyzed using an Agilent 8860 Gas Chromatograph coupled with a 5977B Mass Selective Detector (MSD). Separation was achieved on an Elite-5MS capillary column (30 m × 0.25 mm i.d., 0.25 µm film thickness; 5% diphenyl/95% dimethylpolysiloxane stationary phase). The GC–MS was operated in electron impact (EI) ionization mode at 70 eV. High-purity helium (99.999%) was used as the carrier gas at a constant flow rate of 1.0 mL/min, with an injection volume of 1 µL in split mode (10:1).

The injector temperature was set at 300 °C, while the ion source temperature was maintained at 250 °C. The oven temperature was initially held at 110 °C for 1 min, ramped at 15 °C/min to 310 °C, and maintained for 2 min. Mass spectra were recorded at 70 eV over a scan range of 45–450 Da with a scan interval of 0.5 s. A solvent delay of 0–3 min was applied to prevent solvent interference.

3. Results and Discussion

3.1 Results

GC–MS profiling of *Euphorbia polycnemoides* extracts revealed 8 in ethyl acetate, 11 in ethanol, 14 in methanol and 19 compounds in aqueous extracts. The phytochemicals detected included fatty acids, esters, terpenoids, aldehydes, alcohols, and phenolic derivatives as shown in Tables 1-4 and Figs. 1-4 (Supplementary Information).

Table 1 show that the GC-MS analysis of the ethyl acetate extract resulted in a total of 8 distinct compounds, with four major components dominating the extract. The key compound present in significant quantity is n-Hexadecanoic acid (RT: 13.839 min), which accounts for 6.00% of the sample and has a very high-quality score of 99, indicating a highly confident identification. There was also the presence of Octadecanoic acid (RT: 15.401min) at 1.86%. The extracts also presented several other minor components.

Table 1. Phytochemical constituents of the Ethyl acetate extract of *Euphorbia polycnemoides*

Peak #	Retention Time (RT) (min)	% Area	Compound Identified	Quality Score
1	3.9	0.46	D-Limonene	96
5	10.526	0.71	Nonadecane	90
6	11.132	0.52	Benzene, 1,1'-(1,3-propanediyl)bis	95
8	11.916	0.46	1,2-Diphenylcyclopropane	96
9	12.403	1.09	10-Methylnonadecane	91
15	13.839	6.00	n-Hexadecanoic acid	99
20	15.229	3.70	cis-Vaccenic acid	89
21	15.401	1.86	Octadecanoic acid	98

Table 2 shows a total of 11 distinct peaks. The extract composition is dominated by a single, very large presence at 19.361 min, which accounts for 42.19% of the total area. This indicates a high concentration of the compound identified as Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester, with a high-quality score of 91. Other key compounds present in significant quantities include: n-Hexadecanoic acid (RT: 13.822 min), with a high abundance of 10.86% and an excellent quality score of 99, indicating a very confident identification. Phytol (RT: 14.978 min), which makes up 6.07% of the sample. Octadecanoic acid (RT: 15.407 min), at 5.57%. The list also shows other numerous smaller components of the complex mixture.

Table 2. Phytochemical constituents of the Ethanol extract of *Euphorbia polycnemoides*

Peak #	Retention Time (RT) (min)	% Area	Compounds Identified	Quality Score
1	5.994	1.68	Octanoic acid, ethyl ester	96
2	8.363	1.00	Decanoic acid, ethyl ester	97
3	9.948	0.94	2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-, (R)-	97
5	13.484	1.57	Hexadecanoic acid, methyl ester	98
7	13.822	10.86	n-Hexadecanoic acid	99
8	14.045	2.76	Hexadecanoic acid, ethyl ester	97
9	14.978	6.07	Phytol	86
10	15.098	1.61	Methyl stearate	98
13	15.407	5.57	Octadecanoic acid	90
14	15.476	1.55	2(1H)-Naphthalenone, 4a,5,6,7,8,8a-hexahydro-4a,8a-dimethyl-	91
21	19.361	42.19	Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester	91

Table 3 shows that the analysis of the methanol extract of E. polycnemoides identified 14 distinct compounds. The table lists the major identified compounds, their retention times, and the relative abundance (Area %) of each peak.

There are 5 major compounds present, with an Area % > 4. These compounds make up 70.52%

of the total sample composition. They are: Phytol (RT: 14.978 min, Area%: 10.84%), Hexadecanoic acid, ethyl ester (RT: 14.045 min, Area%: 6.33%), Hexadecanoic acid, methyl ester (RT: 13.484 min, Area%: 5.45%), Neophytadiene (RT: 12.717 min, Area%: 5.09%), and n-Hexadecanoic acid (RT: 13.822 min, Area%: 4.14%).

Table 4 shows that Euphorbia polycnemoides aqueous extract is a complex mixture with 19 compounds. The most abundant compound, Benzene, 1-methyl-4-(1-methylethenyl)-, with an Area% of 12.48%.

Table 3. Phytochemical constituents of the Methanol extract of *Euphorbia polycnemoides*

Peak #	Retention Time (RT) (min)	% Area	Compounds Identified	Quality Score
10	8.569	0.47	9-Borabicyclo[3.3.1]nonane, 9-hydroxy-	87
13	9.954	0.94	2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-, (R)-	96
18	12.717	5.09	Neophytadiene	94
19	12.775	1.20	2-Pentadecanone, 6,10,14-trimethyl-	90
20	12.929	1.08	Neophytadiene	93
22	13.484	5.45	Hexadecanoic acid, methyl ester	98
24	13.822	4.14	n-Hexadecanoic acid	99
25	14.045	6.33	Hexadecanoic acid, ethyl ester	97
26	14.846	1.49	Cyclohexene, 4-(4-ethylcyclohexyl)-1-pentyl-	89
27	14.898	1.95	2-Dodecen-1-yl(-)succinic anhydride	90
28	14.978	10.84	Phytol	86
29	15.098	1.64	Methyl stearate	95
32	15.412	1.75	11,13-Dimethyl-12-tetradecen-1-ol acetate	86
37	16.877	0.57	11,13-Dimethyl-12-tetradecen-1-ol acetate	90

Table 4. Phytochemical constituents of the Aqueous extract of *Euphorbia polycnemoides* identified by GC–MS

Peak #	Retention Time (RT) (min)	% Area	Compounds Identified	Quality Score
1	3.865	3.08	o-Cymene	96
2	4.712	12.48	Benzene, 1-methyl-4-(1-methylethenyl)-	98
3	5.078	1.17	Fenchol	95
4	5.136	8.12	Fenchol	87
5	5.296	2.55	3-Cyclohexen-1-ol, 1-methyl-4-(1-methylethyl)-	95
6	5.822	8.06	Isoborneol	80
8	6.492	1.02	2-Cyclohexen-1-ol, 2-methyl-5-(1-methylethenyl)-	96
9	6.549	1.76	(1S,2R,4R,7R)-4-Isopropyl-7-methyl-3,8-dioxatricyclo[5.1.0.02,4]octane	87
11	6.755	2.02	D-Carvone	91
13	7.27	1.52	Safrole	97
14	7.31	1.04	p-Cymen-7-ol	93
21	8.002	2.38	Tricyclo[5.4.0.0(2,8)]undec-9-ene, 2,6,6,9-	90

			tetramethyl-	
23	8.294	1.36	1,2,4-Methenoazulene, decahydro-1,5,5,8a-tetramethyl-	93
24	8.369	1.95	3-Cyclohexene-1-methanol, 5-hydroxy-.alpha.,.alpha.,4-trimethyl-	86
26	8.695	1.94	Longifolene	99
27	8.895	3.69	S-(+)-5-(1-Hydroxy-1-methylethyl)-2-methyl-2-cyclohexen-1-one	90
30	13.496	1.18	Pentadecanoic acid, 14-methyl-, methyl ester	98
36	15.109	0.83	Heptadecanoic acid, 16-methyl-, methyl ester	98

3.2 Discussion

3.2.1 Phytochemical profiling

The GC–MS analysis of *Euphorbia polycnemoides* extracts revealed a diverse array of phytochemicals, including fatty acids, alcohols, esters, terpenoids, and benzofurans, across the ethyl acetate, methanol, aqueous, and ethanol extracts. This chemical diversity aligns with reports in other Euphorbia species, known for producing a wide spectrum of bioactive metabolites (Rauf *et al.*, 2019). Among the three extracts, (ethyl acetate, ethanol, and methanol), n-hexadecanoic acid (palmitic acid), phytol, and octadecanoic acid were consistently identified, suggesting that the compounds constitute the chemical core of the species.

Phytochemical comparison across the extracts indicated quantitative and qualitative variation. The methanol extract (Table 3) showed a high abundance of phytol (10.84%), hexadecanoic acid (4.14%), and hexadecanoic acid, ethyl ester (6.33%), indicating potential antioxidant and antimicrobial properties (Musa *et al.*, 2015) cytotoxic activity (Nisa et al., 2022). The ethyl acetate extract (Table 1) was dominated by phytol (20.08%) and n-hexadecanoic acid (6%), suggesting significant antioxidant, anti-inflammatory, antibacterial, and anti-malarial potential (Siswadi & Saragih, 2021). The ethanol extract (Table 2) exhibited the highest relative abundance of Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester (42.19%). The specific compound, hexadecanoic acid 2-hydroxy-1-(hydroxymethyl)ethyl ester) is not widely documented for its direct biological activity, but its constituent parts, namely hexadecanoic acid (palmitic acid) and other fatty acids, have shown various beneficial properties. These reported activities for related compounds include antioxidant, anti-inflammatory, antiarthritic, and anticancer effects, as well as roles in cholesterol reduction and as a component of plant metabolites (Gupta *et al.*, 2023). The aqueous extract (Table 4) is rich in terpenes, such as Benzene, 1-methyl-4-(1-methylethenyl)- (12.48%), Fenchol (8.12%), and Isoborneol (8.06%), which have been associated with antimicrobial, anti-inflammatory, and antioxidant properties (Masyita *et al.*, 2022). These findings suggest that solvent polarity significantly influences the extraction of both hydrophilic and lipophilic metabolites.

3.2.2 Pharmacological Relevance

The Phytol, present in three of the extracts, has documented antioxidant, antimicrobial, and anticancer activities (Islam *et al.*, 2015). n-Hexadecanoic acid exhibits antimicrobial, anti-inflammatory, and cytotoxic properties (Ravi & Krishnan, 2016). Octadecanoic acid, along with its ethyl ester derivatives, is associated with anti-inflammatory and lipid-lowering effects (Xie *et al.*, 2022). Notably, the presence of these compounds provides a chemical explanation for the significant antimicrobial and antioxidant activities previously reported in *E. polycnemoides* ethanol and aqueous extracts (Aletan *et al.*, 2025). The activities likely result from the synergistic action of fatty acids, terpenoids, and phenolic constituents, which can disrupt microbial membranes and scavenge free radicals (De Rossi *et al.*, 2025).

The ethanol extract revealed hydroxy-palmitic acid derivatives as predominant constituents. The aqueous extract displayed terpenoids such as fenchol and α-terpineol, which are known for antimicrobial and anti-inflammatory effects (Câmara *et al.*, 2024). These extract-specific compounds may contribute to differential bioactivity observed in antimicrobial and antioxidant assays, highlighting the importance of extraction solvent choice and aligning with the findings of Aletan *et al.* (2025).

Ethnomedicinal relevance is supported by these chemical and bioactivity profiles. Euphorbia species have been traditionally used for treating infections, inflammation, and oxidative stress-related conditions. The combined presence of fatty acids, terpenoids, and benzofurans not only supports these traditional uses but also provides targets for further pharmacological validation. The potential synergistic interactions of these metabolites may enhance overall therapeutic efficacy, as reflected in the antimicrobial and antioxidant capacities reported by Aletan *et al.* (2025), warranting future fractionation and bioassay-guided studies.

4. Conclusion

The GC–MS profiling of Euphorbia polycnemoides revealed eight (8) in the ethyl acetate extract, eleven (11) in the ethanol extract, fourteen (14) compounds in the methanol extract, eighteen (18) compounds in the aqueous extract, and Major constituents such as palmitic acid, stearic acid, phytol, linolenic acid derivatives, and 2-monopalmitin are pharmacologically relevant, exhibiting antioxidant, antimicrobial, anti-inflammatory, and anticancer activities. The findings provide chemical evidence for the plant's ethnomedicinal uses and highlight its potential as a source of bioactive compounds for further drug discovery research. This study confirms that solvent polarity is a critical factor in isolating specific phytochemicals from Euphorbia polycnemoides. The identified compounds, many of which are known for their biological activities, warrant further investigation.

Authors' Contribution

AUI conceived the idea, AHK prepared the first draft, OOO and OJE were in charge of the laboratory analysis, AEE did the literature search. All the authors read and approved the final draft.

Conflict of Interest

The authors declare no conflict of interest.

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Supplementary Information: Chromatograms of the Extracts of *Euphorbia polycnemoides* Aerial Parts Identified by GC–MS

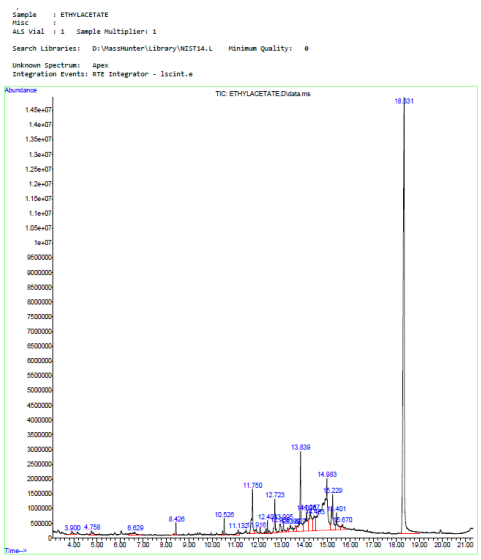

Fig 1: Chromatogram of the Ethyl acetate Extract of *Euphorbia polycnemoides* Identified by GC–MS

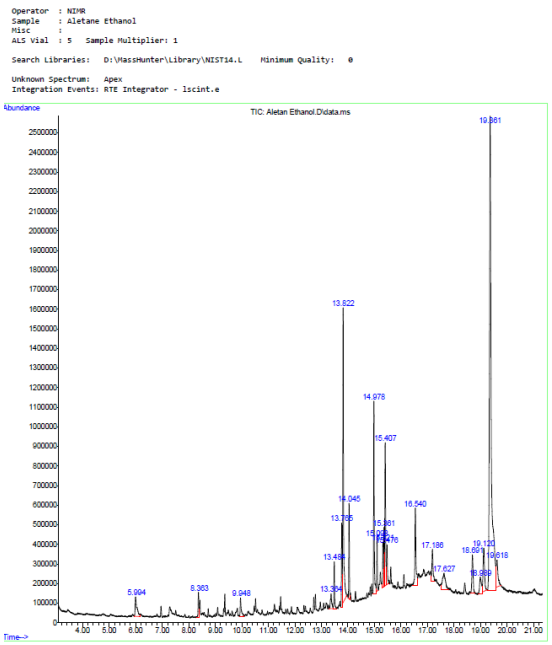

Fig 2: Chromatogram of the Ethanol Extract of *Euphorbia polycnemoides* Identified by GC–MS

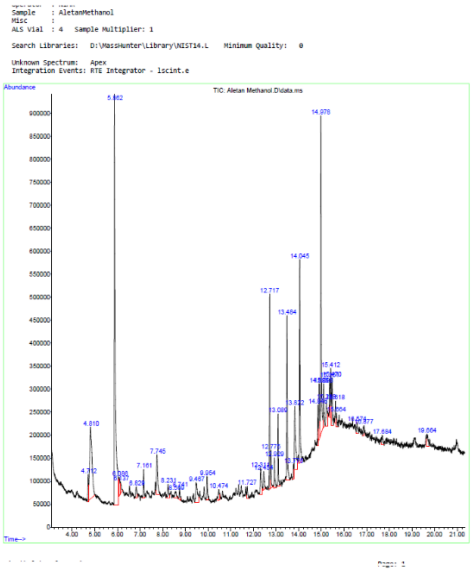

Fig 3: Chromatogram of the Methanol Extract of *Euphorbia polycnemoides* Identified by GC–MS

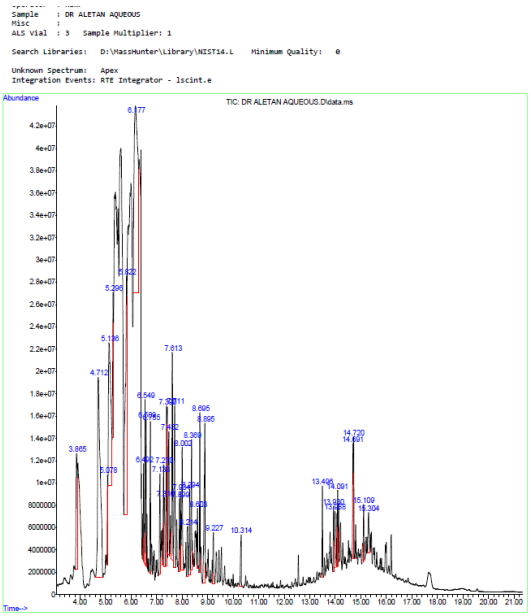

Fig 4: Chromatogram of the Aqueous Extract of *Euphorbia polycnemoides* Identified by GC–MS