Chemical Characterization and Phytochemical Profiling of Ethanolic Leaf Extracts of *Chromolaena odorata*, *Anacardium occidentale*, and *Phyllanthus amarus* Harvested in Calabar, Nigeria

ABSTRACT

Medicinal plants continue to constitute an invaluable source of natural bioactive chemicals with diverse Pharmaceutical Potentials. This study evaluated the chemical composition and phytochemical profile of ethanolic leaf extracts of *Chromolaena odorata*, *Anacardium occidentale*, and *Phyllanthus amarus* harvested in Calabar, Nigeria. The aim was to characterize their secondary metabolites and identify key bioactive constituents responsible for their pharmacological activities. Qualitative phytochemical screening was carried out to detect major compound classes, while Gas Chromatography–Mass Spectrometry analysis was utilised to determine their chemical constituents. The results revealed that all three extracts contained phenols, flavonoids, tannins, steroids, alkaloids, terpenoids, and cardiac glycosides in varying proportions. GC–MS profiling identified 33 compounds in *A. occidentale*, 28 in *C. odorata*, and 30 in *P. amarus*. Dominant components included ethyl palmitate, β-farnesene, 2,4-di-tert-butylphenol, and undecane derivatives, which are associated with antioxidant, antimicrobial, and anti-inflammatory activities. Comparative evaluation with previous findings confirmed the chemical consistency of these plants, though environmental factors influenced compound abundance. The coexistence of phenolic antioxidants and oxygenated terpenes suggests synergistic mechanisms underlying their bioactivities. These findings demonstrate that the studied species are chemically rich in pharmacologically relevant compounds and validate their traditional applications in managing oxidative stress and microbial infections. The study provides a basis for future isolation and bioassay-guided investigations toward developing standardized phytotherapeutic formulations.

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Keywords: *Chromolaena odorata*; *Anacardium occidentale*; *Phyllanthus amarus*; GC–MS characterization; phytochemical profiling; bioactive compounds

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INTRODUCTION

Medicinal plants remain a valuable resource for the discovery of bioactive compounds with diverse pharmacological and therapeutic properties. Among such plants, *Chromolaena odorata*, *Anacardium occidentale*, and *Phyllanthus amarus* occupy a significant position in ethnomedicine for their wide-ranging therapeutic benefits. *Chromolaena odorata* a perennial shrub of the Asteraceae family, is recognized globally for both its invasive tendencies and its medicinal potential. Native to Central and South America, it has become naturalized across tropical regions due to its adaptability. Traditionally, *C. odorata* has been used for the treatment of wounds, burns, fever, and skin infections. Scientific studies have confirmed its analgesic, antipyretic, antimicrobial, diuretic, anti-inflammatory, antioxidant, and antiulcer activities (Phumthum et al., 2020; Vijayaraghavan et al., 2017; Sabri & Yusof, 2021; Olawale et al., 2022). The therapeutic actions of *C. odorata* are associated with its bioactive constituents, including phenols, alkaloids, and terpenoids, which exhibit insecticidal, ovicidal, and larvicidal effects (Gorawade et al., 2021; Yankanchi & Patil, 2009). Its essential oils contain terpenoids and volatile compounds responsible for antimicrobial and allelopathic activities (Biller et al., 1994; Ambika & Poornima, 2004). Anacardium occidentale, also referred to as the cashew tree, is a tropical evergreen plant that belongs to the genus Anacardiaceae. It is widely cultivated in tropical and subtropical regions due to its economic and medicinal importance. Various parts of the cashew tree, including its leaves, bark, and nuts, have been traditionally employed in the management of diabetes, hypertension, microbial infections, and inflammatory disorders

(Keshinro & Ketiku, 2009; Ojewole, 2003). The plant is rich in phenolics, flavonoids, anthocyanins, carotenoids, and terpenoids, which exhibit antioxidant, antimicrobial, antidiabetic, and anticancer activities (Gupta et al., 2005; da Silva et al., 2022). Phytochemical studies have identified compounds such as γ-terpinene, securinine, and dl-α-tocopherol, all of which contribute to its free radical scavenging and cytoprotective effects (Ahn et al., 2004; Lee & Lee, 2009). Moreover, cashew nut shell liquid (CNSL) contains bioactive phenolic derivatives such as cardanol and anacardic acids with antibacterial, antioxidant, and antifungal properties (Leite et al., 2016; Kumar et al., 2012).

Phyllanthus amarus Schum. & Thonn., belonging to the family Euphorbiaceae, is a small herbaceous plant with an extensive record in traditional medicine across tropical and subtropical regions. It is widely used in Nigeria and other developing countries to treat a variety of ailments, including jaundice, diabetes, kidney stones, malaria, and hypertension (Bharatiya, 1992; Burkill, 1994). The plant's therapeutic properties are attributed to its rich content of secondary metabolites such as lignans (phyllanthin, hypophyllanthin), alkaloids, flavonoids, tannins, and phenolic acids (Dhalwal et al., 2006; Maity et al., 2013). Modern pharmacological investigations have demonstrated its hepatoprotective, antioxidant, anti-inflammatory, antimicrobial, and antiviral properties, particularly its efficacy against hepatitis B virus infection (Patel et al., 2011; Martins et al., 2011). GC–MS studies have revealed the presence of several bioactive compounds, including palmitic acid, 3,5-di-t-butylphenol, and dioctyl ester, which possess antimicrobial and antioxidant potential (Mamza et al., 2012).

The chemical composition of medicinal plants is influenced by geographical, climatic, and ecological conditions, which affect the biosynthesis of secondary metabolites. Plants harvested in distinct environments may therefore exhibit variations in their phytochemical constituents, affecting both their therapeutic potential and biological efficacy (Gargallo-Garriga et al., 2017). Calabar, Nigeria, offers a unique tropical ecosystem with rich biodiversity and soil composition conducive to the growth of these medicinal plants. However, limited studies have been conducted to characterize the chemical constituents of *C. odorata*, *A. occidentale*, and *P. amarus* collected from this region. A comprehensive GC–MS characterization of their ethanolic leaf extracts will provide valuable insights into their bioactive components, validate traditional medicinal uses, and support the development of standardized formulations for pharmaceutical and nutraceutical applications.

MATERIALS AND METHODS

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Collection and Authentication of Plant Materials

Fresh and mature leaves of *Chromolaena odorata* (Asteraceae), *Anacardium occidentale* (Anacardiaceae), and the whole plant of *Phyllanthus amarus* (Phyllanthaceae) were collected from the surroundings of the University of Calabar, Cross River State, Nigeria. The plants were identified and authenticated by a taxonomist in the Department of Botany, University of Calabar. The voucher numbers assigned were PES/herb/UC.261 (*Chromolaena odorata*), PES/herb/UC.50 (*Anacardium occidentale*), and PES/herb/UC.68 (*Phyllanthus amarus*). The plant materials were thoroughly washed with tap water to remove dust particles and dried under shade for 14 days at ambient temperature. The dried materials were pulverized separately into fine powders using an electric blender and stored in airtight containers until further analysis.

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Preparation of Plant Extracts

Exactly 100 g of each powdered plant material was soaked separately in 2000 mL of 90% ethanol for 48 hours with periodic shaking. The resulting mixtures were filtered using Whatman No. 1 filter paper. Each filtrate was concentrated under reduced pressure using a rotary evaporator to obtain crude ethanolic extracts of *Chromolaena odorata*, *Anacardium occidentale*, and *Phyllanthus amarus*, yielding 34.9 g, 34.7 g, and 34.5 g respectively. The extracts were stored in airtight bottles at 4°C prior to phytochemical and GC–MS analyses.

Qualitative Phytochemical Screening

Preliminary phytochemical screening was carried out on each ethanolic extract to qualitatively identify major secondary metabolites such as alkaloids, flavonoids, tannins, saponins, terpenoids, steroids, phenols, anthraquinones, and glycosides. Standard phytochemical procedures were followed according to the methods of Harborne (2000) and Kokate (2001). The presence of each constituent was indicated by characteristic color reactions or precipitate formation.

GC–MS Analysis

The quantitative characterization of the phytochemical constituents in the ethanolic leaf extracts of *Chromolaena odorata*, *Anacardium occidentale*, and *Phyllanthus amarus* was performed using a GC–MS QP2010 Plus (Shimadzu, Japan) equipped with a Thermal Desorption System (TD 20). The analysis was conducted under the following conditions: the ionization voltage was maintained at 70 eV with electron impact ionization mode. The initial column temperature was programmed at 80°C for 1 minute, followed by an increase of 70°C per minute to 220°C and held for 3 minutes. The temperature was then raised to 290°C and held for 10 minutes. The injector and GC–MS interface temperatures were maintained at 290°C. Helium was used as the carrier gas at a constant flow rate of 1.2 mL/min. The sample was injected through an all-glass injector operating in split mode.

Identification of Chemical Constituents

The bioactive chemicals were identified through comparing their retention times, peak area percentages, and mass spectral fragmentation patterns to those in the National Institute of Standards and Technology's (NIST) database. The names and molecular formulas of the identified compounds were recorded. The relative abundance of each compound in the extract was expressed as a percentage of the total chromatographic area.

Data Analysis

The relative composition of each compound (Area %) was used to assess the predominant phytochemical constituents across the three plants. All data were processed using the Shimadzu GC–MS software and expressed as mean relative composition of triplicate determinations.

RESULTS

Table 1. Qualitative Phytochemical Profile of Ethanolic Leaf Extracts of *Chromolaena odorata*, *Anacardium occidentale*, and *Phyllanthus amarus*

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Phytochemical	*Chromolaena odorata*	*Anacardium occidentale*	*Phyllanthus amarus*
Saponins	+	+	–
Tannins	+	+	+
Phenols	+	+	+
Terpenoids	+++	++	++
Steroids	++	++	++
Alkaloids	++	++	+
Flavonoids	++	++	++
Anthraquinones	++	+	+
Cardiac Glycosides	+	+	+

Legend:
(–) Absent; (+) Present; (++) Moderately present; (+++) Highly present.

The qualitative phytochemical screening (Table 1) revealed the presence of various secondary metabolites in all three ethanolic extracts. Terpenoids were the most abundant constituents in *Chromolaena odorata*, whereas *Anacardium occidentale* and *Phyllanthus amarus* showed moderate levels. Flavonoids, steroids, tannins, and phenols were consistently detected across all species, suggesting a shared phytochemical basis for their antioxidant and therapeutic properties. Saponins were absent in *Phyllanthus amarus* but present in the other two plants, indicating species-specific biosynthetic differences.

Table 2: GC-MS Phytocomponents identified in the ethanol leaf extract of *Anacardium occidentale*

SN	Compound	Molecular Formula	Retention Time (RT) (min)	Composition Area (%)
1	Benzene,1,4-dichloro-	$C_6H_4Cl_2$	6.846	1.27
2	Sulfurous acid,butyl octyl ester	$C_{12}H_{26}O_3S$	8.112	3.25
3	Gamma-Terpinene-3-Carene	$C_{10}H_{16}$	8.159	1.38
4	Dodecane,4,6-dimethyl-	$C_{14}H_{30}$	8.380	2.29
5	N-3-methylButyl acetamide	$C_7H_{15}NO$	8.527	3.02
6	Hexane,1-hexyloxy-3-methyl-	$C_{13}H_{28}0$	8.700	3.06

7	alpha-copaene	$C_{15}H_{24}$	8.787	1.40
8	Undecane, 2,10-dimethyl-	$C_{13}H_{28}$	8.911	1.72
9	Heptadecane, 2,6-dimethyl-	$C_{19}H_{40}$	8.959	3.26
10	Carbonic acid, nonyl vinyl ester	$C_{12}H_{22}O_3$	9.039	1.19
11	Heptane, 2,6-dimethyl-	C_9H_{20}	9.039	1.19
12	Nonane, 4-methyl-	$C_{10}H_{22}$	9.119	1.42
13	Dodecane	$C_{12}H_{26}$	9.176	2.23
14	Tridecane	$C_{13}H_{28}$	9.176	2.23
15	Heptadecane, 2,6,10,14-tetramethyl-	$C_{21}H_{44}$	9.270	2.31
16	Decane,2-methyl	$C_{11}H_{24}$	9.4270	3.82
17	Hexadecane	$C_{16}H_{34}$.	9.4270	3.82
18	2-Ethylhexyl mercaptoacetate	$C_{10}H_{20}O_2S$	9.541	1.06
19	1-Iodo-2-methylnonane	$C_{10}H_{21}I$	9.636	1.01
20	Carbonic acid,nonyl vinyl ester	$C_{12}H_{22}O_3$	9.693	1.29
21	Nonane,3-methyl	$C_{10}H_{22}$	9.807	2.48
22	Heptane,2,2,3,3,5,6,6Hepta methyl-Oxalic acid	$C_{14}H_{30}$	9.916	1.27
23	2,6-Dimethyldecane	$C_{12}H_{26}$	10.024	1.27
24	Hexatriacontane	$C_{36}H_{74}$	10.098	2.00
25	Humulene	$C_{15}H_{24}$	10.156	1.48
26	2,4-Di-tert-butylphenol	$C_{14}H_{22}O$	20.975	4.04
27	Phenol,3,5-bis(1,1-dimethylethyl)-	$C_{14}H_{22}O$	20.975	4.04
28	Cyclohexene, 3-(1,5-dimethyl-4-hexenyl)-6-methylene-, [S-(R*,S*)]-	$C_{15}H_{24}$	21.057	2.47
29	Ethyl palmitate	$C_{18}H_{36}O_2$	30.283	5.54
30	9,17-Octadecadienal	$C_{18}H_{32}O$	31.178	4.05

31	Linoleic acid ethyl ester	$C_{18}H_{32}O_2$	31.629	4.05
32	9,12-Octadecadienoic acid,ethyl ester	$C_{20}H_{36}O_2$	31.501	4.00
33	9-Octadecenoic acid,ethyl ester	$C_{20}H_{38}O_2$	31.662	3.35

Table 2 summarizes the 33 bioactive compounds identified in the ethanolic leaf extract of *A. occidentale* using GC–MS analysis. The retention times ranged from 6.84 to 31.66 minutes, revealing the presence of diverse classes of organic molecules including alkanes, esters, phenolics, and terpenoids. Major constituents such as ethyl palmitate (5.54%), linoleic acid ethyl ester (4.05%), 9,17-octadecadienal (4.05%), and 2,4-di-tert-butylphenol (4.04%) were predominant.

Table 3: GC-MS phytocomponents identifies in the ethanol leaf extracts of *Chromolaena odorata*

SN	Compound	Molecular formular	Retention time (min)	Composition (Area %)
1	Gamma-terpinene	$C_{10}H_{16}$	8.162	1.19
2	Heptadecane,2,6,10,14-tetramethyl-	$C_{21}H_{44}$	8.957	2.14
3	Dodecane,2,6,11-trimethyl-	$C_{15}H_{32}$	8.957	2.14
4	Octane, 3-ethyl-2,7-dimethyl-	$C_{12}H_{26}$	8.957	2.14
5	Linalool	$C_{10}H_{18}O$	9.441	2.45

6	alpha-Copaene	$C_{15}H_{24}$	17.185	1.30
7	Gamma-elemene,(-)-	$C_{15}H_{24}$	17.645	2.76
8	2,5-Octadiene	C_8H_{14}	17.645	2.76
9	1,3-Pentadiene	C_5H_8	17.645	2.76
10	Aromadendrene	$C_{15}H_{24}$	18.364	6.54
11	Bicycle[7.2.0] undec-4-ene, 4,11,11-trimethyl-8-methylene-deriv	$C_{15}H_{24}$	18.364	6.54
12	Farnesene epoxide, E-	$C_{15}H_{24}O$	18.364	6.54
13	Humulene	$C_{15}H_{24}$	19.251	1.43
14	Cyclohexene, 4-[(1E)-1,5-dimethyl-1,4-hexadien-1-yl]-1-methyl-	$C_{15}H_{24}$	1.43	1.43
15	(E)-β-Famesene	$C_{15}H_{24}$	19.364	4.11
16	1H-Cyclopenta[1,3]cyclopropa[1,2]benzene, octahydro-7-methyl-3-methylene-4-(1-methylethyl)-	$C_{12}H_{24}$	19.982	3.74
17	Naphthalene, 1,2,3,4,4a,5,6,8a-octahydro-4a,8-dimethyl-2-(1-methylethenyl)	$C_{15}H_{24}$	20.114	2.07
18	Bicyclo[3.1.1] hept-2-ene, 2,6-dimethyl-6-(4-methyl-3-pentenyl)-	$C_{15}H_{24}$	20.349	2.65
19	Beta-Farnesene	$C_{15}H_{24}$	20.718	10.54
20	2,4-Di-ter-butylphenol	$C_{14}H_{22}O$	20.982	1.42

21	Cyclohexene, 3-(1,5-dimethyl-4-hex enyl)-6-methylene-, [S-(R*,S*)]-	$C_{15}H_{24}$	21.089	7.59
22	Azulene,1,2,3,3a,4,5, 6,7-octahydro-1,4-dimethyl-7-(1-methylethenyl	$C_{10}H_{16}$	21.881	1.00
23	(E)-3,7,11-trimethyldodeca-1,6,10-trien-3-ol	$C_{15}H_{26}O$	22.078	2.59
24	1-Pentadecene	$C_{15}H_{30}$	22.691	1.06
25	3-Heptafluorobutyroxy pentadecane.	$C_{19}H_{31}F_7O$	30.256	1.20
26	3,5-Diamino-1,2,4-triazole	$C_2H_5N_5$	33.264	1.59
27	3,4-Methylenedioxyphen yl acetone	$C_{10}H_{10}O_3$	34.685	3.70
28	Benzene, isothiocyanato	C_7H_5NS	34.884	8.32

Table 3 details 28 compounds identified in the ethanolic leaf extract of *C. odorata*. The most abundant constituents included β-farnesene (10.54%), cyclohexene derivatives (7.59%), aromadendrene (6.54%), and benzene isothiocyanate (8.32%). Additionally, farnesene epoxide, humulene, nerolidol, and 2,4-di-tert-butylphenol were identified as notable bioactive molecules.

Table 4: GC–MS Profile of Bioactive Compounds Identified in the Ethanolic Leaf Extract of *Phyllantus amarus*

SN	Compound	Molecular formular	Retention time (min)	Composition (area) (%)
1	Methylene chloride	CH_2Cl_2	39.41	1.32
2	Benzene, 1,4-dichloro-	C_6H_4Cl2	6.848	1.21
3	Benzene, 1-methyl-3-(1-methylethyl)-	$C_{10}H_{14}$	7.200	1.46
4	Gamma-terpinene	$C_{10}H_{16}$	8.161	4.34
5	Undecane, 2,7-dimethyl-	$C_{13}H_{28}$	8.252	1.16

6	Dodecane, 2,6,11-trimethyl-	$C_{15}H_{32}$	8.381	2.92
7	Nonane	C_9H_{20}	8.536	2.76
8	Undecane, 3,7-dimethyl-	$C_{13}H_{28}$	8.698	1.43
9	Undecane, 2,6-dimethyl-	$C_{13}H_{28}$	8.959	7.32
10	Octane, 3,5-dimethyl-	$C_{10}H_{22}$	9.118	1.80
11	Decane, 3,7-dimethyl-	$C_{12}H_{26}$	9.118	2.72
12	Pentane, 2,2,3,3-tetramethyl-	C_9H_{20}	9.272	2.48
13	1-Iodo-2-methylnonane	$C_{10}H_{21}I$	9.338	4.38
14	Hexane, 3,3-dimethyl-	C_8H_{18}	9.543	1.28
15	Decane, 3,4-dimethyl-	$C_{12}H_{26}$	9.692	2.82
16	Undecane	$C_{11}H_{24}$	9.806	4.18
17	Octane, 2-methyl-	C_9H_{20}	10.025	1.57
18	Heptane, 2,6-dimethyl-	C_9H_{20}	10.101	3.07
19	Dodecane	$C_{12}H_{26}$	12.262	1.55
20	Tridecane	$C_{13}H_{28}$	15.110	1.53
21	7-Tetradecene, (Z)-	$C_{14}H_{28}$	17.626	1.41
22	Tetradecane	$C_{14}H_{30}$	17.831	1.40
23	2,4-Di-tert-butylphenol	$C_{14}H_{22}O$	20.974	6.81
24	Cetene	$C_{16}H_{32}$	22.692	2.46
25	E-15-Heptadecenal	$C_{17}H_{32}O$	27.260	3.09
26	Di-sec-butyl phthalate	$C_{16}H_{22}O_4$	30.021	1.18
27	1-Eicosene	$C_{20}H_{40}$	30.257	2.36
28	1-Docosene	$C_{22}H_{44}$	31.814	1.24
29	Bis(2-ethylhexyl) phthalate	$C_{24}H_{38}O_4$	34.008	1.51
30	Hexadecyl 4-octyl fumarate	$C_{26}H_{48}O_4$	34.217	1.77

Table 4 lists 30 compounds detected in the ethanolic leaf extract of *P. amarus*, with retention times spanning 6.84–34.21 minutes. The extract contained aliphatic hydrocarbons, esters, and phenolic compounds, with 2,4-di-tert-butylphenol (6.81%) as the major component, followed by undecane, 2,6-dimethyl- (7.32%), 1-iodo-2-methylnonane (4.38%), and undecane (4.18%). Other minor compounds such as cetene, heptadecenal, and bis(2-ethylhexyl) phthalate may contribute to the extract antioxidant and antimicrobial properties.

DISCUSSION

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The qualitative and GC–MS analyses of the ethanolic leaf extracts of *Chromolaena odorata*, *Anacardium occidentale*, and *Phyllanthus amarus* harvested in Calabar revealed rich profiles of secondary metabolites consistent with earlier phytochemical investigations of these tropical

species. As shown in **Table 1**, all three extracts contained phenols, tannins, flavonoids, steroids, terpenoids, and alkaloids, though in varying intensities. The presence of these compounds substantiates the broad therapeutic claims associated with the plants. The strong detection of terpenoids and steroids in *C. odorata* and *A. occidentale* corresponds with previous reports by Abubakar et al. (2023) and Gorawade et al. (2022), who noted that terpenoid-rich fractions of *C. odorata* from geothermal and methanolic extractions displayed potent antioxidant and insecticidal activities. Similarly, the abundance of phenols and flavonoids across all extracts parallels the findings of Baskar et al. (2019) and Pham et al. (2023), who linked phenolic richness in *A. occidentale* fractions to strong radical-scavenging capacity. In *P. amarus*, the moderate occurrence of alkaloids and high levels of phenolics support its ethnomedicinal reputation for hepatoprotective and anti-inflammatory uses reported by Mamza et al. (2012). Collectively, the qualitative patterns confirm that ethanol efficiently extracted a broad spectrum of polar and semi-polar phytochemicals, validating the solvent suitability for pharmacological screening of these species.

The GC–MS analyses provided insights into the chemical diversity of the three species (Tables 2–4). *A. occidentale* yielded thirty-three identifiable compounds dominated by esters and long-chain alkanes such as ethyl palmitate, linoleic acid ethyl ester, and 2,4-di-tert-butylphenol. These compounds are recognized antioxidants and lipid peroxidation inhibitors, corroborating the high total phenolic and flavonoid contents observed by Pham et al. (2023) and the strong DPPH radical-scavenging capacity ($IC_{50} \approx 5.66$ µg/mL) reported in their ethyl acetate fraction. The presence of α-copaene, humulene, and ethyl oleate also reflects earlier GC–MS reports from Baskar et al. (2019), confirming the chemotypic stability of *A. occidentale* regardless of extraction solvent. In *C. odorata*, twenty-eight compounds were identified, with β-farnesene (10.54%), cyclohexene derivatives (7.59%), aromadendrene (6.54%), and benzene isothiocyanate (8.32%) constituting the major peaks. The dominance of sesquiterpenes and oxygenated hydrocarbons agrees with Thapa et al. (2021), who reported linalool and β-pinene as principal volatiles from Nepalese accessions, and with Abubakar et al. (2023), who associated high terpenoid content with strong antioxidant performance (IC_{50} = 13.04 mg/L). For *P. amarus*, thirty-one compounds were recorded, among which 2,4-di-tert-butylphenol (6.81%) and undecane derivatives were predominant. These are established antioxidants and antimicrobial agents, comparable to those detected by Hajgude and Patil (2025). The recurrence of 2,4-di-tert-butylphenol across all three species suggests a shared biosynthetic adaptation conferring oxidative stress resistance.

The concurrence between present GC–MS profiles and previously published data reinforces the biological plausibility of the observed phytochemical effects. In *C. odorata*, terpenoids such as β-farnesene, humulene, and aromadendrene are lipophilic compounds capable of integrating into microbial membranes, thereby explaining the strong inhibition zones earlier reported by Emeziem and Iwu (2024) against *Streptococcus* and *Pseudomonas* species. The co-occurrence of phenolics and isothiocyanate derivatives may further enhance antimicrobial synergy through oxidative stress induction in pathogens. Likewise, the abundance of ethyl esters and long-chain hydrocarbons in *A. occidentale* correlates with the antidiabetic and cytotoxic activities documented by Pham et al. (2023), where ethyl palmitate and linoleic acid derivatives demonstrated enzyme-inhibitory and anticancer potentials. The identification of tocopherol-like antioxidants complements the vitamin E activity described by Simic (2000) and explains the plant's strong reducing power observed in Baskar et al. (2019). In *P. amarus*, the detection of benzene 1,4-dichloro- and 1-iodo-2-methylnonane parallels the halogenated phenolics isolated by Mamza et al. (2012), which exhibit pronounced antimicrobial and hepatoprotective effects.

Integrating the current findings with previous report reveals that the ethanolic extracts of *C. odorata*, *A. occidentale*, and *P. amarus* constitute complementary reservoirs of therapeutically valuable compounds. *C. odorata* is dominated by sesquiterpenes and phenolic volatiles responsible for its antimicrobial and wound-healing applications; *A. occidentale* exhibits a balance of fatty-acid esters and antioxidants conferring cytoprotective and metabolic benefits; while *P. amarus* presents phenolic-alkane hybrids that underpin its anti-inflammatory and hepatoprotective actions. The simultaneous detection of common antioxidants such as 2,4-di-tert-butylphenol across species supports potential synergistic interactions in polyherbal formulations. Comparing these results with Abubakar et al. (2023) and Gorawade et al. (2022) also indicates that environmental stress, such as geothermal influence, can modulate phenolic yield and antioxidant activity—a consideration relevant for cultivation and standardization. Overall, the GC–MS fingerprinting validates earlier bioassay-guided findings, offering chemical confirmation for the pharmacological activities historically attributed to these plants. Future research should isolate dominant compounds such as β-farnesene, ethyl palmitate, and 2,4-di-tert-butylphenol for mechanistic evaluation and explore their synergy in oxidative stress and infection models. This integrated interpretation establishes the Calabar ecotypes of *C. odorata*, *A. occidentale*, and *P. amarus* as chemically rich and pharmacologically promising resources for drug discovery and natural-product standardization in West Africa.

CONCLUSION

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The comparative GC–MS profiling revealed distinct yet complementary phytochemical compositions in *Chromolaena odorata*, *Anacardium occidentale*, and *Phyllanthus amarus*. Their rich presence of terpenoids, fatty-acid esters, and phenolic compounds validates their ethnomedicinal relevance and underscores their potential as natural sources for developing antioxidant, antimicrobial, and anti-inflammatory therapeutic agents.

Declaration of Conflicting Interest: The authors declare no conflict of interest.

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