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Antibacterial Activity and Gc-Ms Analysis of Gouania Longispicata Eng. Root Extract Against Bacterial Pathogens

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Abstract
This study investigates the phytochemical composition and antibacterial activity of the n-hexane root extract of Gouania longispicata, a medicinal plant traditionally used across East Africa. Gas chromatography-mass spectrometry (GC-MS) analysis identified 39 compounds (99.193% total composition), predominantly esters (92.074%), including hexadecanoic acid, 3-hydroxy-methylester (28.73%), hexadecanoic acid, methyl ester (24.02%), and octadecanoic acid, 3-hydroxy-methyl ester (13.33%). The extract demonstrated dose-dependent antibacterial activity against Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus, and Bacillus cereus, with inhibition zones ranging from 6.8 ± 0.15 mm (25 mg/mL) to 15.6 ± 0.32 mm (100 mg/mL). B. cereus exhibited the highest susceptibility, while P. aeruginosa was the least responsive. Comparative analysis revealed the root extract's superior antibacterial efficacy over leaf extracts from prior studies, likely attributable to its high ester content. Gentamicin (10 µg/disc) controls showed significantly larger inhibition zones (19.5–23.1 mm), underscoring the extract's moderate yet promising activity. The findings align with ethno medicinal claims and highlight the potential of G. longispicata root extracts as a natural antimicrobial resource, particularly against drug-resistant pathogens. Further research is warranted to isolate and characterize specific bioactive compounds for therapeutic development.

Keywords: Gouania Longispicata, Antibacterial, Phytochemical, Extract Root

Background
Gouania longispicata Eng. is a climbing liana belonging to the family of Rhamnaceae comprising 58 genera with about 900 species worldwide [1]. In Africa, Gouania longispicata Eng. is commonly found along forest edges and clearings in countries like Ethiopia, Nigeria, Uganda, Democratic Republic of the Congo (DRC), and Sudan [2]. Different parts of G. longispicata are ingredients of herbal remedies used in traditional medicine by various ethnicities. For instance, its leaf preparations are used to treat oral thrush in Ethiopia, to treat fetal issues in Rwanda, to hasten childbirth, to treat stomachache, and malaria in Tanzania [3,4]. In the DRC, children of Mufti pygmies are given G. longispicata stem sap to make them grow strong [3]. In Uganda, G. longispicata leaves, stems, and roots treat more than forty ailments, including stomachache [3]. Previous phytochemical analyses of G. longispicata showed phenolic, flavonoids, cardiac

glycosides, steroids, sterols, saponins, and resins in its leaf, stem, and whole plant extracts. Antimicrobial activity against harmful bacteria and fungi, including some strains that were resistant to multiple drugs, was demonstrated by these extracts both alone and in combination with common medicines [5]. Despite these wider medicinal claims of the plant, its phytochemicals and GC-MS Analysis of different crude extract reports are limited. Therefore, this study aimed to analyze the phytochemical constituent of the n-hexane root extract of G. longispicata and evaluate its antibacterial activities.

Objectives

This investigation focuses on the n-hexane extraction of G. longispicata root, identifying its chemical composition, and assessing the antibacterial activity of the n-hexane crude extract against specific bacterial strains, including Bacillus cereus, Pseudomonas aeruginosa, Escherichia coli, and Staphylococcus aureus for the first time.

Material and Methods

Preparation of Plant Materials and Extraction

The roots of G. longispicata were collected from Amuru district, Horro Guduru college zone, Oromia, Ethiopia. The plant was identified by a botanist Prof. Sileshi Nemomsa from Addis Ababa University Herbarium. The voucher specimen number (DB 002/14) was kept at the Ethiopian National Herbarium. The plant materials were shade-dried and then pulverized into fine particles. The pulverized root (100 g) was macerated using n-hexane for 48 hr. The extracts were filtered through Whatman No 1 filter paper and solvents were evaporated in a vacuum using a rotary evaporator and the extract was stored in the refrigerator at 20 oC.

Experimental Procedure

The 0.2 g of the extract was added to 3 mL of 2N KOH in methanol and refluxed for one hour. Then it was cooled to room temperature, and 5% 5 mL HCL in methanol was added, again refluxed for one hour. After 1 hour, the content in the container cooled to room temperature. Then the content in the flask was transferred to a separatory funnel; to this, an equal volume of n-hexane and water was added (3 mL each). Finally, discard the aqueous part and transfer the organic phase into GC-MS vials, then inject it into the GC-MS for analysis.

GC-MS Analysis

GC-MS analysis was done using a GC (7890B, Agilent Technologies, China) coupled with an MS (5977B Network, Agilent Technologies, USA). The GC had an HP-5MS column (non-polar column, Agilent Technologies), 30 m × 250 µm internal diameter, and 0.25 µm film thickness. The carrier gas helium was flowing at a rate of 1 mL/min. The injector temperature was 250 °C, and the injection mode was split mode with a split ratio of 7:1. The initial oven temperature was programmed from 50˚C and held for 1 min. It was raised to 180 ˚C at 15 ˚C/min and then ramped by 1.5 ˚C/min to reach 210 ˚C and finally raised to 280 with the rate of 20 ˚C/min at this temperature held for 6 min. Mass spectra were recorded in EI mode at 70 eV, scanning the 45-550 m/z range.

Antibacterial Activity Test

Bacteria Include: Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus, and Bacillus cereus. The bacteria strains were obtained from the microbiology laboratory of the Department of Biology, Wollega University, Ethiopia.

The antibacterial activity of the root extract of G. longispicata was evaluated by the disc diffusion method and performed according to the guidelines of the National Committee for Clinical Laboratory Standards. A 24-hour-old culture of selected bacteria was mixed with sterile physiological saline (0.85%) and the turbidity was adjusted to the standard inoculum of MacFarland scale 0.5 [~106 colony forming units (CFU) per milliliter]. Petri dishes containing 20µL of Mueller-Hinton agar were used to inoculate bacterial suspension. Filter paper discs (Whatman no. 1, diameter = 6 mm) impregnated with the extract solutions prepared in DMSO (100mg/ml, 50mg/ml, and 25mg/ml) were placed on the inoculated plates, and Petri dishes were incubated for 24 h at 37°C. A paper disc impregnated with gentamicin (10µg/disc) was used as a positive control. The inhibition zone diameters were measured in millimeters as indicated (Table 2).

Results and Discussion

Phytochemical Composition of N-Hexane Extract

The results of GC-MS analysis led to the identification of 39 compounds accounting for about 99.193% of n-hexane root extracts of G. longispicata. These compounds were identified through gas chromatography attached to mass spectrometry and the results are shown in Figure 1 and Table 1. These components are classified as a variety of natural compounds, including ethers (0.289%), aldehydes (0.4%), alkanes (0.374%), phenols (0.292%), esters (92.074%), ketones (3.191%), and steroids (1.171%). According to this information, 92.074% was the Ester group of compounds. Most of the identified compounds belonged to ester compounds (92.074%) which include hexadecanoic acid, 3-hydroxy-, methyl ester (28.73%), hexadecanoic acid, methyl ester (24.02%), octadecanoic acid, 3-hydroxy-, methyl ester (13.33%), methyl stearate (5.58%), and methyl 11-docosenoate (3.72%).

No	RT	Peak Area (%)	Compound name	Molecular Formula	Group
1	7.245	2.662	Acetophenone, 4'-hydroxy-	$C_8H_8O_2$	ketone
2	10.422	0.292	Phenol, 2,4-bis(1,1-dimethylethyl)-	$C_{14}H_{22}O$	phenol
3	10.745	0.742	Azelaic acid, dimethyl ester	$C_{11}H_{20}O_4$	Ester
4	11.202	0.199	Cyclohexadecane	$C_{16}H_{32}$	alkane
5	11.288	0.289	Undecanal dimethyl acetal	$C_{13}H_{28}O_2$	Ether
6	11.473	0.400	Tetradecanal	$C_{14}H_{28}O$	Aldehyde
7	12.963	0.275	Myristic acid, methyl ester	$C_{15}H_{30}O_2$	Ester
8	14.089	0.175	Cyclohexadecane	$C_{16}H_{32}$	Alkane
9	14.719	0.253	Pentadecanoic acid, methyl ester	$C_{16}H_{32}O_2$	Ester
10	15.741	0.209	Methyl 3-hydroxytetradecanoate	$C_{15}H_{30}O_3$	Ester
11	17.144	24.020	Hexadecanoic acid, methyl ester	$C_{17}H_{34}O_2$	Ester
12	18.317	0.508	Cholestan-15-one, 3-(acetyloxy)-14-butyl-, (3.beta.,5.alpha.)-	$C_{33}H_{56}O_3$	Steroid
13	19.715	0.633	Heptadecanoic acid, methyl ester	$C_{18}H_{36}O_2$	Ester
14	20.275	0.622	Methyl 3-acetylhydroxypalmitate	$C_{19}H_{36}O_4$	
15	21.707	28.733	Hexadecanoic acid, 3-hydroxy-, methyl ester	$C17H_34O_3$	Ester
16	23.128	5.578	Methyl stearate	$C_{19}H_{38}O_2$	Ester
17	23.445	0.332	Linoleic acid	$C_{18}H_{32}O_2$	Fatty acid
18	23.676	0.172	9,12,15-Octadecatrienoic acid, methyl ester	$C_{19}H_{32}O_2$	Ester
19	26.697	0.683	Nonadecanoic acid, methyl ester	$C_{20}H_{40}O_2$	Ester
20	27.592	1.150	4,7-Octadecadienoic acid, methyl ester	$C_{19}H_{34}O_2$	Ester
21	27.719	0.474	Methyl 9.cis.,11.trans.t,13.trans.-octadecatrienoate	$C_{19}H_{32}O_2$	-
22	28.978	13.330	3-hydroxy-, methyl ester	$C_{19}H_{38}O_3$	Ester
23	30.509	2.407	Eicosanoic acid, methyl ester	$C_{21}H_{42}O_2$	Ester
24	32.16	0.772	Heneicosanoic acid, methyl ester	$C_{22}H_{44}O_2$	Ester
25	32.409	0.183	Isolongifolol	$C_{15}H_{26}O$	Alcohol
26	32.478	2.495	cis-11,14-Eicosadienoic acid, methyl ester	$C_{21}H_{38}O_2$	Ester
27	32.952	3.721	Methyl 11-docosenoate	$C_{23}H_{44}O_2$	Ester
28	33.171	1.477	Methyl 20-methyl-heneicosanoate	$C_{23}H_{46}O_2$	Ester
29	33.321	0.393	2-Eicosen-5-olide	$C_{20}H_{36}O_2$	Ester
30	34.02	0.871	Tricosanoic acid, methyl ester	$C_{24}H_{48}O_2$	Ester
31	34.095	0.233	Campesterol	$C_{28}H_{48}O$	Steroid
32	34.921	1.292	Tetracosanoic acid, methyl ester	$C_{25}H_{50}O_2$	Ester
33	35.395	0.702	11,13-Dimethyl-12-tetradecen-1-ol acetate	$C_{18}H_{34}O_2$	Ester
34	35.92	0.654	Pentacosanoic acid, methyl ester	$C_{26}H_{52}O_2$	Ester
35	36.307	0.722	Methyl 2-hydroxy-tetracosanoate	$C_{25}H_{50}O_3$	Ester
36	36.365	1.171	(24R)-Stigmast-5-en-3.beta.-ol	$C_{29}H_{50}O$	Steroid
37	37.075	0.480	Hexacosanoic acid, methyl ester	$C_{27}H_{54}O_2$	Ester
38	37.549	0.529	(2R,3R,4aR,5S,8aS)-2-Hydroxy-4a,5-dimethyl-3-(prop-1-en-2-yl) octahydronaphthalen-1(2H)-one	$C_{15}H_{24}O_2$	Ketone
39	38.895	0.146	gamma.-Tocopherol	$C_{28}H_{48}O_2$	Ether
Abbreviation: RT Retention Time					

Table 1 : Chemical Composition of Gouania Longispicata N-Hexane Extract

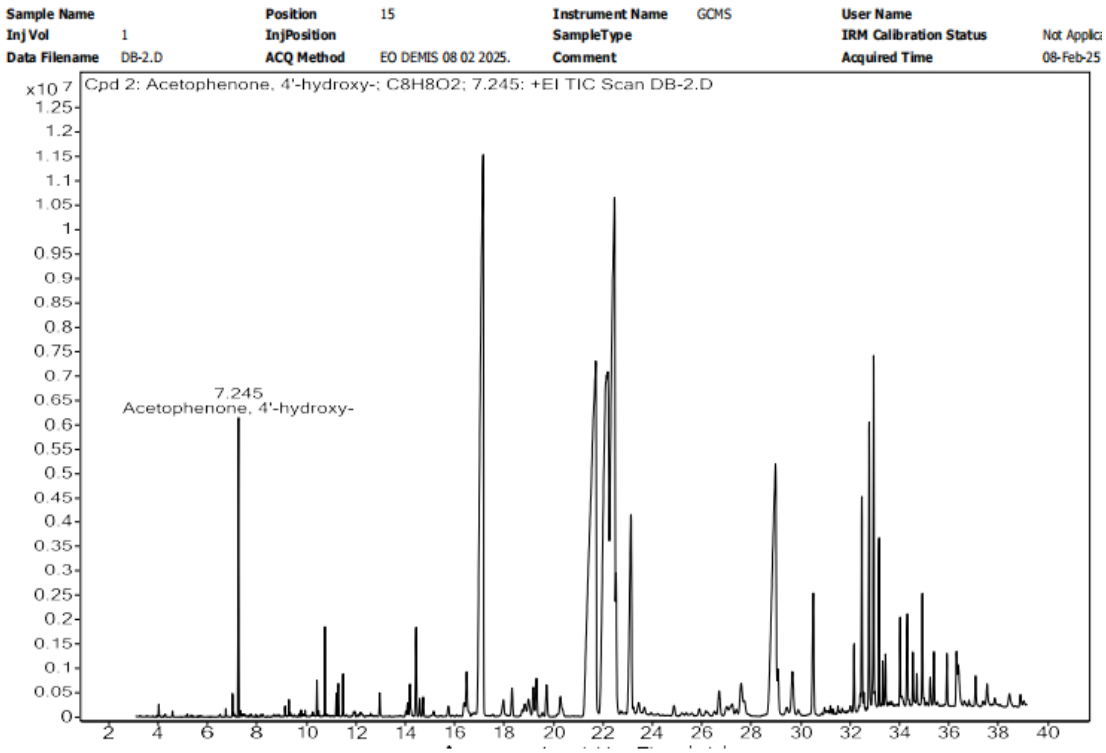

Figure 1: GC-MS Chromatography of N-Hexane Crude Extract of G. Longispicata

Antimicrobial Activity Test

The extract showed antimicrobial activity against selected pathogenic bacteria at a concentration of 100mg/mL, 50 mg/mL, and 25 mg/mL. The results are presented in Table 2.

Discussion

The results of GC-MS analysis led to the identification of 39 compounds accounting for about 99.979% of n-hexane root extracts of G. longispicata. These compounds were identified through gas chromatography attached to mass spectrometry and the results are shown in Figure1 and Table 1. These components are classified as a variety of natural compounds, including ethers (0.289%), aldehydes (0.4%), alkanes (0.374%), phenols (0.292%), esters (92.074%), ketones (3.191%), and steroids (1.171%). Most of the identified compounds belonged to ester compounds (92.074%) which include hexadecanoic acid, 3-hydroxy-, methyl ester (28.73%), hexadecanoic acid, methyl ester (24.02%), octadecanoic acid, 3-hydroxy-, methyl ester (13.33%), methyl stearate (5.58%), and methyl 11-docosenoate (3.72%). The GC/MS study of G. longispicata n-hexane extract revealed a higher ester group composition, which is advantageous for the production of medicines and cosmetics as well as for antioxidant and antimicrobial activities [6].

Sample	**Concentration mg/ml**	**The zone of inhibition diameter (mm) including disc diameter (6mm)**			
		E. coli	*P. aeruginosa*	*S. aureus*	*B. cereus*
n-hexane extract	100	13.3±0.43	8.7±0.56	12.5±0.12	14.6±0.32
	50	11.2±0.17	7.2±0.02	11.2±0.19	11.7±0.01
	25	9.0±0.15	6.9±0.48	9.3±0.34	10.4±0.11
gentamicin	10µg/disc	21.8±0.02	19.5±0.31	20.2±0.24	23.1±0.32
DMSO	10%	6.0±0.00	6.0±0.00	6.0±0.00	6.0±0.00

Table 2: Inhibition Zone Diameter (mm) of Gouania Longispicata Against Selected Bacteria

Values are expressed as mean± SD. 6.0±00=no inhibition, DMSO- dimethyl sulfoxide.

The octadecanoic acid, Hexadecanoic acid, 3-hydroxy-, methyl ester, Hexadecanoic acid, 3-hydroxy-methyl ester, and methyl stearate showed antibacterial activity in previous studies while Hexadecanoic acid, 3-hydroxy-, methyl ester and methyl stearate showed antioxidant and anti-inflammatory activity [6,7]. Because of the rise in antimicrobial-resistant microorganisms, the use of compounds extracted from medicinal plants may be helpful in the development of antimicrobial agents 65. This study result showed promising antibacterial activities of the n-hexane extract against Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus, and Bacillus cereus, with a zone of inhibition 13.3± 0.43 mm, 8.7 ± 0.56 mm, and 12.6 ± 0.32 mm respectively at 100mg/ml. The extract demonstrated dose-dependent antibacterial activity against E. coli, P. aeruginosa, S. aureus, and B. cereus, with inhibition zones ranging from 6.8 ±

0.15 mm (25 mg/mL) to 15.6 ± 0.32 mm (100 mg/mL). B. cereus exhibited the highest susceptibility, while P. aeruginosa was the least responsive. The positive standard, ceftriaxone, gave the zones of inhibition for E. coli (21.8 ± 0.02 mm), P. aeruginosa (19.5 ± 0.31mm), S. aureus (20.2 ± 0.24 mm), and B. cereus (23.1 ± 0.32 mm). The extract exhibited good antibacterial activities against all studied bacteria when compared with a previous study [1]. Further comparison with literature reported for the leaf extract that reported zone inhibition was lower than the current study. However, this study's results show better antibacterial activities compared to the n-hexane leaf extracts of G. longispicata. The difference in antimicrobial activities with other studies might be possibly due to the difference in concentration and types of antimicrobial agents in different parts of the study plant, environmental factors, and the laboratory method.

Conclusion

In this study 39 compounds were identified from the root of G. longispicata. The major chemical constituents included ester compounds (92.074%), which include hexadecanoic acid, 3-hydroxy-, methyl ester (28.73%), hexadecanoic acid, methyl ester (24.02%), octadecanoic acid, 3-hydroxy-, methyl ester (13.33%), methyl stearate (5.58%), and methyl 11-docosenoate (3.72%) and were n-hexane crude extract to explore its antibacterial activities. The root of G. longispicata demonstrated good antibacterial potential against E. coli, S. aureus, P. aeruginosa, and B. cereus. Overall, the results of the study suggest that the composition of the root of G. longispicata is responsible for its antibacterial properties. Therefore, the root of G. longispicata will be a potential source for the discovery of antibacterial drugs. However, the compounds responsible for the antibacterial activity are currently unclear [8]. Therefore, further investigation is needed to isolate, identify, and characterize the antibacterial compounds present in the roots of the studied plant.

Data Availability

The data used to support the findings of the present study are available from the corresponding author.

Author Contributions

Diriba Borena performed experiments, analyzed the data, interpreted the data, and Wakshum Adugna, wrote the paper. Negera Abdisa and Zelalem Abdisa supervise research. All authors read and approved the final manuscript.

Competing Interests

The authors declare no competing interests. Additional information Correspondence and requests for materials should be addressed to B.A.

References

1. Gumisiriza H, Olet EA, Mwikali L, Akatuhebwa R, Omara T, Lejju JB, et al. Antibacterial and Antioxidant Activities of Flavonoids , Phenolic and Flavonoid Glycosides from Gouania longispicata Leaves. 2024;2085–101.
2. Gumisiriza H, Birungi G, Omara T, Lejju JB, Sesaazi CD. Polyphenolic content, antioxidant activity and acute toxicity of Gouania longispicata Engl. leaves. LIANBS. 2025.
3. Gumisiriza H, Birungi G, Lejju JB, Olet EA, Kembabazi O, Duncan C. Ethnobotany and Antimicrobial Activity of Gouania longispicata Engl . Ethnobotany and Antimicrobial Activity of Gouania longispicata Engl . 2020;(July).
4. Gumisiriza H, Apio E, Mwikali L, Akatuhebwa R, Kembabazi O, Omara T, et al. Heliyon Ethnopharmacology , phytochemistry , pharmacology and toxicity of the genus Gouania. Heliyon [Internet]. 2024;10(24):e40933.
5. Gumisiriza H. Polyphenolic Content , Antioxidant Activity and Acute Toxicity of Gouania longispicata Engl . Leaves. 2025;14(2):1–11.
6. Ashoka GB, Shivanna MB. In vitro Antioxidant and Antibacterial Activities of Jatropha heynei Leaf Extract and GC-MS Profiling. 2023;(February).
7. Reza ASMA, Haque A, Sarker J, Samima M, Rahman M, Montakim A, et al. Antiproliferative and antioxidant potentials of bioactive edible vegetable fraction of Achyranthes ferruginea Roxb . in cancer cell line. 2021;(March):3777–805.
8. Sellami M, Châari A, Aissa I, Bouaziz M, Gargouri Y, Miled N. Newly synthesized dopamine ester derivatives and assessment of their antioxidant, antimicrobial and hemolytic activities. Process Biochemistry. 2013 Oct 1;48(10):1481-7.