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Research Article

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Chemical composition and antioxidant activity of acetone extract from *Chlorophytum laxum*

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ABSTRACT

The genus *Chlorophytum* is renowned for its ornamental value and applications in traditional medicine, especially in tropical regions. This study presents the first report on the phytochemical screening, volatile compound profiling, and antioxidant activity of acetone extracts from the leaves and roots of *Chlorophytum laxum*. Gas chromatography-mass spectrometry (GC-MS) analysis revealed 34 and 22 volatile compounds in the leaf and root extracts, respectively. In the leaf extract, the major components identified were 2-pentanone, 4-methoxy-4-methyl-; 1,2,4-butanetriol; 9,12,15-octadecatrienoic acid, (Z,Z,Z)-; n-hexadecanoic acid; neophytadiene; and 5-hydroxymethylfurfural. Conversely, the root extract predominantly contained 2-pentanone, 4-hydroxy-4-methyl-, and 5-hydroxymethylfurfural. Furthermore, the antioxidant capacity was evaluated using the ABTS radical scavenging assay. The leaf and root acetone extracts exhibited IC_{50} values of 29.82±1.08 µg/mL and 52.46±2.51 µg/mL, respectively. These findings suggest potential applications for *C. laxum* in pharmaceutical and food industries and warrant further investigation into its bioactive properties and potential utilization.

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INTRODUCTION

The genus *Chlorophytum* (Liliaceae) comprises over 215 species widely distributed across pantropical regions, including China, India, Australia, and Africa (Li *et al.*, 1990; Dabur *et al.*, 2007; Katoch *et al.*, 2010; Chao *et al.*, 2017; Vakele *et al.*, 2022). While primarily cultivated for their ornamental value (Kaushik, 2005), species within this genus have garnered significant attention for their medicinal properties. However, research on the chemical composition and biological activities of the *Chlorophytum* genus has predominantly focused on two species: *Chlorophytum comosum* and *Chlorophytum borivilianum*.

C. comosum has been recognized for its phytoremediation capabilities, demonstrating the ability to purify indoor air by absorbing various hazardous gases, including carbon monoxide, formaldehyde, xylene, and benzene (Li *et al.*, 2021; Khan *et al.*, 2023). In addition, some studies have documented the cytotoxic activity of *C. comosum*. For example, the ethanolic root extract exhibited significant cytotoxicity against HepG2 and Caco-2 cell lines (Vakele *et al.*, 2022). The aqueous root and leaf extracts showed antiproliferative effects on lung cancer (A549) and breast cancer (MCF-7) cell lines (Adhami *et al.*, 2021). Methanolic leaf and root extracts demonstrated high apoptotic activity in leukemia K562 cells (Padayachee *et al.*, 2021), while the water fraction of the methanolic leaf extract also showed cytotoxicity towards human cervical epithelioid carcinoma (HeLa) cells (Rzhepakovsky *et al.*, 2022). Antioxidant activity of methanolic extracts from tubers (Deore *et al.*, 2015) and leaves (Rzhepakovsky *et al.*, 2022) of *C. comosum* has also been reported.

Studies on *C. borivilianum* have revealed its positive effects on animal models. Aqueous root extract has been shown to prevent deterioration of testicular function in mice and preserve human sperm function under hydrogen peroxide-induced oxidative stress (Giribabu *et al.*, 2014a; Mararajah *et al.*, 2024). Treatment with *C. borivilianum* root aqueous extract maintained near-normal levels of blood glucose, insulin, and lipids in diabetic rats, while preventing oxidative stress-induced pancreatic damage (Giribabu *et al.*, 2014b). *C. borivilianum* root powder demonstrated hypolipidemic and hypocholesterolemic effects in mice by increasing fecal cholesterol excretion and

endogenous cholesterol conversion to bile acid (Visavadiya & Narasimhacharya, 2007). Another study reported that *C. borivilianum* root extract significantly increased the activity of reduced glutathione, catalase, and superoxide dismutase, while decreasing hepatic MDA levels and tumor incidence in mice (Kumar *et al.*, 2010).

A few other *Chlorophytum* species have also demonstrated different biological activities. Ethanolic extract of *C. tuberosum* dried roots exhibited antioxidant activity (Chittama *et al.*, 2016). Ethyl acetate extract of *C. alismifolium* showed antihyperglycemic activity in type 2 diabetes by increasing insulin secretion (Abubakar *et al.*, 2021), while its methanol extract demonstrated antinociceptive activity (Abubakar *et al.*, 2020). Hexane extract of *C. inornatum* inhibited fast-growing mycobacteria, including *M. fortuitum*, *M. smegmatis*, *M. phlei*, and *M. aurum* (O'Donnell *et al.*, 2006).

Chlorophytum laxum, besides its use in decoration, is a traditional medicinal plant used by Kani tribes in Kerala, India, for treating inflammation, insect bites, snake bites, swelling, pain, diarrhea, and dysentery (Arundhathy & Suja, 2018). Studies on the chemical composition and biological activities of *C. laxum* are very limited, with only reports on the anti-inflammatory activity of ethanolic tuber extract (Arundhathy & Suja, 2018) and antibacterial activity of acetone extract against *P. aeruginosa* (Dabur *et al.*, 2007). Therefore, in order to further explore its potential applications, this study aims to investigate the phytochemical profile and antioxidant properties of acetone extracts from *C. laxum* roots and leaves for the first time.

MATERIALS AND METHODS

Plant Material

The fresh roots and leaves of *Chlorophytum laxum* R. Br. were collected from Binh Chau - Phuoc Buu Nature Reserve, Xuyen Moc District, Ba Ria-Vung Tau Province, Vietnam (Figure 1).

Screening of Primary Phytochemicals of the Acetone Extract

The fresh roots and leaves of *C. laxum* were delicately washed, dried at 50 □, and ground into powder using a grinder. For the extraction process, 0.5 g of the powdered sample was immersed in 99% acetone at a ratio of 1:30 (w/v) for 8 hours. Following extraction, the supernatant (designated as fraction 1) was separated via filtration. This extraction procedure was repeated twice on the residual material under identical conditions, yielding fractions 2 and 3. To facilitate a comprehensive qualitative analysis of bioactive constituents, all fractions were combined to obtain the final extract. Phytochemical screening was subsequently performed as Table 1.

Determination of Volatile Compounds

For the extraction process, 50 g of the powder sample was soaked in 250 mL of acetone 99% for 72 hours at room temperature.

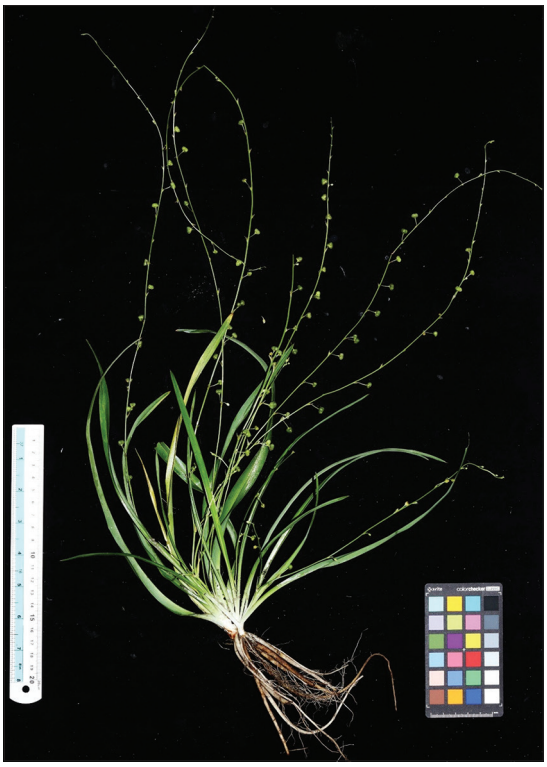

Figure 1: Morphology of *Chlorophytum laxum*

Table 1: Preliminary phytochemical screening for *Chlorophytum laxum*

Metabolites	Roots	Leaves
Phenols	+	+++
Tannins	+	+++
Flavonoids	+	+++
Coumarins	-	-
Alkaloids	+	+++
Terpenoids	+	++
Steroids	+	++
Saponins	+	-

(-) absent, (+) low quantities, (++) moderate quantities, (+++) high quantities

The extracts were filtered, and the process was repeated twice for the residual material. The combined filtrate was concentrated at low heat (45 □) under reduced pressure to ensure complete elimination of residual acetone.

Chemical composition analysis of the acetone extract was performed using a TRACE 1310 Gas Chromatograph coupled with an ISQ 7000 Mass Spectrometer (Thermo Fisher Scientific, USA) with the DB-5MS column used as stationary phase. Helium was used as carrier gas with 1.2 mL/min flow rate. The injector was held at 250 □ with 36 mL/min flow rate, 30:1 split ratio, and splitless time 1 minute. The oven was programmed as: 80 □ for 5 minutes, increased (20 □/min) to 280 □, 280 □ for 10 minutes. The ionization voltage was 70 eV, and the source temperature was 250 □. The acquisitions scan range was 29-650 m/z at a frequency of 2 scans/sec and chemical compositions were determined based on comparison with the NIST 2017 library.

Determination of Antioxidant Activity of Extract

ABTS (2,29-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid)) radical scavenging property of the root and leaf acetone extracts were identified as described by Maeng *et al.* (2017). First, a mixture of 7.0 mM ABTS and 2.45 mM $K_2S_2O_8$ was prepared and incubated at 37 ⍰ in the dark for 18 hours to form solution A. Then, 0.1 mL of the extract was added to 3 mL of solution A. This new mixture was diluted to a final volume of 5.0 mL with acetone, incubated in the dark for 15 minutes, and subjected to absorbance measurement at 734 nm by a UVS 2800 spectrophotometer (Labome, USA). Ascorbic acid was used as the reference standard, and a standard curve for ascorbic acid was generated. The concentration of the sample was determined using this standard curve and expressed in µg/mL ascorbic acid equivalent.

RESULTS AND DISCUSSION

Primary Phytochemicals of Acetone Extracts from *C. laxum*

The qualitative phytochemical analysis of *C. laxum* acetone extracts are presented in Table 2. Of the eight bioactive compounds assessed (phenols, tannins, flavonoids, coumarins, alkaloids, terpenoids, steroids, and saponins), the root extract contained seven constituents, with coumarins being the sole exception while both coumarins and saponins were found absent in the leaf extract.

Volatile Compounds of the Leaf Acetone Extract

The volatile compound profile of the acetone extract from *C. laxum* leaves is presented in Figure 2 and Table 3, revealing 34 distinct compounds. The most abundant compounds included 2-pentanone, 4-methoxy-4-methyl-; 1,2,4-butanetriol; 9,12,15-octadecatrienoic acid, (Z,Z,Z)-; n-hexadecanoic acid; neophytadiene; and 5-hydroxymethylfurfural.

Table 2: Methods for phytochemical screening of *Chlorophytum laxum*

Phytochemical	Reaction composition	Positive observation
Phenolic and tannin	2 mL extract+2 mL H_2O+2-3 drops FeCl3 5%	Color change to brownish- or blackish green (Deka *et al.*, 2017)
Alkaloid	2 mL extract+3-4 drops Wagner reagent	Formation of reddish brown precipitate (Bodi *et al.*, 2014)
Flavonoid	2 mL extract+2 mL Pb $(COOH)_2$ 10%	Formation of yellow precipitate (Nguyen *et al.*, 2017)
Saponin	2 mL extract+10 mL H20+2-minute boiling	Formation of foam (Odebiyi & Sofowora, 1978)
Terpenoid and steroid	5 mL extract+2 mL chloroform+3 mL H_2SO_4 concentrate	Color change to brownish red (Llauradó *et al.*, 2013)
Coumarin	2 mL extract+3 mL NaOH (10%)	Color change to yellow or dark yellow (Ngan *et al.*, 2017)

Volatile Compounds of the Root Acetone Extract

Gas chromatographic analysis of the acetone extract from *C. laxum* roots revealed 22 distinct volatile compounds, as illustrated in Figure 3 and detailed in Table 4. The most prominent chemical components were identified as 2-pentanone, 4-hydroxy-4-methyl-; 5-hydroxymethylfurfural; 2-furanmethanol; and 4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-.

Antioxidant activity of *C. laxum* acetone extracts

Both leaf and root acetone extracts of *C. laxum* exhibited ABTS radical scavenging activity in a concentration-dependent manner (Figure 4a & b). The IC_{50} values for *C. laxum* leaf and root acetone extracts were determined to

Table 3: Phytochemical profile of acetone leaf extract of *Chlorophytum laxum*

S. No.	RT	Compounds	Formula
1	2.16	2-Pentanone, 4-methoxy-4-methyl-	$C_7H_{14}O_2$
2	5.59	Benzeneacetaldehyde	C_8H_8O
3	5.79	Furaneol	$C_6H_8O_3$
4	6.94	2-Butenamide, N, N-diethyl-3-methyl-	$C_9H_{17}NO$
5	7.31	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-	$C_6H_8O_4$
6	8.35	5-Hydroxymethylfurfural	$C_6H_6O_3$
7	10.36	1,2,4-Butanetriol	$C_4H_{10}O_3$
8	11.24	3,4-Altrosan	$C_6H_{10}O_5$
9	11.84	Ethyl α-d-glucopyranoside	$C_8H_{16}O_6$
10	12.17	(E)-4-(3-Hydroxyprop-1-en-1-yl)-2-methoxyphenol	$C_{10}H_{12}O_3$
11	12.41	6-Hydroxy-4,4,7a- trimethyl-5,6,7,7a-tetrahydrobenzofuran-2 (4H)-one	$C_{11}H_{16}O_3$
12	12.69	Neophytadiene	$C_{20}H_{38}$
13	12.82	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	$C_{20}H_{40}O$
14	12.92	Neophytadiene	$C_{20}H_{38}$
15	13.15	Hexadecanoic acid, methyl ester	$C_{17}H_{34}O_2$
16	13.34	n-Hexadecanoic acid	$C_{16}H_{32}O_2$
17	14.04	9,12,15-Octadecatrienoic acid, methyl ester, (Z, Z, Z)-	$C_{19}H_{32}O_2$
18	14.08	Phytol	$C_{20}H_{40}O$
19	14.19	10(E),12(Z)-Conjugated linoleic acid	$C_{18}H_{32}O_2$
20	14.23	9,12,15-Octadecatrienoic acid, (Z, Z, Z)-	$C_{18}H_{30}O_2$
21	14.31	Octadecanoic acid	$C_{18}H_{36}O_2$
22	15.95	Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl) ethyl ester	$C_{19}H_{38}O_4$
23	17.05	9,12-Octadecadienoic acid (Z, Z)-, 2-hydroxy-1-(hydroxymethyl) ethyl ester	$C_{21}H_{38}O_4$
24	17.10	Linolenic acid, 2-hydroxy-1-(hydroxymethyl) ethyl ester (Z, Z, Z)-	$C_{21}H_{36}O_4$
25	21.83	Vitamin E	$C_{29}H_{50}O_2$
26	22.93	Phenol, 4,4'-(tetrahydro-1H,3H-furo[3,4-c] furan-1,4-diyl) bis[2-methoxy-	$C_{20}H_{22}O_6$
27	23.25	Dehydrodiconiferyl alcohol	$C_{20}H_{22}O_6$
28	23.57	Ergost-5-en-3-ol, (3β)-	$C_{28}H_{48}O$
29	24.11	Stigmasterol	$C_{29}H_{48}O$
30	25.02	Neotigogenin	$C_{27}H_{44}O_3$
31	25.39	Tigogenin	$C_{27}H_{44}O_3$
32	27.34	dl-α-Tocopherol	$C_{29}H_{50}O_2$
33	28.47	(25R)-5α-Spirostan-2α,3β-diol	$C_{27}H_{44}O_4$
34	28.83	Rockogenin	$C_{27}H_{44}O_4$

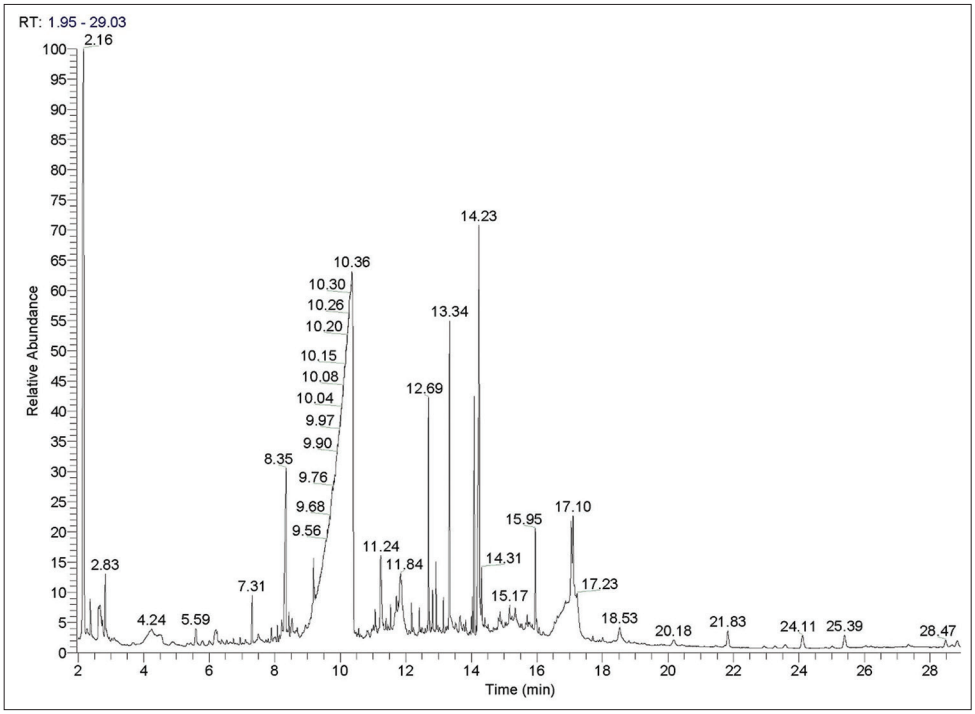

Figure 2: GC chromatogram of acetone leaf extract of *Chlorophytum laxum*

Table 4: Phytochemical profile of acetone root extract of *Chlorophytum laxum*

S. No.	RT	Compounds	Formula
1	2.18	2-Pentanone, 4-hydroxy-4-methyl-	$C_6H_{12}O_2$
2	2.36	2-Furanmethanol	$C_5H_6O_2$
3	2.76	Ethanol, 2-butoxy-	$C_6H_{14}O_2$
4	3.16	2-Cyclopenten-1-one, 2-hydroxy-	$C_5H_6O_2$
5	5.17	2-Cyclopenten-1-one, 2-hydroxy-3-methyl-	$C_6H_8O_2$
6	6.04	2,5-Dimethylfuran-3,4 (2H,5H)-dione	$C_6H_8O_3$
7	6.38	Thymine	$C_5H_6N_2O_2$
8	6.45	Furaneol	$C_6H_8O_3$
9	6.80	6-Azabicyclo[3.2.1]octane	$C_7H_{13}N$
10	6.87	Phenylethyl Alcohol	$C_8H_{10}O$
11	7.05	2 (3H)-Furanone, 5-acetyldihydro-	$C_6H_8O_3$
12	7.20	Ethanamine, N-ethyl-N-nitroso-	$C_4H_{10}N_2O$
13	7.40	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-	$C_6H_8O_4$
14	8.51	5-Hydroxymethylfurfural	$C_6H_6O_3$
15	11.59	Benzenepropanoic acid, 4-hydroxy-	$C_9H_{10}O_3$
16	13.31	n-Hexadecanoic acid	$C_{16}H_{32}O_2$
17	13.33	Dibutyl phthalate	$C_{16}H_{22}O_4$
18	13.63	1-Naphthalenecarbonitrile, 8-amino-	$C_{11}H_8N_2$
19	22.92	Phenol, 4,4'-(tetrahydro-1H,3H-furo[3,4-c] furan-1,4-diyl) bis[2-methoxy-	$C_{20}H_{22}O_6$
20	23.26	Dehydrodiconiferyl alcohol	$C_{20}H_{22}O_6$
21	25.38	Neotigogenin	$C_{27}H_{44}O_3$
22	28.46	(25R)-5α-Spirostan-2α,3β-diol	$C_{27}H_{44}O_4$

be 29.82±1.08 µg/mL and 52.46±2.51 µg/mL, respectively (Figure 4c). The root extract demonstrated lower antioxidant capacity compared to the leaf extract, which may be attributed to the higher abundance of phenolic compounds in the leaf extract (Table 1).

DISCUSSION

A few studies have reported on the preliminary phytochemistry of different extracts from *C. laxum* and other *Chlorophytum* species. Deore *et al.* (2015) demonstrated that both aqueous and ethanolic extracts of *C. laxum* tubers contained carbohydrates, alkaloids, flavonoids, triterpenoids, while saponins and saponin glycoside only present in the aqueous extract. Similar results were obtained for the same extracts from other *Chlorophytum* species with exceptions such as no detection of alkaloids in *C. comosum* and *C. tuberosum*, no detection of triterpenoids in *C. borivilianum* and *C. arundinaceum*, and no tannins were recorded in *C. laxum* and *C. tuberosum* (Deore *et al.*, 2015). In addition, the petroleum ether-methanol extract of *C. borivilianum* leaves and stems consisted of alkaloids, saponin glycosides, glycosides, tannins, and steroids (Chakraborthy *et al.*, 2014). Moreover, alkaloids, carbohydrates, reducing sugars, starch, proteins, tannins, and saponins have been reported as the preliminary phytochemistry in the methanol extract of *C. kolhapurens* and *C. bharuchae* (Sharma & Thakare, 2019).

Building upon these phytochemical screenings, a closer comparison of volatile compound profiles reveals notable similarities between the acetone leaf extract of *C. laxum* and the methanolic and ethyl acetate extracts of *C. comosum*, suggesting the presence of conserved bioactive constituents across species. Both extracts contain n-hexadecanoic acid; hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester; 9,12-octadecadienoic acid (Z,Z)-; stigmasterol;

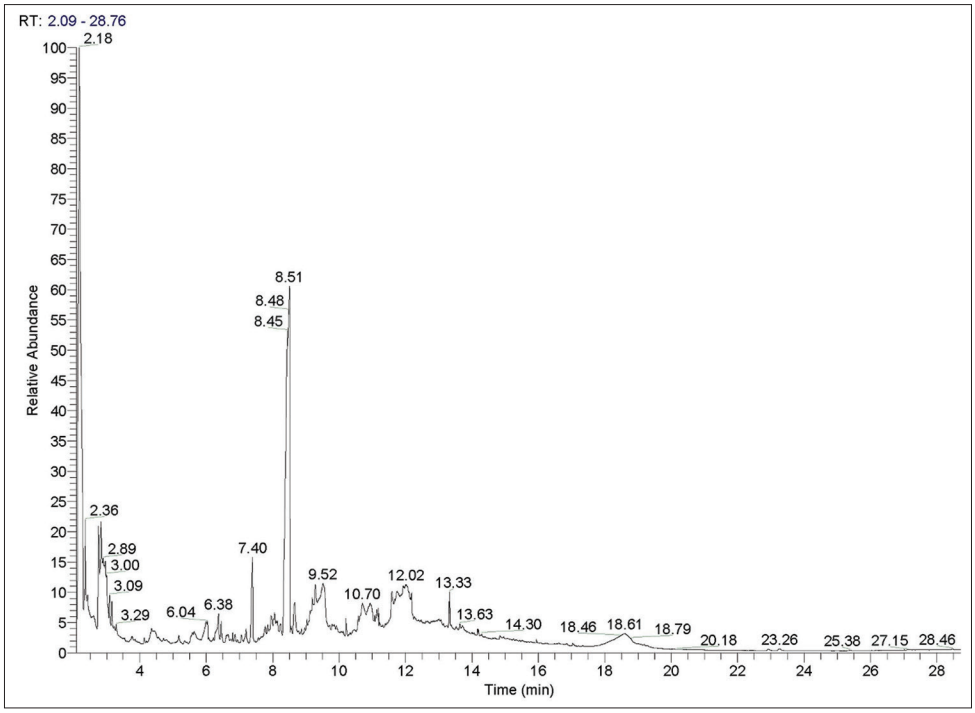

Figure 3: GC chromatogram of acetone root extract of *Chlorophytum laxum*

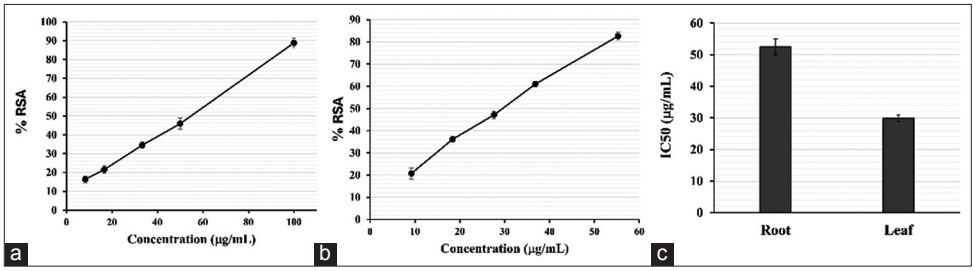

Figure 4: ABTS radical scavenging activity (% RSA) of *Chlorophytum laxum* acetone extracts from a) root, b) leaf, and c) the corresponding IC_{50}

octadecanoic acid; 3,7,11,15-tetramethyl-2-hexadecen-1-ol; and 5-hydroxymethylfurfural (Rzhepakovsky *et al.*, 2022). In addition, in the main components of *C. comosum* root and leaf ethyl acetate extracts, n-hexadecanoic acid, octadecanoic acid were also found along with other compounds such as 9(E),11(E)-conjugated linoleic acid, acetamide, N-[4-(trimethylsilyl)phenyl]-, oleic acid in root ethyl acetate extracts and 9,12,15-octadecatrienoic acid, 1-(1,5- dimethyl-4-hexenyl)-4-methyl-, fumaric acid, 2-methoxyphenyl 2,3-dichlorophenyl ester, and γ-sitosterol in leaf ethyl acetate extract (Kavya *et al.*, 2024). The obtained results indicate some commonality in the phytochemical profiles across different *Chlorophytum* species. In contrast, the aqueous extract of *C. comosum* leaves has been reported to contain 34 compounds, with major components including 2-tert-butyl-5-(hydroxymethyl)-5-methyl-1,3-dioxolan-4-one; 3-(hydroxymethyl)phenol; and butoxy(trimethyl)silane (Adhami *et al.*, 2021). Furthermore, the ethyl acetate extract of whole *C. alismifolium* plants has been shown to contain volatile compounds such as isothiazole; isoxazolidine; 2(3H)-naphthalenone; N-ethylformamide; and 1,7-octadiyne (Abubakar *et al.*, 2021).

Notably, 2-pentanone, 4-hydroxy-4-methyl- has also been reported in the methanol tuber extract of *C. alismifolium* (Abubakar *et al.*, 2020). In contrast, analysis of the aqueous root extract from *C. comosum* identified 17 compounds, with bis(2-ethylhexyl) hexanedioate; neotigogenin; yuccagenon; and dodecane being the most abundant (Adhami *et al.*, 2021). These findings highlight the diversity of volatile compounds present in various *Chlorophytum* species and emphasize the potential influence of both the species and the extraction solvent on the resulting phytochemical profile. Such variations may contribute to the different biological activities observed among *Chlorophytum* species and extracts.

Antioxidant effects of some extracts from other *Chlorophytum* species were also reported. The ABTS free radical scavenging activities of various fractions of *C. comosum* methanolic leaf extract, expressed as mg Trolox Equivalents (TEs)/mL, were 0.27±0.01, 0.36±0.01, 0.81±0.05, and 0.27±0.02 for n-hexane, chloroform, n-butanol, and water fractions, respectively (Rzhepakovsky *et al.*, 2022). Additionally, the ethyl acetate extracts of *C. comosum* root and leaf exhibited ABTS free

radical scavenging activity, with IC_{50} values of 126.24±0.13 µM and 264.41±0.08 µM, respectively, which are comparable to those of the *C. laxum* acetone extracts in this study (Kavya *et al.*, 2024). For DPPH free radical scavenging, *C. borivilianum* aqueous root extract demonstrated antioxidant potential with an IC_{50} value of 46.37 µg/mL (Vyas *et al.*, 2022). A comparative analysis of DPPH free radical scavenging activities among methanolic tuber extracts from various *Chlorophytum* species also revealed a hierarchical order of antioxidant potency. *C. arundinaceum* exhibited the strongest activity, followed by *C. comosum* and *C. tuberosum*, which demonstrated equivalent potency. *C. borivilianum* showed lower activity while *C. laxum* probably exhibited the least potent (Deore *et al.*, 2015). While studies on the antioxidant activity of *C. laxum* are limited, particularly regarding acetone extracts, the observed radical scavenging ability of both extracts can be partially attributed to the presence of phenolic compounds in the leaf and root (Table 1), as well as the presence of 5-hydroxymethylfurfural and n-hexadecanoic acid (Tables 3 & 4), which are known for their antioxidant properties (Zhao *et al.*, 2013; Ullah *et al.*, 2020; Ganesan *et al.*, 2022).

CONCLUSION

The phytochemical screening of *Chlorophytum laxum* acetone extracts from leaves and roots revealed the presence of diverse bioactive compounds, including phenols, tannins, flavonoids, alkaloids, terpenoids, and steroids. Gas chromatography-mass spectrometry analysis further elucidated the volatile component profiles of these extracts, providing a comprehensive characterization of their chemical composition. Moreover, both leaf and root extracts demonstrated notable antioxidant properties, as evidenced by their ABTS radical scavenging capacity. Further research is warranted to isolate and characterize specific bioactive constituents, elucidate their mechanisms of action, and explore additional biological activities. Such investigations could pave the way for the development of novel natural products derived from *C. laxum*, potentially expanding its utility beyond ornamental purposes for broader applications, particularly in the pharmaceutical and food industry.

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