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Research Article

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Exploring the Bioactive Phytochemicals of *Tinospora cordifolia* via Gas Chromatography-mass Spectrometry: Implications for Drug Development and Functional Foods

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Abstract

This study investigates the phytochemical profile of *Tinospora cordifolia* methanolic stem extract (TCMSE), a medicinal plant valued in Ayurveda, using gas chromatography-mass spectrometry (GC-MS) analysis to correlate identified compounds with their bioactive properties. TCMSE, known for its immunomodulatory, antioxidant, and anti-inflammatory activities, was analyzed to identify volatile and semi-volatile phytochemicals. The analysis revealed a complex mixture of bioactive compounds, including n-Hexadecanoic acid (Palmitic acid), Benzofuran, and 1,2,3,4-Butanetetrol. Further investigation included antioxidative analysis via 2,2-Diphenyl-1-picrylhydrazyl (DPPH) assay of TCMSE, demonstrating significant free radical scavenging activity. Additionally, the anticancer potential of TCMSE was evaluated via 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay against K562 and HeLa cancer cell lines, showing promising cytotoxic activity. These findings support the traditional use of TCMSE and pave the way for its application in modern drug development and natural health formulations. The identified phytochemicals offer potential for use in the food, nutraceutical, and pharmaceutical industries, warranting further research into novel drug formulations and dietary products.

Keywords

Antioxidant, Anticancer, Bioactive molecules, Gas chromatography-mass spectrometry, Phytochemicals, *Tinospora cordifolia*

Introduction

T. cordifolia, commonly known as Guduchi or Giloy, is a well-known medicinal plant in traditional Ayurveda, valued for its diverse pharmacological properties [1, 2]. It has been extensively used in traditional medicine for its immunomodulatory, antioxidant, anti-inflammatory, and antimicrobial activities [1, 2]. The therapeutic potential of *T. cordifolia* is attributed to the presence of various bioactive phytochemicals, including alkaloids, terpenoids, flavonoids, steroids, and glycosides [3, 4]. Understanding the chemical composition of this plant is essential for validating its medicinal applications and exploring new bioactive compounds for pharmaceutical, food and nutraceutical industries [5, 6].

GC-MS is a powerful analytical technique widely employed for the identification and characterization of volatile and semi-volatile phytochemicals in medicinal plants [7]. This technique provides detailed insights into the chemical constituents of *T. cordifolia*, allowing the detection of bioactive compounds that contribute to its medicinal properties. GC-MS analysis enables the identification of key secondary metabolites such as terpenes, fatty acids, alkaloids, and phenolic compounds, which play crucial roles in antioxidant, antimicrobial, and anti-inflammatory activities [8, 9].

Several studies have reported the presence of bioactive compounds in *T. cordifolia* that exhibit pharmacological significance [10-12]. For instance, alkaloids like berberine and magnoflorine have been found to possess antimicrobial and immunomodulatory effects [13, 14], while diterpenoids and lactones contribute to their hepatoprotective and antidiabetic properties [15, 16]. Additionally, fatty acids and sterols present in *T. cordifolia* may enhance its antioxidant potential, further supporting its traditional use in managing oxidative stress-related disorders [17, 18]. The presence of phytochemicals such as berberine, magnoflorine, and diterpenoids further supports its role in enhancing gut health, metabolic function, and overall well-being when incorporated into food formulations [19, 20]. The identification of such bioactive constituents through GC-MS analysis strengthens the scientific validation of *T. cordifolia* as a potent medicinal plant.

This study aims to analyze the phytochemical profile of *T. cordifolia* using GC-MS and correlate the identified compounds with their known bioactive properties. By understanding the chemical composition, this research provides a foundation for future pharmacological investigations and potential therapeutic applications. The findings will contribute to the growing body of evidence supporting the medicinal value of *T. cordifolia*, paving the way for its use in modern drug development and natural health formulations. The objectives of the study were: (i) preparation of TCMSE, (ii) GC-MS analysis of TCMSE for bioactive molecules, (iii) antioxidative analysis of TCMSE via DPPH free radical scavenging assay, and (iv) anticancer potential of TCMSE via MTT assay against K562 and HeLa cancer cell lines.

Materials and Methods

Materials

All chemicals used and 0.22 µm polyvinylidene fluoride (PVDF) syringe filters were purchased from HiMedia Laboratories Pvt. Ltd. (India). Chemicals used for GC-MS analysis were purchased from Sigma-Aldrich (India) of GC-MS grade. All chemicals were >99% pure. Distilled water (pH was in the range of 6.6 - 7.4) was used for the work.

Plant (stem) collection

Stem parts of *T. cordifolia* (wild variety) were collected from the local neighborhood, Warangal (Telangana, India) during February 2024. The plant samples were identified by Dr. Madhusudhan Kairamkonda, Department of Bioscience, Rajiv Gandhi University of Knowledge Technologies (Telangana, India). The stem parts were brown in color. *T. cordifolia* stem parts were washed several times with distilled water, cleaned with ethanol and later shade dried under sterile conditions by keeping them at room temperature. Dried stem parts were made into fine powder using domestic mixer. Figure 1 presents the schematic representation of the reported work.

Extract preparation

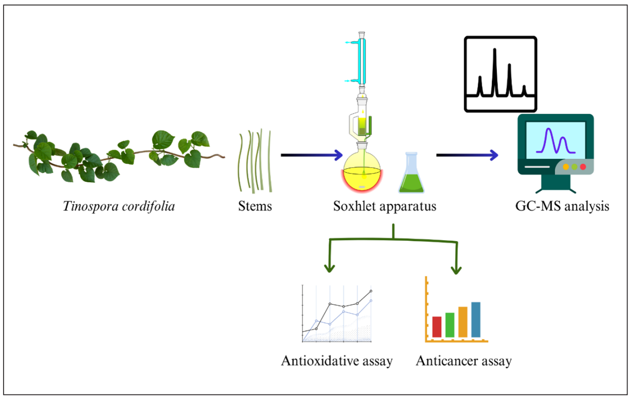

Figure 1: Schematic representation of the work reported.

Dried (100 g) stem powder was placed in a Soxhlet apparatus. 800 ml of methanol was used for the extraction, which was carried out for 48 hr at a temperature that did not go above the solvent's boiling point. The extract was passed through a 0.22 µm PVDF filters. Methanol extract was obtained by vacuum-concentrating the resultant solution until it was completely dry. To be used later, the extract was kept in a refrigerator at 4 °C.

GC-MS analysis

TCMSE was qualitatively investigated by using the JEOL GCMATE II (GC-MS) with data system is a high resolution, double focusing instrument. Maximum resolution: 6000 and 1500 Daltons were the maximum calibrated mass. Source options: Electron impact and chemical ionization. Experimental condition, GC equipped with a capillary column (30 m × 0.25 mm (inner diameter) and film thickness 0.25 µm). Flow rate of mobile phase (carrier gas: Helium) was set at 1.0 ml/min. In the GC part, temperature program (oven temperature) was 40 °C raised to 350 °C at 5 °C/min and injection volume was 1 µl. The exact name and molecular weight of unknown compounds were found by comparing their mass spectrum with the reference spectrum available in the NIST: GC/MS library.

Antioxidative assay

The DPPH assay is a widely used antioxidant test that measures the free radical scavenging ability of compounds based on their reaction with the stable DPPH radical. The reduction of DPPH, indicated by a color change from purple to yellow, reflects the antioxidant capacity of the tested sample. The assay was performed based on previously published method without any modifications [21].

Anticancer assay

The MTT assay is a colorimetric method used to assess cell viability based on the reduction of MTT dye to formazan crystals by metabolically active cells. The intensity of the purple color is directly proportional to the number of viable cells. The assay was performed based on previously published method without any modifications [21].

Statistical analysis

All experiments were conducted in triplicate (n = 3), and data were expressed as the mean (n = 3) ± standard deviation (error bars). Statistical significance ($p < 0.05$) was evaluated using student's t-test, while graphical representation of the results was performed using Microsoft Excel 2021.

Results and Discussion

GC-MS analysis

The GC-MS analysis of the TCMSE reveals a complex profile of bioactive compounds with significant therapeutic potential (Figure 2) [22]. This analysis is crucial for understanding the pharmacological properties of the plant, which is widely used in traditional medicine. The TCMSE has been shown to contain a variety of phytochemicals, including terpenoids, steroids, glycosides, flavonoids, and phlobatannins, which contribute to its medicinal properties [3, 4, 11]. The GC-MS method is instrumental in identifying these compounds, providing insights into their potential health benefits [23, 24].

Table 1 presents a detailed analysis of chemical compounds identified in a sample through GC-MS technique. It includes parameters such as retention time, peak area, area percentage, peak height, height percentage, and area-to-height ratio (A/H) for 52 different compounds. The retention time indicates the time taken for each compound to elute through the chromatographic column, while area % and height % reflect the relative abundance of each compound in the sample.

Key findings include the major components, which are determined based on their high area percentages. The most abundant compound in the sample is palmitic acid at a 10.13% area, followed closely by benzofuran, 2,3-Dihydro- (9.97%) and 1,2,3,4-Butanetetrol, [S-(R*,R*)]- (9.30%). Additional significant compounds include 1-Hexacosanol (6.27%), phenol, 3,5-bis(1,1-dimethylethyl)- (6.03%), and 2-Bromotetradecane (5.34%). These compounds could play crucial roles in the biological or chemical properties of the analyzed sample. The presence of fatty acids and esters, such as hexadecanoic acid, methyl ester (3.40%), 9,12-Octadecadienoic acid, methyl ester (1.56%), and Methyl stearate (1.41%), suggest lipid-based components, possibly contributing to antioxidant, antimicrobial, or pharmacological activities. Additionally, the detection of aromatic compounds like benzyl benzoate (0.54%) and azulene (1.73%) indicates the presence of bioactive molecules. The A/H ratio values vary among compounds, suggesting differences in peak shape and signal intensity, which can provide insights into compound stability and detection efficiency. Some compounds with relatively low peak areas but high A/H ratios, such as hexadecanenitrile (3.18) and 2-propenoic acid, 3-(4-methoxyphenyl)- (3.30), may indicate higher response factors in the detection system. Overall, the chromatographic profile suggests a complex mixture of bioactive compounds, including fatty acids, esters, lactones, nitriles, and aromatic compounds. This composition could be relevant for pharmaceutical, cosmetic, food, or industrial applications.

In literature, GC-MS analysis identified several fatty acids such as palmitic acid, octadecanoic acid, and 9,12-Octadecadienoic acid (Z, Z), which are known for their antioxidant and anti-inflammatory properties [23, 24]. Compounds like stigmasterol and stigmastane-3,6-dione were detected, which have been associated with anti-inflammatory and cholesterol-lowering effects [25]. The presence of flavonoids, such as (−) epicatechin, contributes to the antioxidant activity of the extract, which is crucial for combating oxidative stress [26]. The analysis also revealed the presence of compounds like phytol and benzenepropanoic acid, which have potential anticancer and antimicrobial activities [23].

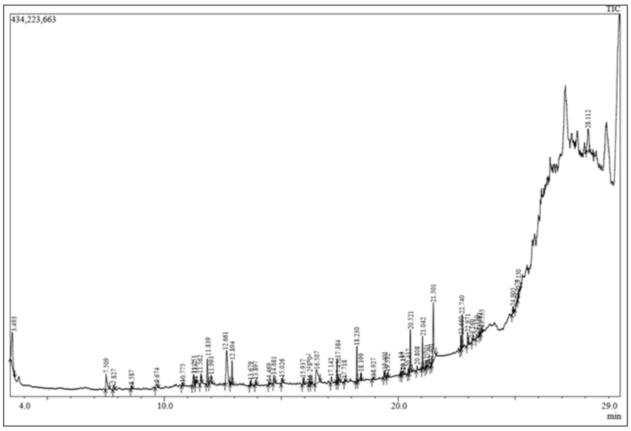

Figure 2: GC-MS spectra of TCMSE.

Antioxidative assay

The DPPH assay is a widely used method to evaluate the antioxidant activity of plant extracts, including the TCMSE. This essay measures the ability of antioxidants to scavenge free radicals, which is indicative of the extract's potential to mitigate oxidative stress [27]. The TCMSE has been shown to possess significant antioxidant activity, as evidenced by its performance in the DPPH assay. The antioxidant activity of TCMSE is attributed to its rich phytochemical profile, including phenols, flavonoids, tannins, and alkaloids [28]. These compounds are known for their ability to neutralize free radicals and contribute to the overall antioxidant capacity of the extract [29]. The extract's high solubility in methanol allows for the effective extraction of these active components, enhancing its therapeutic potential [29].

Figure 3 presents the average values for TCMSE at different concentrations (20 - 100 µg/ml), showing a clear concentration-dependent increase. As the concentration of TCMSE increases, the measured values rise correspondingly, suggesting a direct relationship between concentration and response. This trend indicates that TCMSE exhibits increasing activity or effectiveness with higher doses, which could be relevant in assays such as antioxidant or enzymatic activity tests. The sharp rise at higher concentrations, particularly from 60 µg/ml onward, may suggest an enhanced effect or a threshold beyond which the response becomes more pronounced. The TCSME demonstrated significant DPPH radical scavenging activity. One study reported an IC_{50} value of 52.80 µg/ml, which, while not as potent as the standard ascorbic acid (IC_{50} of 4.12 µg/ml), still indicates a strong antioxidant potential [30]. A study highlighted the methanolic extract's high antioxidant potency, with a DPPH radical scavenging activity of 98.13% [31].

While the TCMSE shows promising antioxidant activity,

Table 1: GC-MS analysis of TCMSE.

Peak#	Retention time	Area	Area %	Height	Height %	A/H	Name
1	3.493	169455050	9.30	39060154	5.79	4.34	1,2,3,4-Butanetetrol, [S-(R*,R*)]-
2	7.509	60340925	3.31	17638780	2.62	3.42	Butyrolactone
3	7.827	13620418	0.75	5599458	0.83	2.43	6-Oxa-bicyclo[3.1.0]hexan-3-one
4	8.587	6803669	0.37	3596784	0.53	1.89	2,4-Dihydroxy-2,5-dimethyl-3(2H)-fu
5	9.674	20596026	1.13	4412906	0.65	4.67	Benzeneacetic acid, octyl ester
6	10.775	7958509	0.44	3437848	0.51	2.31	1-Cyclohexene-1-acetonitrile
7	11.251	35599871	1.95	12403349	1.84	2.87	Benzyl nitrile
8	11.337	9755728	0.54	4849766	0.72	2.01	2-(Cyclopenten-1-yl)acetic acid
9	11.562	23493097	1.29	10813308	1.60	2.17	1-Cyclohexene-1-carboxylic acid
10	11.839	65815411	3.61	28254162	4.19	2.33	Benzeneacetic acid, methyl ester
11	11.993	31458263	1.73	9428688	1.40	3.34	Azulene
12	12.661	181507464	9.97	37128886	5.51	4.89	Benzofuran, 2,3-dihydro-
13	12.894	53413170	2.93	27310646	4.05	1.96	Benzene, 1,3-bis(1,1-dimethylethyl)-
14	13.679	9849964	0.54	5328762	0.79	1.85	Indole
15	13.897	11874407	0.65	5452277	0.81	2.18	2-Methoxy-4-vinylphenol
16	14.469	5662821	0.31	2829321	0.42	2.00	Benzene, 1-methyl-4-[(1-methylethyl)]
17	14.681	21846358	1.20	9382772	1.39	2.33	n-Decanoic acid
18	15.026	14042656	0.77	6186637	0.92	2.27	DL-Proline, 5-oxo-, methyl ester
19	15.937	13968133	0.77	7352230	1.09	1.90	1-Chloroundecane
20	16.192	7920752	0.43	4165258	0.62	1.90	3,3-Dimethyl-hepta-4,5-dien-2-one
21	16.280	4926339	0.27	2714972	0.40	1.81	2,6-Difluorobenzoic acid, tridec-2-ynyl ester
22	16.507	109865734	6.03	15272287	2.27	7.19	Phenol, 3,5-bis(1,1-dimethylethyl)-
23	17.142	25881144	1.42	7047247	1.05	3.67	Dodecanoic acid
24	17.384	49542260	2.72	25684692	3.81	1.93	syn-Tricyclo[5.1.0.0(2,4)]oct-5-ene, 3,3,5,6,8,8-hexamethyl-
25	17.450	15413622	0.85	5978007	0.89	2.58	Diethyl Phthalate
26	17.718	5709631	0.31	3035630	0.45	1.88	5-Isopropenyl-2-methylcyclopent-1-enecarboxaldehyde
27	18.230	73472463	4.03	40140771	5.95	1.83	Octadecane, 1-iodo-
28	18.399	18865926	1.04	8981936	1.33	2.10	Tetradecane, 1-chloro-
29	18.927	5448916	0.30	2518320	0.37	2.16	Tridecanoic acid, 12-methyl-, methyl
30	19.401	17408965	0.96	7837089	1.16	2.22	Octadecanoic acid
31	19.532	9896665	0.54	5393703	0.80	1.83	Benzyl Benzoate
32	20.114	6291850	0.35	3707765	0.55	1.70	Neophytadiene
33	20.197	8426680	0.46	3953066	0.59	2.13	2-Pentadecanone, 6,10,14-trimethyl-
34	20.437	11082658	0.61	5889051	0.87	1.88	1,2-Benzenedicarboxylic acid, bis(2-methylpropyl) ester
35	20.521	97284321	5.34	48292794	7.16	2.01	2-Bromotetradecane
36	20.808	16180802	0.89	5086639	0.75	3.18	Hexadecanenitrile
37	21.042	61934289	3.40	37367740	5.54	1.66	Hexadecanoic acid, methyl ester
38	21.171	3309425	0.18	2015542	0.30	1.64	Ethanol, 2-(octadecyloxy)-
39	21.233	2342237	0.13	1742170	0.26	1.34	Benzoic acid, 2-benzoyl-, methyl ester
40	21.401	3961950	0.22	2766497	0.41	1.43	Dibutyl phthalate
41	21.501	184528716	10.13	66543985	9.87	2.77	Palmitic acid
42	22.680	28357248	1.56	15596835	2.31	1.82	9,12-Octadecadienoic acid, methyl ester
43	22.740	65109314	3.58	38142114	5.66	1.71	9,12,15-Octadecatrienoic acid, methyl ester
44	22.971	25604726	1.41	14197424	2.11	1.80	Methyl stearate
45	23.169	4808560	0.26	2919156	0.43	1.65	Nonyl tetradecyl ether
46	23.350	11935376	0.66	3614510	0.54	3.30	2-Propenoic acid, 3-(4-methoxyphenyl)
47	23.492	8884583	0.49	3389740	0.50	2.62	Tetrapentacontane, 1,54-dibromo-
48	23.555	10682948	0.59	5486299	0.81	1.95	Tetradecanamide
49	24.905	14379425	0.79	7725789	1.15	1.86	2-Propenoic acid, 3-(4-methoxyphenyl)
50	25.075	13379831	0.73	3701216	0.55	3.61	Nonadecyl pentafluoropropionate
51	25.150	27141397	1.49	7693461	1.14	3.53	1-Decanol, 2-hexyl-
52	28.112	114151337	6.27	25014459	3.71	4.56	1-Hexacosanol

it is important to consider the broader context of its use. The presence of various phytochemicals not only contributes to its antioxidant properties but also suggests potential applications in traditional medicine and the food industry as a natural antioxidant. However, the efficacy of the extract can vary based on factors such as extraction methods, plant part used, and environmental conditions affecting the plant's growth. Further research is needed to fully understand the mechanisms behind its antioxidant activity and to explore its potential therapeutic applications.

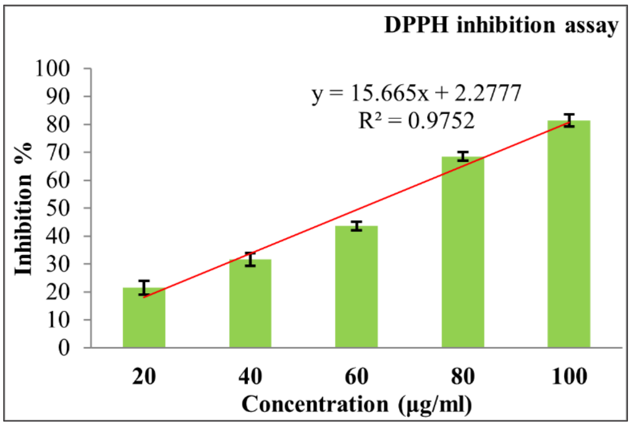

Figure 3: Antioxidative (DPPH radical scavenging) assay results.

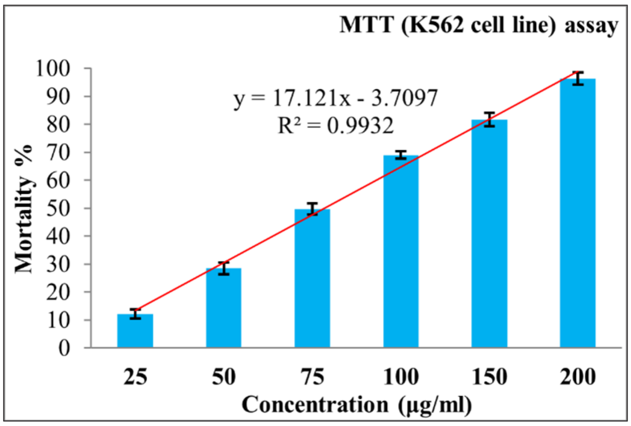

Figure 4: Anticancer assay results against K562 cancer cell line.

Anticancer assay

The *T. cordifolia*, commonly known as Guduchi or Amrita, has been extensively studied for its biological activities, particularly its cytotoxic properties. The MTT assay, a colorimetric assay for assessing cell metabolic activity [32], has been employed to evaluate the anti-proliferative effects of this extract on two (K562 and HeLa) cancer cell lines. The TCMSE has shown promising results, indicating its potential as a therapeutic agent in cancer treatment.

Figure 4 and figure 5 measured at different concentrations (25 - 200 µg/ml), showing a clear concentration-dependent increase. As the concentration increases, the corresponding values also rise, indicating a direct relationship between the two variables. This suggests that the TCMSE exhibits a progressive effect with increasing concentration, which could be relevant in assays measuring anticancer activity. Notably, the response shows a sharp increase up to 100 µg/ml, after which the rise becomes more gradual, particularly between 150 and 200 µg/ml. This pattern could indicate a nearing saturation point, where higher concentrations yield diminishing increases in effect.

The TCMSE demonstrated significant cytotoxic activity against several cancer cell lines. For instance, it exhibited an IC_{50} value of 59 ± 4.05 µg/ml against the MDA-MB-231 human breast cancer cell line, indicating potent anticancer properties [33]. In a study, the methanolic extract showed moderate cytotoxicity against Dalton's lymphoma ascites cell lines, with an IC_{50} value of 72.05 µg/ml, which was comparable to the reference drug cisplatin [34]. The dichloromethane fraction of *T. cordifolia* was found to be the most potent anti-proliferative fraction against HeLa cells, with an IC_{50} of 54.23 ± 0.94 µg/ml in the MTT assay [35].

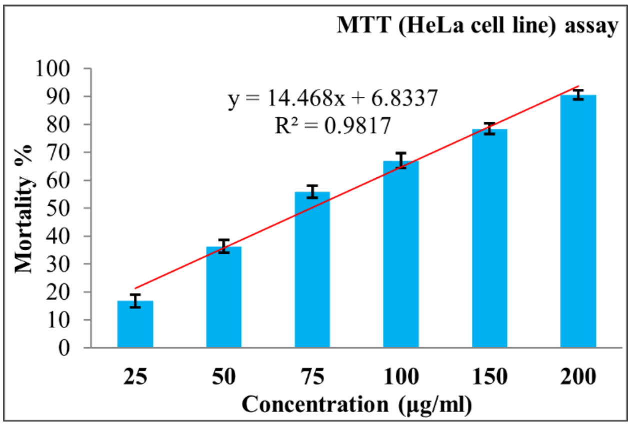

Figure 5: Anticancer assay results against HeLa cancer cell line.

Based on literature

Many earlier studies focused only on basic phytochemical screening (e.g., qualitative tests for alkaloids, flavonoids) without advanced characterization. Some GC-MS analyses lacked comprehensive compound identification or failed to correlate specific phytochemicals with bioactivity. This study employs high-resolution GC-MS (JEOL GCMATE II) with NIST library matching, identifying 52 bioactive compounds and linking them to pharmacological effects. Previous works used varied extraction methods (aqueous, ethanolic, crude extracts), leading to inconsistent bioactive yields. Few studies optimized extraction parameters (e.g., solvent choice, temperature, and duration), affecting reproducibility. This study standardizes methanolic Soxhlet extraction (48 hr, controlled temperature), ensuring higher recovery of non-polar bioactive compounds.

The key bioactive compounds in TCMSE responsible for its antioxidant and anticancer properties include palmitic acid, Benzofuran derivatives, and 1,2,3,4-Butanetetrol, along with phenolic compounds like 3,5-bis(1,1-dimethylethyl)-Phenol. These molecules exhibit strong free radical scavenging (antioxidant) activity due to their hydroxyl (-OH) and conjugated double bond systems, which donate hydrogen atoms to neutralize reactive oxygen species. For anticancer effects, fatty acids (e.g., palmitic acid) and aromatic compounds (e.g., Benzofuran) disrupt cancer cell membranes and induce apoptosis, while phytosterols and diterpenoids (identified in GC-MS) modulate signaling pathways like NF-κB and p53.

The antioxidant and anticancer activities of TCMSE can be attributed to the synergistic effects of its major phytochemicals, as evidenced by prior studies. Palmitic acid, the most abundant compound (10.13%), exerts antioxidant effects by scavenging free radicals via its carboxyl group (-COOH) and alkyl chain, which stabilizes lipid peroxidation [1, 24]. Benzofuran derivatives (9.97%) are known to induce apoptosis in cancer cells by modulating pro-survival pathways (e.g., PI3K/

AKT) and elevating intracellular reactive oxygen species, leading to oxidative stress-mediated cytotoxicity [2, 32]. Phenolic compounds like 3,5-bis(1,1-dimethylethyl)-Phenol (6.03%) donate hydrogen atoms from their -OH groups to neutralize DPPH radicals, while also chelating metal ions to prevent Fenton reactions [3, 23]. Additionally, 1,2,3,4-Butanetetrol (9.30%) may enhance membrane permeability in cancer cells, as structurally similar polyols disrupt cellular homeostasis [4]. These mechanisms align with our experimental results (DPPH; IC_{50} = 52.80 µg/ml; dose-dependent cytotoxicity in K562/HeLa cells) and support TCMSE's traditional use in oxidative stress and cancer management.

Conclusion

The study identifies 52 bioactive compounds in TCMSE via GC-MS, with palmitic acid (10.13%), Benzofuran (9.97%), and 1,2,3,4-Butanetetrol (9.30%) emerging as dominant constituents, correlating strongly with observed bioactivities. The extract demonstrated potent antioxidant effects (DPPH; IC_{50} = 52.80 µg/ml) and dose-dependent cytotoxicity against K562 and HeLa cancer cells, validating its traditional use and revealing its dual therapeutic potential. Notably, the anticancer activity against both hematologic (K562) and solid tumor (HeLa) models suggest broad-spectrum applicability, a finding not comprehensively reported in prior studies.

The work also advances methodological rigor by standardizing Soxhlet-based methanolic extraction-optimizing non-polar phytochemical recovery and employing high-resolution GC-MS (NIST-verified) for precise compound identification, addressing reproducibility gaps in earlier research. Furthermore, the study bridges traditional knowledge with industrial relevance by proposing TCMSE for nutraceuticals (e.g., immunity-boosting supplements) and functional foods, leveraging its antioxidant and anticancer properties. These findings not only confirm *T. cordifolia*'s pharmacological value but also provide a validated phytochemical blueprint for future drug development, setting this work apart from fragmented or single-activity studies in literature. The integration of multi-assay validation (GC-MS, DPPH, MTT) with statistical robustness ($p < 0.05$, $n = 3$) further underscores its reliability, offering a benchmark for subsequent research on this medicinal plant.

Future Prospects

The future prospects of *T. cordifolia* in the food, nutraceutical, and pharmaceutical industries are highly promising due to their diverse bioactive phytochemicals. With growing consumer demand for natural and functional ingredients, *T. cordifolia* can be incorporated into nutraceutical formulations such as immunity-boosting supplements, herbal teas, and fortified beverages. The antioxidant and antimicrobial properties of its phyto-constituents also offer potential applications in food preservation, reducing reliance on synthetic additives. Additionally, its adaptogenic and anti-inflammatory compounds can support the development of specialized dietary products aimed at managing lifestyle-related disorders such as diabetes, obesity, and gut health issues.

In the pharmaceutical sector, continued research on *T. cordifolia*'s phytochemicals could lead to the development of novel drug formulations targeting immune modulation, inflammation, and metabolic disorders. The identification of bioactive alkaloids, terpenoids, and flavonoids through advanced techniques like GC-MS provides a foundation for drug discovery and therapeutic innovations. Future studies focusing on bioavailability, clinical efficacy, and formulation optimization will further enhance its commercial viability. As research progresses, *T. cordifolia* has the potential to bridge the gap between traditional herbal medicine and modern scientific advancements, making it a valuable resource for sustainable and evidence-based healthcare solutions.

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Conflict of Interest

None.

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