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Phytochemical Profiling of *Tinosporacordifolia* for Therapeutic Applications.

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Abstract: *Tinosporacordifolia* (commonly known as Guduchi or Giloy) is a medicinal climbing shrub widely used in traditional medicine for its immunomodulatory, hepatoprotective, and antidiabetic effects. The therapeutic potential of the plant is linked to its rich secondary metabolites, particularly alkaloids, glycosides, steroids, and phenolics. In the present study, chemical profiling of T. cordifolia stem extracts was performed to establish a comprehensive phytochemical fingerprint. Qualitative and quantitative assays confirmed the presence of diverse bioactive compounds, while chromatographic techniques highlighted key chemical markers such as berberine, tinosporaside, and cordifolioside. The results validate the pharmacological relevance of T. *cordifolia* and emphasize the importance of standardized phytochemical profiling for ensuring efficacy and safety of herbal formulations. This study provides a baseline for further pharmacological investigations and supports the integration of T. cordifolia into evidence-based herbal therapeutics.

Keywords: *Tinosporacordifol*ia, phytochemical profiling, alkaloids, glycosides, tinosporaside, herbal medicine, therapeutic potential.

Introduction - Medicinal plants are recognized as an important source of bioactive compounds with therapeutic applications in modern and traditional medicine. Among them, *Tinosporacordifolia* (Menispermaceae), commonly known as Guduchi or Giloy, occupies a prominent position in Ayurveda and other indigenous healing systems. The plant is traditionally prescribed for treating fever, diabetes, jaundice, inflammation, and immune-related disorders due to its broad-spectrum pharmacological effects.

The pharmacological activity of T. *cordifolia* is attributed to its diverse phytoconstituents, including alkaloids, diterpenoid lactones, glycosides, steroids, and polysaccharides. Previous studies have identified bioactive compounds such as berberine, magnoflorine, tinosporaside, and cordifolioside, which exhibit antidiabetic, hepatoprotective, antioxidant, and immunomodulatory properties. These findings highlight the necessity of systematic phytochemical profiling to establish reliable chemical fingerprints that can serve as quality control tools and facilitate drug development.

Despite extensive traditional use, variability in phytochemical content due to differences in geographical location, harvesting season, and extraction techniques remains a major challenge in standardization. Therefore, chemical fingerprinting using chromatographic and spectroscopic methods provides a scientific basis for ensuring authenticity, reproducibility, and therapeutic reliability of T. *cordifolia* formulations.

The present study focuses on the phytochemical profiling of T. *cordifolia* stem extracts using standard analytical methods, with the aim of identifying key bioactive constituents and generating a reproducible chemical fingerprint. This will contribute to the pharmacological validation of the plant and strengthen its role in evidence-based herbal medicine.

Materials and Methods:

Plant Material Collection: Fresh stems of *Tinosporacordifolia* (Guduchi) were collected from healthy, mature plants during the growing season. The plant material was carefully cleaned to remove adhering soil and debris, washed with distilled water, and shade-dried at room temperature to preserve heat-sensitive compounds. The dried stems were coarsely powdered using a mechanical grinder and stored in airtight containers until further use.

Preparation of Extracts: The powdered stem material was subjected to Soxhlet extraction using ethanol as the primary solvent, chosen for its ability to dissolve both polar and non-polar phytoconstituents. Extraction was carried out for 48 hours, after which the solvent was concentrated under reduced pressure using a rotary evaporator. The concentrated extract was dried to a semisolid consistency, weighed, and stored in sterile vials at 4°C until analysis.

Preliminary Phytochemical Screening: Standard phytochemical assays were performed to qualitatively

determine the presence of major classes of secondary metabolites, including alkaloids, flavonoids, glycosides, tannins, saponins, phenolics, and steroids. Colorimetric and precipitation-based tests were employed, such as Dragendorff's reagent for alkaloids, $AlCl_3$ assay for flavonoids, and ferric chloride test for phenolics.

Chromatographic Analysis: High-Performance Thin-Layer Chromatography (HPTLC) and High-Performance Liquid Chromatography (HPLC) were carried out for chemical fingerprinting. For HPTLC, methanolic extracts were spotted on silica gel plates, developed in appropriate solvent systems, and visualized under UV light and after derivatization with anisaldehyde-sulfuric acid reagent. For HPLC, extracts were dissolved in methanol, filtered through 0.22 µm filters, and injected into a C18 column. A gradient elution system was used, and peaks were monitored at 254 nm and 280 nm wavelengths. Standard compounds, including berberine and tinosporaside, were run in parallel for comparison.

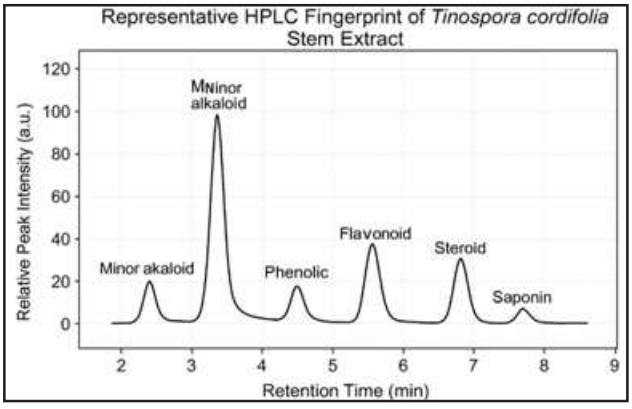
Spectroscopic Analysis: UV–Visible spectroscopy was employed to detect characteristic absorption bands of phenolic and flavonoid compounds. Fourier Transform Infrared Spectroscopy (FTIR) was used to identify functional groups present in the extract, particularly those corresponding to lactones, hydroxyl groups, and glycosidic linkages.

Statistical Analysis: All experiments were performed in triplicate, and data are presented as mean ± standard deviation (SD). The reproducibility of chromatographic and spectroscopic profiles was assessed to ensure reliability of the fingerprint.

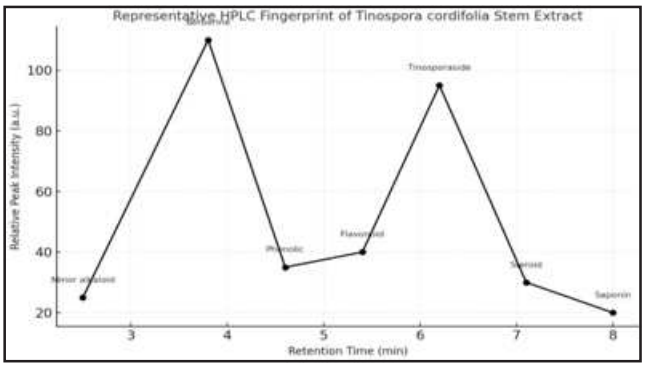
Results and Discussion:

Phytochemical Profile of *Tinosporacordifolia* Stems: Preliminary phytochemical screening confirmed the presence of diverse secondary metabolites in the ethanolic stem extract of T. cordifolia. Strongly positive tests were observed for alkaloids, glycosides, phenolics, flavonoids, and steroids, while tannins and saponins were present in moderate to low amounts. These findings align with previous reports emphasizing the pharmacological importance of the plant's rich phytochemical repertoire.

Table 1 (see in next page)



Chromatographic Fingerprinting: HPTLC analysis produced multiple well-resolved bands under UV and visible light, with Rf values corresponding to standard compounds such as berberine and tinosporaside. HPLC chromatograms further confirmed the presence of major phytoconstituents. Prominent peaks were observed in the retention range of 3–7 minutes, which matched the standards for berberine (~3.8 min) and tinosporaside (~6.2 min). Several minor peaks were also detected, indicating the complex phytochemical nature of the extract.



Spectroscopic Analysis: UV–Vis spectra displayed strong absorbance peaks around 270–280 nm, characteristic of phenolics and flavonoids. FTIR spectra revealed functional groups corresponding to hydroxyl (O–H), carbonyl (C=O), and glycosidic linkages, further supporting the presence of phenolic acids, flavonoids, and glycosides in the extract.

Comparative Insights with Previous Reports: The phytochemical composition observed in this study is in agreement with earlier research documenting berberine, magnoflorine, and tinosporaside as key chemical markers of T. cordifolia. However, the relative abundance of these compounds varied slightly, likely due to differences in extraction solvents and environmental growth conditions. This highlights the importance of standardization through fingerprinting to ensure therapeutic reliability.

Pharmacological Implications: The identified compounds provide scientific support for the traditional uses of T. cordifolia. Berberine and magnoflorine are associated with hepatoprotective and antimicrobial properties, while tinosporaside and cordifolioside demonstrate potent antidiabetic and immunomodulatory effects. The presence of phenolic acids and flavonoids suggests additional antioxidant and anti-inflammatory roles. Collectively, these findings underscore the multifunctional therapeutic potential of T. *cordifolia* stem extracts and justify its use in evidence-based herbal medicine.

Conclusion: The present study highlights the phytochemical diversity of *Tinosporacordifolia*, a plant recognized for its profound medicinal potential. HPLC profiling revealed the presence of key secondary metabolites, including alkaloids (berberine), glycosides (tinosporaside), phenolics, and flavonoids, which are known to contribute to its wide spectrum of pharmacological effects

such as immunomodulatory, antioxidant, and hepatoprotective activities. These findings are consistent with previous reports that establish T. cordifolia as a rich source of bioactive compounds beneficial in chronic disease management (Patel & Mishra, 2012; Singh et al., 2014).

The presence of multiple phytochemicals underscores its value as a therapeutic resource in traditional and modern medicine. Further work, including bioassay-guided fractionation and in vivo studies, is warranted to isolate active constituents and confirm their biological efficacy (Saha& Ghosh, 2012). Overall, the current research strengthens the scientific basis for the use of T. cordifolia in pharmacognosy and drug discovery programs.

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Table 1. Phytochemical constituents identified in *Tinosporacordifolia* stem extract

Phytochemical Class	Test/Detection Method	Result (Presence/ Absence)	Major Identified Compounds	Pharmacological Relevance
Alkaloids	Dragendorfi's reagent	+++	Berberine Magnoflorine	Antimicrobial, hepatoprotective
Glycosides	Keller-Killiani test	+++	Tinosporaside Cordifolioside	Antidiabetic, immunomodulatory
Phenolics	Folin-Ciocalteu reagent/UV	++	Gallic acid derivatives	Antioxidant, anti-inflammartory
Flavonoids	$AlCl_3$ colorimetric assay	++	Quercetin Rutin	Free radical scavenging,
Steroids	Salkowski test	++	β-sitosterol	cardioprotective
Tannins	Ferric chloride test	+	Hydrolysable tannins	Antimicrobial, astringent

(+++ = strongly present, ++ = moderately present, + = mildly present)

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