Pharmacogn. Res., 2026; 18(1):59-67.
https://www.phcogres.com

Original Article

# LC-MS Guided Discovery of Pharmacologically Active Metabolites from Ethanol Extracts of Bark and Leaf of *Vincetoxicum iphisia* Meve and Liede

**Ashitha Sugunan*, Jeeshna Meethaley Valappil, Sreelakshmi Thachan, Sarga Abhijith**

Department of Botany, Sree Narayana College, Kannur, Kerala, INDIA.

## ABSTRACT

**Background:** *Vincetoxicum iphisia* Meve and Liede is a lesser-known medicinal plant species with limited phytochemical characterization despite its traditional uses. This study aimed to investigate the bioactive metabolite profile of ethanol extracts from the bark and leaves of *V. iphisia* using Liquid Chromatography-Mass Spectrometry (LC-MS). **Materials and Methods:** Ethanol extracts were prepared from the bark and leaves and analyzed using LC-MS. Metabolites were identified based on retention time, molecular weight, and database comparisons. Total Ion Chromatograms (TICs) and Base Peak Intensity (BPI) profiles were used to assess shared and tissue-specific compounds. **Results:** LC-MS profiling revealed a range of pharmacologically relevant secondary metabolites including flavonoids (e.g., quercetin, isorhamnetin derivatives), phenolic glycosides, coumarins, alkaloids (e.g., L-kynurenine), and linear diarylheptanoids. Key compounds such as cinobufagin, catechin gallate, and triptophenolide known for their antioxidant, anti-inflammatory, cardioprotective, and anticancer effects were identified. **Conclusion:** This first LC-MS-based investigation of *V. iphisia* highlights its rich metabolite diversity and supports its potential as a source of pharmacologically important natural products, warranting further biological and mechanistic studies.

**Keywords:** *Vincetoxicum iphisia* Meve and Liede, Phytochemical Profiling, LC-MS Analysis, Secondary Metabolites.

**Correspondence:**
**Ms. Ashitha Sugunan**
Department of Botany, Sree Narayana College, Kannur, Kerala, INDIA.
Email: ashithasugunan7@gmail.com

**Received:** 24-06-2025;
**Revised:** 18-08-2025;
**Accepted:** 09-10-2025.

## INTRODUCTION

Medicinal plants have been integral to traditional healthcare systems such as Ayurveda, Unani, and Siddha for centuries, where they are predominantly used in polyherbal formulations composed of crude plant extracts. These extracts are abundant in phytochemicals, which are responsible for treating a broad range of human ailments. The pharmacological activities of these traditional remedies are primarily attributed to the presence of various secondary metabolites. While primary metabolites including amino acids, sugars, and nucleotides support basic cellular functions and plant development, secondary metabolites such as alkaloids, flavonoids, terpenoids, and glycosides play a central role in mediating therapeutic effects.[1] The advent of modern analytical tools, especially Liquid Chromatography Mass Spectrometry (LC-MS), has revolutionized the chemical profiling of complex plant matrices. LC-MS enables the sensitive and accurate detection of bioactive secondary metabolites, even at trace levels, and has proven particularly efficient in identifying phenolics, alkaloids, and flavonoids classes of compounds frequently associated with significant pharmacological activities.[2]

Despite their extensive use in ethnomedicine, many medicinal plants have not been thoroughly studied in terms of their phytochemical makeup and pharmacological potential. Estimates suggest that only around 15% of plant species have been chemically profiled, and less than 5% have undergone biological screening.[3] This underscores a pressing need to bridge traditional knowledge with contemporary scientific methods for the discovery of novel bioactive molecules.[4] Comprehensive phytochemical characterization is vital, not only to substantiate traditional therapeutic claims but also to support the development of new pharmaceutical agents.[5] Plants of the genus *Vincetoxicum* (family Apocynaceae) have exhibited notable biological activities, including antimicrobial, antioxidant, and anticancer effects. However, *Vincetoxicum iphisia* Meve and Liede remains an obscure and understudied species, despite anecdotal evidence supporting its traditional usage and possible pharmacological value. The absence of detailed phytochemical data on this species represents a critical knowledge gap. Accordingly, the present investigation aims to conduct a comprehensive phytochemical

**DOI:** 10.5530/pres.20260039

analysis of *V. iphisia* using LC-MS techniques. By integrating ethnomedicinal perspectives with advanced chemical analysis, this study endeavors to elucidate the chemical constituents of *V. iphisia* and assess its potential relevance in modern therapeutic applications.

## MATERIALS AND METHODS

### Collection of Plant Materials

Mature plant parts such as leaf and stem of *Vincetoxicum iphisia* Meve and Liede were collected from the Nilagiri hills in Coimbatore district, Tamil Nadu, India. The plants were authenticated by Dr. Ravichandran, Botanical Survey of India, and voucher specimens were deposited at the Madras Herbarium.

### Sample Preparation

The collected plant materials were washed, shade-dried, and powdered. Ethanol was used for solvent extraction using a *Soxhlet apparatus*. The extracts were filtered, concentrated under reduced pressure, and prepared for LC-MS analysis.

### LC-MS Analysis

LC-MS analysis was performed using a Waters Acquity UPLC system coupled with a Xevo G2-XS QTof mass spectrometer (Waters, USA). Chromatographic separation was achieved on an Acquity BEH C18 column (50 × 2.1 mm, 1.7 µm particle size). The mobile phases used were: (A) 0.1% formic acid in water and (B) acetonitrile. A gradient elution program was applied as follows: 0-1 min, 95% A and 5% B; 8 min, 50% A and 50% B; 12-17 min, 5% A and 95% B; followed by a return to initial conditions (95% A and 5% B) by 18 min, and held until 20 min. The flow rate was maintained at 0.400 mL/min, and the injection volume was 10 µL. For sample preparation, 10 mg of *V. iphisia* extract was dissolved in 2 mL of methanol, sonicated for 10 min, and filtered through a 0.22 µm syringe filter prior to LC-MS injection. Mass spectrometric analysis was conducted using Electrospray Ionization (ESI) in both positive and negative ion modes. The capillary voltage was set at 3.0 kV. Collision energy was applied in two stages: a fixed 20 V and a ramped range of 30-90 V. The source temperature was set to 150°C, and the desolvation temperature to 450°C. The cone gas flow was maintained at 50 L/h, and the desolvation gas flow at 800 L/h. Data acquisition and processing were conducted using MassLynx V4.1 software. Compound identification was based on accurate mass (m/z), Retention Time (RT), and fragmentation patterns, supported by spectral databases and literature reports.[6]

## RESULTS

### Phytochemical Profile

The LC-MS analysis of ethanol extracts from the bark and leaf of *V. iphisia* revealed a complex and diverse array of secondary metabolites (Figure 1). Chromatograms obtained in both positive and negative ionization modes displayed distinct peaks corresponding to various phytochemicals. In the bark extract, major compounds were detected at retention times of 1.25, 3.08, 4.44, 6.51, and 8.68 min, corresponding to flavonoid-7-O-glycosides, L-kynurenine, cis-zeatin, 4-hydroxycoumarin, and cinobufagin. Additional compounds identified in the bark included catechin gallate, aspalathin, isorhamnetin-3-O-rutinoside, and 7-methoxy-6-(1, 2, 3-trihydroxy-3-methylbutyl) chromen-2-one, along with phenolic glycosides and linear diarylheptanoids. The ethanol extract of the leaf showed prominent peaks at retention times of 3.53, 4.18, 6.51, 8.03, and 10.30 min. These peaks corresponded to compounds such as quercetin, 3-methylquercetin, catechin, capillarisin, and semivioxanthin. Negative ionization mode further confirmed the presence of apigenin-7-O-glucoside, quercetin-3, 4′-O-di-β-glucopyranoside, isorhamnetin-3, 7-di-O-glucoside, caffeoyl lysine, triptophenolide, additional coumarin derivatives, and phenolic glycosides. The compounds identified through LC-MS analysis in the bark and leaves are presented in Tables 1 and 2, respectively.

A comparative analysis of both extracts revealed several overlapping phytoconstituents, indicating a conserved phytochemical profile across different plant parts. Shared compounds included catechin or its derivatives, 7-methoxy-6-(1,2,3-trihydroxy-3-methylbutyl)chromen-2-one, the diarylheptanoid 7-(3,4-dihydroxyphenyl)-5-hydroxy-1-(4-hydroxyphenyl)-3-heptanone, and phenolic glycosides such as (2S,3R,4S,5S,6R)-2-[4-(3-hydroxybutyl)phenoxy]-6-(hydroxymethyl)oxane-3,4,5-triol. Flavonoid glycosides including isorhamnetin derivatives were also detected in both bark and leaf samples. The presence of these shared metabolites reflects the metabolic connectivity between vegetative and woody tissues and supports the traditional use of multiple plant parts in ethnomedicine. At the same time, the presence of unique peaks in each extract highlights the tissue-specific accumulation of certain bioactive compounds, which may contribute to differential therapeutic applications.

### Total Ion Chromatogram (TIC)

The Total Ion Chromatogram (TIC) profiles obtained from LC-MS analysis of the ethanol extracts of *V. iphisia* bark and leaf revealed a diverse array of phytochemicals spanning multiple retention times (Figures 2 and 3). The TIC for the bark extract exhibited prominent peaks corresponding to compounds such as (2R,3S)-7-[(2S,3R,4R,5S)-3,4-dihydroxy-5-(hydroxymethyl)oxolan-2-yl]oxy-2-(3,4-dihydroxyphenyl)-3,4-dihydro-2H-chromene-3,5-diol (RT 1.255 min), L-kynurenine (RT 3.075 min), cis-zeatin (RT 3.581 min), 4-hydroxycoumarin (RT 4.440 min), and cinobufagin (RT 8.684 min), among others. Notably, several unknown peaks with significant intensities were also detected, indicating the presence of potentially novel constituents. In the ethanol leaf extract, the

TIC exhibited distinct peaks at retention times corresponding to known bioactive flavonoids and phenolics, including quercetin (RT 3.935 min), 3-methylquercetin (RT 4.188 min), capillarisin (RT 4.895 min), catechin (RT 8.029 min), and semivioxanthin (RT 8.737 min), along with multiple glycosidic forms such as quercetin-3,4-O-di-β-glucopyranoside and isorhamnetin-3,7-di-O-glucoside. The chromatographic data, visualized through both positive and negative ion TICs, illustrate the phytochemical complexity of the species and provide a foundational profile for subsequent compound isolation and bioactivity studies, particularly focusing on anti-inflammatory potential.

## Base Peak Intensity (BPI) Chromatogram

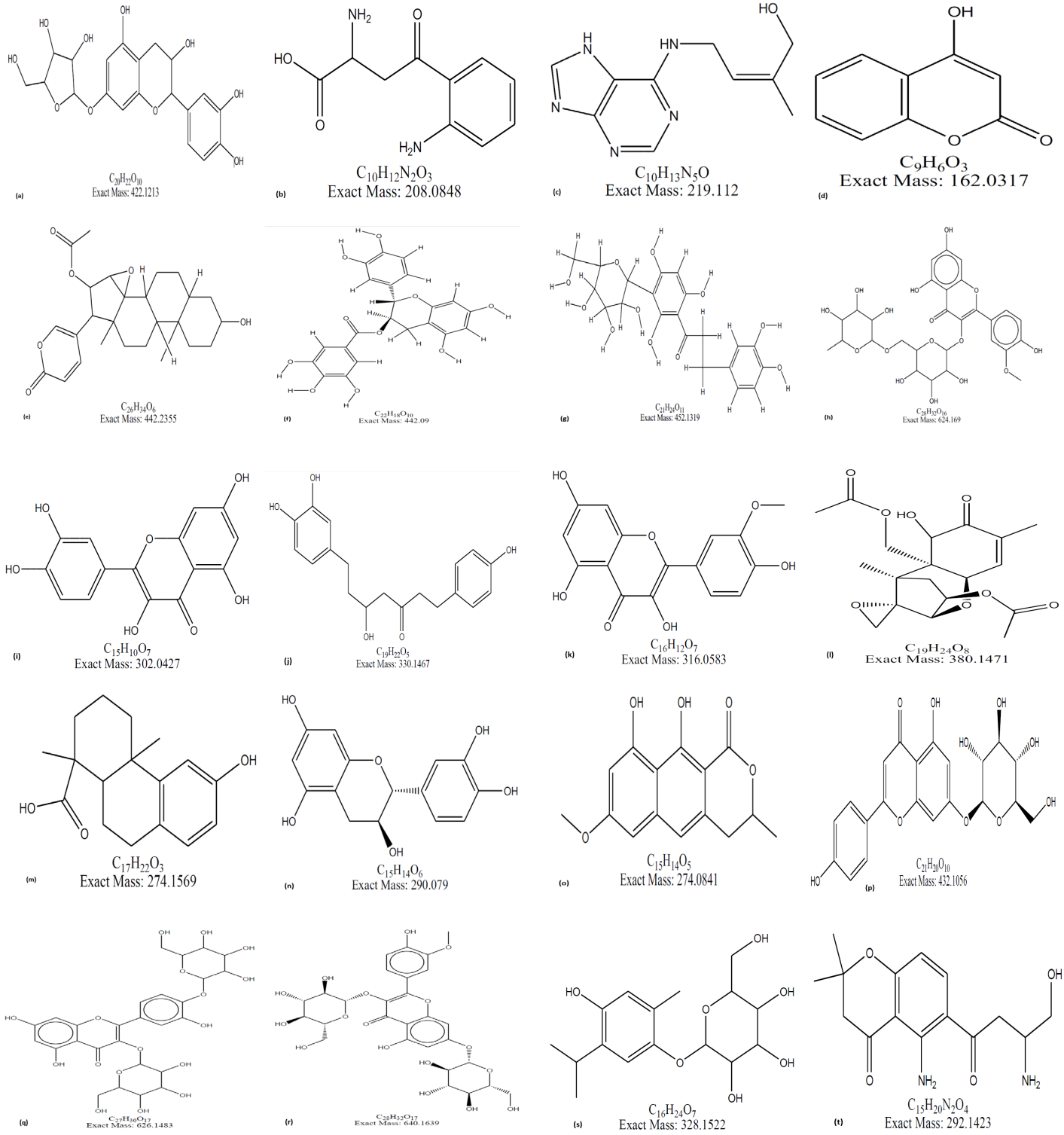
The Base Peak Intensity (BPI) chromatograms for both bark and leaf ethanol extracts of *V. iphisia* provided a refined view of the most intense ions at each retention time, enhancing the resolution of key bioactive constituents (Figures 1 and 2). In the bark extract, the BPI chromatogram displayed sharp and well-defined peaks at RT 1.255, 3.075, 3.581, and 8.684 min, corresponding respectively to flavonoid-7-O-glycosides, L-kynurenine, cis-zeatin, and the steroidal compound cinobufagin. These high-intensity peaks suggest a high abundance or superior ionization efficiency of these molecules under the applied ESI conditions. Similarly, the BPI profile of the leaf extract showed dominant peaks at RT 3.935, 4.188, 4.895, and 8.029 min, which were attributed to quercetin, 3-methylquercetin, capillarisin, and catechin. The intensity and distinctness of these peaks affirm the presence of potent antioxidant and anti-inflammatory flavonoids. The BPI chromatogram also revealed several minor yet distinct peaks that may correspond to less abundant but biologically relevant compounds, including phenolic glycosides and hydroxycinnamic acid derivatives. The clarity and peak separation observed in the BPI data highlight its utility for selecting marker compounds for targeted quantification and for guiding further pharmacological evaluations.

**Table 1: LC-MS Identified Compounds from Ethanol Bark Extract of *Vincetoxicum iphisia*.**

| Sl. No. | Compound Name | RT (min) | Precursor m/z | Adduct | Molecular Formula | Ontology |
|---|---|---|---|---|---|---|
| 1 | (2R,3S)-7-[(2S,3R,4R,5S)-3,4-dihydroxy-5-(hydroxymethyl) oxolan-2-yl]oxy-2-(3,4-dihydroxyphenyl)-3,4-dihydro-2H-chromene-3,5-diol | 1.255 | 445.1202 | [M+Na]+ | $C_{20}H_{22}O_{10}$ | Flavonoid-7-O-glycosides |
| 2 | L-Kynurenine | 3.075 | 209.0872 | [M+H]+ | $C_{10}H_{12}N_2O_3$ | Alkyl-phenylketones |
| 3 | cis-Zeatin | 3.581 | 220.1845 | [M+H]+ | $C_{10}H_{13}N_5O$ | N/A |
| 4 | 4-hydroxycoumarin | 4.440 | 163.0486 | [M+H]+ | $C_9H_6O_3$ | 4-hydroxycoumarins |
| 5 | Cinobufagin | 8.684 | 443.1747 | [M+H]+ | $C_{26}H_{34}O_6$ | Steroid |
| 6 | Catechin gallate | 1.255 | 441.0257 | [M−H]− | $C_{22}H_{18}O_{10}$ | N/A |
| 7 | Aspalathin | 3.075 | 451.0545 | [M−H]− | $C_{21}H_{24}O_{11}$ | N/A |
| 8 | Isorhamnetin-3-O-rutinoside | 4.440 | 623.0668 | [M−H]− | $C_{28}H_{32}O_{16}$ | Flavonoid |
| 9 | (2S,3R,4S,5S,6R)-2-[4-(3-hydroxybutyl)phenoxy]-6-(hydroxymethyl)oxane-3,4,5-triol | 6.513 | 327.1510 | [M−H]− | $C_{16}H_{24}O_7$ | Phenolic glycosides |
| 10 | 7-(3,4-dihydroxyphenyl)-5-hydroxy-1-(4-hydroxyphenyl)-3-Heptanone | 6.917 | 329.1652 | [M−H]− | $C_{19}H_{22}O_5$ | Linear diarylheptanoids |
| 11 | 7-methoxy-6-(1,2,3-trihydroxy-3-methylbutyl) chromen-2-one | 10.405 | 293.1529 | [M−H]− | $C_{15}H_{18}O_6$ | Coumarins |

## Pharmacological Relevance

The ethanol extracts derived from both the bark and leaves of the plant revealed a diverse array of bioactive secondary metabolites with notable pharmacological significance (Table 3). Among the principal constituents identified were flavonoid glycosides, including (2R,3S)-7-[(2S,3R,4R,5S)-3,4-dihydroxy-5-(hydroxymethyl)oxolan-2-yl]oxy-2-(3,4-dihydroxyphenyl)-3,4-dihydro-2H-chromene-3,5-diol, quercetin-3,4'-di-O-glucoside, and isorhamnetin-3-O-rutinoside. These compounds are well-recognized for their potent antioxidant, anti-inflammatory, and anticancer activities.[7-9]



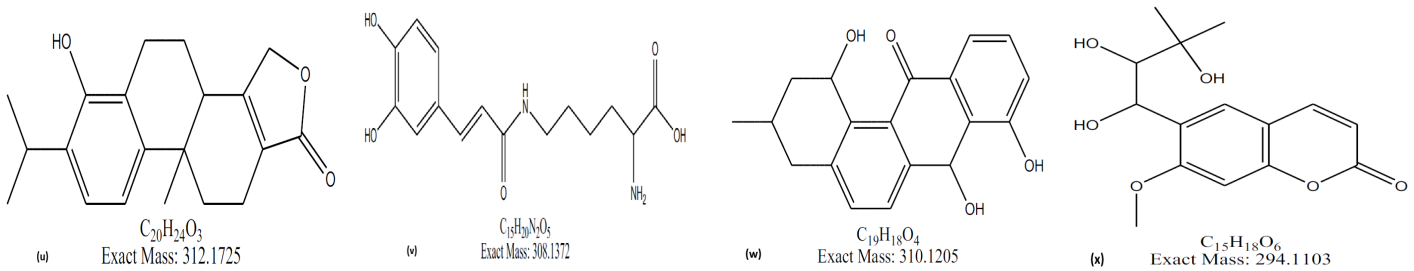



**Figure 1:** Chemical stuctures of selected phytocompounds: a) Quercetin, b) Isorhamnetin, c) Catechin, d) Apigenin-7-O-glucoside, e) Cinobufagin, f) Aspalathin, g) Triptophenolide, h) L-Kynurenine, i) Capillarisin, j) Semivioxanthin, k) 4-Hydroxycoumarin, l) Caffeoyl lysine, m) 7-methoxy-6-(1,2,3-trihydroxy-3-methylbutyl) chromen-2-one, n) Quercetin-3,4′-O-di-β-glucopyranoside, o) Isorhamnetin-3,7-di-O-glucoside, p) Catechin gallate, q) 7-(3,4-Dihydroxyphenyl)-5-hydroxy-1-(4-hydroxyphenyl)-3-heptanone, r) Podocarpic acid, s) 1,7,8-Trihydroxy-3-methyl-2,3,4,7-tetrahydro-1H-benzo[a]anthracen-12-one, t) cis-Zeatin, u) Phenolic glycoside, v) 3-Methylquercetin, w) Fusarochromanone, and x) 3,15-Diacetyldeoxynivalenol.

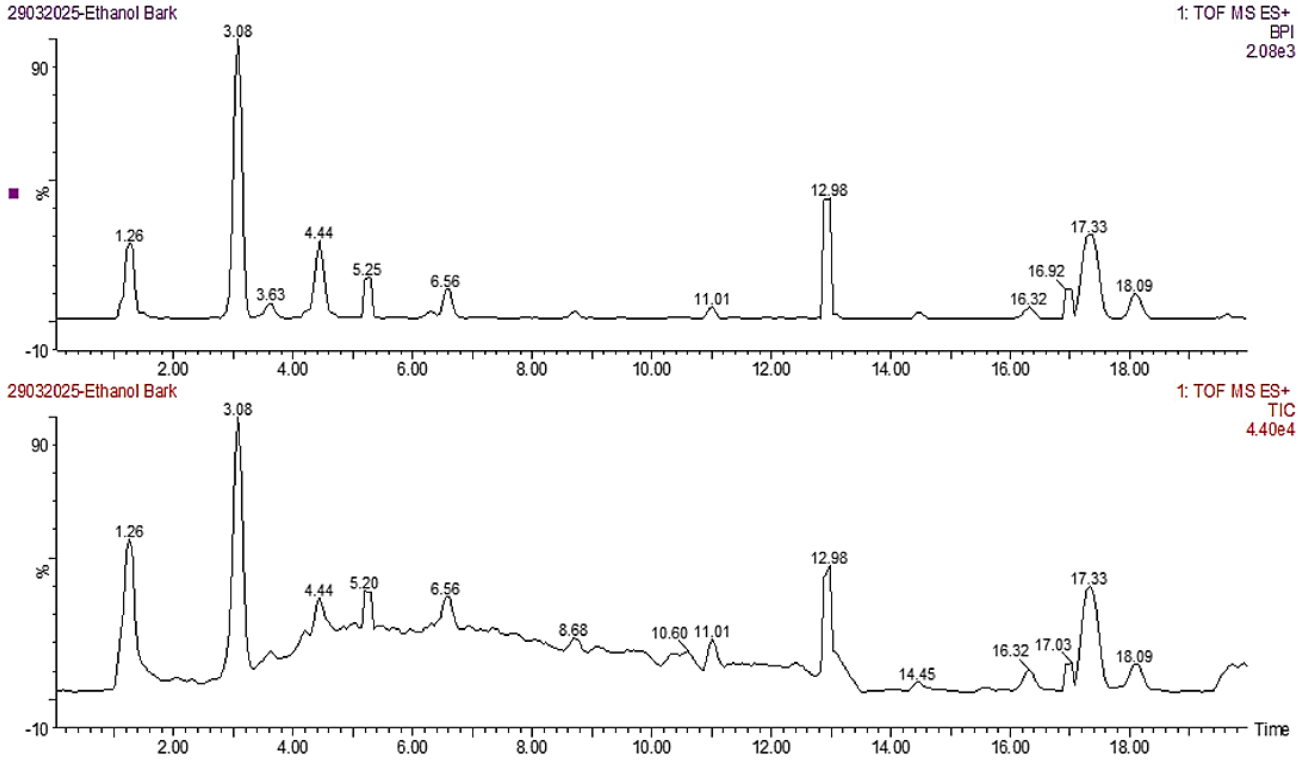



**Figure 2:** Positive mode TIC and BPI Chromatograms of bark of *Vincetoxicum iphisia*. The X- axis represents retention time (minutes), and the y-axis indicates compound intensity.

Additionally, aspalathin, a flavonoid present in the extract, has been shown to exhibit significant antidiabetic effects and free radical scavenging capacity.[10] The identification of phenolic compounds such as quercetin, catechin gallate, and capillarisin further supports the extract's therapeutic promise, given their reported cardioprotective, antihypertensive, and anticancer effects.[11-13] A coumarin derivative, 7-methoxy-6-(1, 2, 3-trihydroxy-3-methylbutyl) chromen-2-one, known for its antioxidant and anticancer properties, was also detected.[14] The presence of linear diarylheptanoids, particularly 7-(3, 4-dihydroxyphenyl)-5-hydroxy-1-(4-hydroxyphenyl)-3-heptanone, indicates anti-inflammatory and neuroprotective actions comparable to curcuminoids.[15] Moreover, the diterpenoid triptophenolide, found in both bark and leaf samples, is recognized for its immunosuppressive and anticancer activities.[16] The bark extract also contained the steroidal compound cinobufagin, which possesses cardiotonic and antineoplastic properties.[17]

Other bioactive compounds identified include L-kynurenine, a neuroactive molecule involved in immune regulation,[18] and 4-hydroxycoumarin, a known precursor in the synthesis of anticoagulants like warfarin.[19] Additionally, caffeoyl lysine, observed in the leaf extract, has demonstrated antioxidant potential and nitric oxide-scavenging capacity.[20] The diterpenoid podocarpic acid, also found in the leaf, is notable

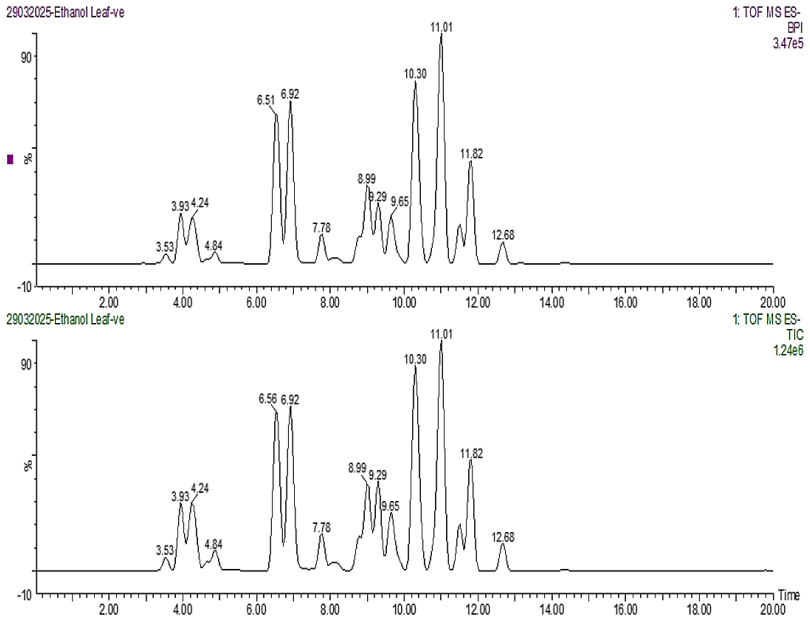



**Figure 3:** Positive mode TIC and BPI Chromatograms of leaf of *Vincetoxicum iphisia*, The X-axis represents retention time (minutes), and the y-axis indicates compound intensity.

**Table 2: LC-MS Identified Compounds from Ethanol Leaf Extract of *Vincetoxicum iphisia*.**

| Sl. No. | Proposed Compounds | Retention Time (min) | Precursor | Adduct | Formula | Ontology |
|---|---|---|---|---|---|---|
| 1 | 3,15-Diacetyldeoxynivalenol | 1.154 | 381.1005 | [M+H]+ | $C_{19}H_{24}O_8$ | N/A |
| 2 | Quercetin | 3.935 | 303.0676 | [M+H]+ | $C_{15}H_{10}O_7$ | Flavonoid |
| 3 | 3-Methylquercetin | 4.188 | 317.0837 | [M+H]+ | $C_{16}H_{12}O_7$ | Flavonoid |
| 4 | Capillarisin | 4.895 | 317.0837 | [M+H]+ | $C_{16}H_{12}O_7$ | Chromone |
| 5 | Fusarochromanone | 6.512 | 275.2178 | [M+H]+ | $C_{15}H_{20}N_2O_4$ | N/A |
| 6 | Podocarpic acid | 7.726 | 275.1057 | [M+H]+ | $C_{17}H_{22}O_3$ | N/A |
| 7 | Catechin | 8.029 | 291.2096 | [M+H]+ | $C_{15}H_{14}O_6$ | Catechins |
| 8 | Semivioxanthin | 8.737 | 275.2144 | [M+H]+ | $C_{15}H_{14}O_6$ | N/A |
| 9 | Apigenin-7-O-glucoside | 3.530 | 431.1010 | [M−H]− | $C_{21}H_{20}O_{10}$ | Flavonoid-7-O-glycosides |
| 10 | Quercetin-3,4'-O-di-β-glucopyranoside | 3.934 | 625.0055 | [M−H]− | $C_{27}H_{30}O_{17}$ | Flavonoid-3-O-glycosides |
| 11 | Isorhamnetin-3,7-di-O-glucoside | 4.238 | 639.0180 | [M−H]− | $C_{28}H_{32}O_{17}$ | Flavonol glycosides |
| 12 | 2-(Hydroxymethyl)-6-(4-hydroxy-2-methyl-5-propan-2-ylphenoxy)oxane-3,4,5-triol | 6.563 | 327.1473 | [M−H]− | $C_{16}H_{24}O_7$ | Phenolic glycosides |
| 13 | 7-(3,4-Dihydroxyphenyl)-5-hydroxy-1-(4-hydroxyphenyl)-3-heptanone | 6.917 | 329.1652 | [M−H]− | $C_{19}H_{22}O_5$ | Linear diarylheptanoids |
| 14 | 1,7,8-Trihydroxy-3-methyl-2,3,4,7-tetrahydro-1H-benzo[a] anthracen-12-one | 7.776 | 309.1425 | [M−H]− | $C_{19}H_{18}O_4$ | Phenanthrols |
| 15 | Caffeoyl lysine | 8.130 | 307.1259 | [M−H]− | $C_{15}H_{20}O_5$ | Hydroxycinnamic acids |
| 16 | Triptophenolide | 9.293 | 311.1584 | [M−H]− | $C_{20}H_{24}O_3$ | Oxosteroids |
| 17 | 7-Methoxy-6-(1,2,3-trihydroxy-3-methylbutyl) chromen-2-one | 10.304 | 293.1529 | [M−H]− | $C_{15}H_{18}O_6$ | Coumarins |

**Table 3: Pharmacological relevance of identified compounds.**

| Sl. No. | Compound Name | Pharmacological Relevance | Citation |
|---|---|---|---|
| 1 | Flavonoid-7-O-glycoside (e.g., catechin derivatives) | Antioxidant, anti-inflammatory, anti-cancer | [7] |
| 2 | L-Kynurenine | Neuroprotective, immune modulation, anti-inflammatory | [18] |
| 3 | cis-Zeatin (cytokinin class) | Plant growth regulator; limited direct human pharmacology | [26] |
| 4 | 4-Hydroxycoumarin | Anticoagulant (warfarin precursor), antimicrobial | [19] |
| 5 | Cinobufagin (steroidal compound) | Cardiotonic, anti-cancer | [17] |
| 6 | Catechin gallate | Strong antioxidant, antimicrobial | [8] |
| 7 | Aspalathin | Antidiabetic, antioxidant | [10] |
| 8 | Isorhamnetin-3-O-rutinoside | Antioxidant, hepatoprotective, cardioprotective | [9] |
| 9 | Phenolic glycoside | Antioxidant, anti-inflammatory | [27] |
| 10 | Linear diarylheptanoid (e.g., curcuminoid-like) | Anti-inflammatory, anti-tumor, neuroprotective | [15] |
| 11 | 7-methoxy-6-(1,2,3-trihydroxy-3-methylbutyl) chromen-2-one | Antioxidant, anticancer (coumarin class) | [14] |
| 12 | Quercetin | Antioxidant, antihypertensive, antidiabetic | [13] |
| 13 | Quercetin methyl derivatives (e.g., 3-Methylquercetin) | Antioxidant, anti-inflammatory | [23] |
| 14 | Fusarochromanone | Mycotoxin, antimicrobial | [25] |
| 15 | Podocarpic acid | Antibacterial, anticancer (terpenoid-like) | [7] |
| 16 | Catechin, semivioxanthin | Antioxidant, anti-inflammatory | [20] |
| 17 | Semivioxanthin | Antioxidant (flavonoid-related) | [21] |
| 18 | Apigenin-7-O-glucoside | Antioxidant, anti-inflammatory, anti-cancer | [22] |
| 19 | Quercetin diglucoside | Strong antioxidant, anti-inflammatory | [24] |
| 20 | Isorhamnetin-3,7-di-O-glucoside | Cardioprotective, anti-inflammatory | [28] |
| 21 | Phenanthrols (e.g., triptophenolide class) | Anticancer, apoptosis inducer | [29] |
| 22 | Caffeoyl lysine | Antioxidant, nitric oxide inhibitor | [30] |
| 23 | Triptophenolide | Immunosuppressive, anticancer | [16] |

for its antibacterial and anticancer properties.[21] Collectively, these identified phytochemicals highlight the plant's substantial pharmacological potential, especially in the development of novel natural therapeutics with antioxidant, anti-inflammatory, anticancer, immunomodulatory, and cardioprotective properties.

## DISCUSSION

The present study elucidated the phytochemical composition of *V. iphisia* via LC-MS profiling of ethanol extracts from its bark and leaves. The findings reveal a rich repository of bioactive compounds, prominently including flavonoids, phenolic glycosides, coumarins, alkaloids, and diarylheptanoids, which collectively contribute to its pharmacological potential. Flavonoid-7-O-glycosides such as quercetin and isorhamnetin derivatives were predominant and are of particular interest due to their roles in antioxidative defense mechanisms. These compounds have demonstrated free radical scavenging activity and are implicated in the prevention of cardiovascular and neurodegenerative diseases.[22] The glycosylation of flavonoids is known to enhance their solubility and absorption, improving bioefficacy, as seen in apigenin-7-O-glucoside and quercetin-3-O-glucoside detected in the samples. Coumarin derivatives identified in both plant parts, including methoxylated and hydroxylated chromen-2-one compounds, align with reported anticoagulant and anti-inflammatory activity in related taxa.[23] Their consistent presence in both tissues suggests a potential systemic role in the plant's defense or signaling pathways. Notably, diarylheptanoids and phenolic ketones

identified in the extracts contribute to anti-inflammatory potential, as compounds in this class have shown cyclooxygenase inhibition and radical-quenching properties.[24] Additionally, the presence of alkaloids, including potential indole-based constituents, may explain the traditional usage of *V. iphisia* for immune modulation and microbial infections. The detection of neuroactive metabolites such as L-kynurenine suggests possible neuromodulatory or immunoregulatory functions. Kynurenine derivatives have been implicated in neuroprotective pathways and immunological crosstalk, further supporting the versatility of *V. iphisia* in Ethnomedicine.[25] Importantly, several flavonoids, coumarins, and glycosides were detected across both bark and leaf extracts, indicating conserved biosynthetic mechanisms. However, the occurrence of tissue-specific peaks also points to metabolic specialization, offering opportunities for tissue-targeted extraction in pharmaceutical applications.

## ACKNOWLEDGEMENT

The authors extend their sincere gratitude to Sree Narayana College, Kannur for their support in facilitating this research. Dr. Jeeshna MV acknowledges the University Grants Commission (UGC) for awarding the Junior Research Fellowship (RDC A/A1/2241/2023) to Ashitha Sugunan, which enabled the successful completion of this study.

## ABBREVIATIONS

***V. iphisia*:** *Vincetoxicum iphisia*; **LC-MS:** Liquid Chromatography-Mass Spectrometry; **TIC:** Total Ion Chromatogram; **BPI:** Base Peak Intensity; **RT:** Retention Time; **ESI:** Electrospray Ionization; **UPLC:** Ultra Performance Liquid Chromatography; **QToF:** Quadrupole Time of Flight; **BEH:** Bridged Ethyl Hybrid; **m/z:** Mass-to-Charge Ratio.

## CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

## FUNDING

This work was supported by the Council of Scientific and Industrial Research (CSIR) and the University Grants Commission (UGC), Government of India, through their financial assistance.

## SUMMARY

This study provides the first detailed LC-MS based phytochemical assessment of ethanol extracts from the bark and leaves of *V. iphisia*. The analysis revealed a wide spectrum of secondary metabolites, demonstrating notable variation between plant parts. Chromatographic data indicated both common and unique constituents, reflecting the species' metabolic diversity. The identified metabolites are associated with various therapeutic properties, aligning with the plant's traditional medicinal use. Overall, these findings suggest that *V. iphisia* holds considerable potential as a natural source for future pharmacological exploration.

## REFERENCES

1. Seigler DS. Plant secondary metabolism. Berlin: Springer; 1998. doi: 10.1007/978-1-4615-4913-0.
2. Wolfender JL, Ndjoko K, Hostettmann K. Analysis of natural products by liquid chromatography-mass spectrometry. J Chromatogr A. 2005; 1000: 437-55.
3. Verpoorte R. Plant secondary metabolites and their role in plant physiology. J Pharm Pharmacol. 2000; 52(3): 253-62. doi: 10.1211/0022357001773931, PMID 10757412.
4. Tambe R, Bhambar R. Pharmacognostic review on **Vitex negundo**. Res Rev J Pharmacogn Phytochem. 2014; 2: 41-7.
5. Devi KP, Rajavel T, Habtemariam S, Nabavi SF. Antioxidant potential of selected medicinal plants. J Pharm Pharmacol. 2012; 64: 1347-59.
6. Singh A, Kumar S, Bajpai V, Reddy TJ, Rameshkumar KB, Kumar B. Identification of bioactive compounds by mass spectrometry. Rapid Commun Mass Spectrom. 2015; 29(12): 1095-106. doi: 10.1002/rcm.7202, PMID 25981540.
7. Yao LH, Jiang YM, Shi J, Tomás-Barberán FA, Datta N, Singanusong R, *et al.* Flavonoids in food and their health benefits. Plant Foods Hum Nutr. 2004; 59(3): 113-22. doi: 10.1007/s11130-004-0049-7, PMID 15678717.
8. Rice-Evans CA, Miller NJ, Paganga G. Structure-antioxidant activity relationships of flavonoids and phenolic acids. Free Radic Biol Med. 1996; 20(7): 933-56. doi: 10.1016/0891-5849(95)02227-9, PMID 8743980.
9. Miyake Y, Yamamoto K, Morimitsu Y, Osawa T. Protective effects of flavonoids on oxidative stress. J Agric Food Chem. 2007; 55: 2143-8.
10. Kawano A, Nakamura H, Hata S, Yamada M, Mikami K, Seki T, *et al.* Rooibos tea and aspalathin suppress hyperglycemia and oxidative stress in diabetic mice. Phytomedicine. 2009; 16: 437-43.
11. Li Y, Yao J, Han C, Yang J, Chaudhry MT, Wang S, *et al.* Quercetin, inflammation and immunity. Oxid Med Cell Longev. 2016; 2016: 9851239.
12. Zhou Y, Zheng J, Li S, Zhou T, Zhang P, Li HB, *et al.* Natural polyphenols for prevention and treatment of cancer. Food Funct. 2014; 5: 1272-8.
13. Middleton E Jr, Kandaswami C, Theoharides TC. The effects of plant flavonoids on mammalian cells: implications for inflammation, heart disease, and cancer. Pharmacol Rev. 2000; 52(4): 673-751. doi: 10.1016/S0031-6997(24)01472-8, PMID 11121513.
14. Venugopala KN, Rashmi V, Odhav B. Review on natural coumarin lead compounds for their pharmacological activity. Bioorg Med Chem Lett. 2013; 23: 1710-5.
15. Shishodia S, Sethi G, Aggarwal BB. Curcumin: getting back to the roots. Ann N Y Acad Sci. 2005; 1056: 206-17. doi: 10.1196/annals.1352.010, PMID 16387689.
16. Kupchan SM, Court WA, Dailey RG, Gilmore CJ, Bryan RF. Triptolide and triptophenolide: new antileukemic diterpenoid triepoxides from **Tripterygium wilfordii**. J Med Chem. 1972; 15: 339-40.
17. Qi F, Li A, Inagaki Y, Gao J, Li J, Kokudo N, *et al.* Antitumor activity of cinobufagin. J Ethnopharmacol. 2011; 135: 594-600.
18. Leipnitz G, Schumacher RF, Scussiato K, Dalcin KB, Seminotti B, Dutra-Filho CS, *et al.* Evidence for an antioxidant role of L-kynurenine. Toxicol Vitro. 2005; 19: 603-9.
19. Link KP. The discovery of dicoumarol and its development as an anticoagulant. Circulation. 1959; 19(1): 97-107. doi: 10.1161/01.cir.19.1.97, PMID 13619027.
20. Satue-Gracia MT, Heinonen M, Frankel EN. Antioxidant activity of anthocyanins in LDL oxidation. J Agric Food Chem. 1997; 45: 3120-6.
21. Steyn PS, Van Heerden FR. Podocarpic acid derivatives and their biological activities. Phytochemistry. 1983; 22: 539-65.
22. Bhalodia NR, Shukla VJ. Antimicrobial and antioxidant activities of some medicinal plants. J Young Pharm. 2011; 3: 107-11.
23. Rathore M, Juyal V, Bhatt A. Pharmacognostic and phytochemical studies of selected medicinal plants. Int J Pharm Sci Res. 2013; 4: 913-20.
24. Soni H, Sharma S, Singh S. Phytochemical and pharmacological evaluation of medicinal plants. Indian J Nat Sci. 2015; 6: 10582-8.
25. Gupta R, Sharma AK, Kumar R. Phytochemical analysis and antioxidant activity. Indian J Nat Prod Resour. 2017; 8: 288-99.
26. Mok DW, Mok MC. Cytokinin metabolism and action. Annu Rev Plant Physiol Plant Mol Biol. 2001; 52: 89-118. doi: 10.1146/annurev.arplant.52.1.89, PMID 11337393.

27. Pandey KB, Rizvi SI. Plant polyphenols as dietary antioxidants in human health and disease. Oxid Med Cell Longev. 2009; 2(5): 270-8. doi: 10.4161/oxim.2.5.9498, PMID 20716914.
28. Mukherjee PK. Quality control of herbal drugs. New Delhi: Business Horizons; 2002.
29. Sofowora A. Medicinal plants and traditional medicine in Africa. Ibadan, Nigeria: Spectrum Books; 1993.
30. Evans WC. Trease and Evans pharmacognosy. 16th ed. London: Saunders Elsevier; 2009.

**Cite this article:** Sugunan A, Valappil JM, Thachan S, Abhijith S. LC-MS Guided Discovery of Pharmacologically Active Metabolites from Ethanol Extracts of Bark and Leaf of *Vincetoxicum iphisia* Meve and Liede. Pharmacog Res. 2026;18(1):59-67.