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Pharmacognostic and Phytochemical Evaluation of Root and Root Bark of *Tephrosia purpurea* Linn Pers

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ABSTRACT

Tephrosia purpurea Linn. Pers., commonly known as wild indigo or purple tephrosia, belongs to the family Leguminosae and has been extensively used in traditional medicine systems for various therapeutic applications. This study presents a comprehensive pharmacognostic and phytochemical evaluation of the root and root bark of *T. purpurea* to establish standardization parameters and identify bioactive constituents. The morphological characteristics, microscopic features, and phytochemical profiles were systematically analysed using standard protocols. Macroscopic examination revealed characteristic features including cylindrical roots with greyish-brown exterior and yellowish interior. Microscopic analysis showed the presence of cork cells, cortical parenchyma, medullary rays, and xylem vessels with specific arrangements. Phytochemical screening confirmed the presence of alkaloids, flavonoids, tannins, phenolic compounds, saponins, carbohydrates, proteins, and amino acids in both root and root bark extracts. High-performance liquid chromatography (HPLC) analysis identified rotenone as the major marker compound in roots, while tephrosin was predominant in root bark. The study establishes essential pharmacognostic parameters and validates the traditional medicinal use of *T. purpurea* through scientific evidence of its bioactive constituents.

Key Words *Tephrosia purpurea*, Pharmacognosy, Phytochemistry, Root, Root bark, Rotenone, Tephrosin, Standardization

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INTRODUCTION

Tephrosia purpurea Linn. Pers., a perennial herb of the Leguminosae family, has been recognized as a valuable medicinal plant in various traditional systems of medicine including Ayurveda, Unani, and folk medicine. The plant is

widely distributed across tropical and subtropical regions and has gained significant attention due to its diverse pharmacological properties including hepatoprotective, anti-inflammatory, antimicrobial, antioxidant, and immunomodulatory activities¹.

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The root and root bark of *T. purpurea* are particularly valued for their therapeutic potential and have been traditionally used for treating liver disorders, respiratory ailments, digestive problems, and skin diseases. The World Health Organization (WHO) has emphasized the importance of standardization of herbal medicines to ensure their quality, safety, and efficacy². Pharmacognostic evaluation serves as the primary step in standardization, providing essential parameters for identification and quality control of medicinal plants.

Despite extensive traditional use, comprehensive pharmacognostic and phytochemical documentation of *T. purpurea* root and root bark remains limited in scientific literature. This study aims to fill this knowledge gap by providing detailed morphological, anatomical, and chemical characterization to establish standardization parameters and support evidence-based utilization of this valuable medicinal plant.

Pharmacognostical and Phytochemical Evaluation

Pharmacognostical evaluation encompasses the systematic study of medicinal plants involving macroscopic and microscopic examination, phytochemical analysis, and establishment of quality parameters. This multidisciplinary approach combines botanical identification, anatomical characterization, and chemical profiling to ensure authenticity and quality of herbal drugs³.

The methodology employed in this study follows standard protocols established by the Indian

Pharmacopoeia Commission and WHO guidelines for herbal drug standardization^{2,4}. Fresh plant material was collected, authenticated, and processed according to established procedures for morphological and anatomical studies. Phytochemical analysis was conducted using both qualitative and quantitative methods to identify and characterize bioactive constituents.

Geographical Distribution

Tephrosia purpurea exhibits a wide geographical distribution across tropical and subtropical regions of the world. The species is native to Asia and Africa but has been naturalized in various other regions including Australia, the Americas, and Pacific islands. In India, the plant is commonly found throughout the plains and lower hills up to an altitude of 1,200 meters above sea level⁵.

The species shows remarkable adaptability to diverse climatic conditions and soil types, thriving in both cultivated and waste lands. It is particularly abundant in the states of Rajasthan, Gujarat, Maharashtra, Karnataka, Tamil Nadu, Andhra Pradesh, and West Bengal. The plant also occurs naturally in countries such as Sri Lanka, Myanmar, Thailand, Malaysia, Indonesia, Philippines, and various African nations including Kenya, Tanzania, and South Africa.

Environmental factors such as temperature, rainfall, soil composition, and altitude significantly influence the growth pattern and phytochemical composition of *T. purpurea*. Plants growing in arid and semi-arid regions

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typically show higher concentrations of secondary metabolites, possibly as an adaptive response to environmental stress.

Botanical Description and Varieties

Tephrosia purpurea is a perennial herb or small shrub belonging to the subfamily Papilionoideae of the Leguminosae family. The plant typically

grows to a height of 30-90 cm and exhibits characteristic features of leguminous plants including compound leaves and papilionaceous flowers.

Several varieties of *T. purpurea* have been recognized based on morphological variations (Table 1).

Table 1: Varieties of *Tephrosia purpurea* and their distinguishing features

Variety	Distinguishing Features	Distribution	Medicinal Use
<i>T. purpurea</i> var. <i>purpurea</i>	Purple flowers, 5-9 leaflets	Widespread in India	Traditional medicine
<i>T. purpurea</i> var. <i>glabra</i>	Glabrous leaves and stems	Coastal regions	Liver disorders
<i>T. purpurea</i> var. <i>pumila</i>	Dwarf habit, smaller leaves	Dry regions	Respiratory ailments

Morphology

Macroscopic Characteristics

Root: The roots of *T. purpurea* are typically woody, cylindrical to sub-cylindrical, and range from 10-30 cm in length and 0.5-2.5 cm in diameter. The external surface is greyish-brown to dark brown, showing longitudinal wrinkles and occasional transverse cracks. The texture is hard and tough, with a fibrous fracture. When broken, the root shows a yellowish to pale yellow interior with distinct annual rings. The odor is characteristic and slightly aromatic, while the taste is bitter and astringent.

Root Bark: The root bark appears as curved or quilled pieces, 2-8 cm in length and 2-5 mm in thickness. The outer surface is rough, greyish-brown with darker patches, and shows longitudinal striations. The inner surface is smooth, yellowish-brown, and shows fine longitudinal striations. The fracture is short and granular in the outer portion and fibrous in the inner portion. The bark has a characteristic odor and bitter taste.

Microscopic Characteristics

Root (Transverse Section): The transverse section of *T. purpurea* root shows a typical dicotyledonous structure with the following layers:

- **Cork:** 8-12 layers of thin-walled, rectangular cells filled with brownish contents
- **Cortex:** 15-20 layers of parenchymatous cells containing starch grains and calcium oxalate crystals
- **Endodermis:** Single layer of barrel-shaped cells with Casparian strips
- **Pericycle:** 2-3 layers of sclerenchymatous cells
- **Phloem:** Consists of sieve tubes, companion cells, and phloem parenchyma
- **Cambium:** 2-4 layers of meristematic cells
- **Xylem:** Shows vessels, tracheids, wood fibers, and medullary rays

Root Bark (Transverse Section): The root bark shows the following microscopic features:

- **Outer Cork:** 10-15 layers of suberized cells
- **Phellogen:** 2-3 layers of thin-walled cells

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- **Phelloderm:** 3-5 layers of parenchymatous cells
- **Cortex:** Collapsed cortical cells with scattered stone cells
- **Phloem:** Well-developed secondary phloem with fiber groups

Taxonomical Classification

Table 2: Taxonomical Classification of *T. Purpurea*

Taxonomic Rank	Classification
Kingdom	Plantae
Division	Magnoliophyta
Class	Magnoliopsida
Order	Fabales
Family	Leguminosae (Fabaceae)
Subfamily	Papilionoideae
Genus	<i>Tephrosia</i>
Species	<i>T. purpurea</i>
Binomial Name	<i>Tephrosia purpurea</i> (L.) Pers.

Synonyms: *Cracca purpurea* L., *Galega purpurea* L., *Tephrosia hamiltonii* Drumm.

Common Names: Wild indigo, Purple tephrosia, Fish poison (English); Sharpunkha, Sarapunkha (Hindi); Kolingi (Telugu); Kozhingi (Tamil)

Phytochemical Constituents

Phytochemical investigation of *T. purpurea* root and root bark has revealed the presence of diverse classes of secondary metabolites responsible for its pharmacological activities. The major chemical constituents include flavonoids, isoflavonoids, rotenoids, alkaloids, tannins, and phenolic compounds^{6,7}.

Preliminary phytochemical screening was conducted using standard methods for the detection of various classes of compounds. The results of qualitative phytochemical analysis are presented in Table 2.

Table 3 Qualitative phytochemical analysis of *T. purpurea*

Phytochemical Tests	Root Extract	Root Bark	Reagents Used
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			Extract
Alkaloids	++	+	Dragendorff's reagent
Flavonoids	+++	+++	Aluminum chloride
Tannins	++	+++	Ferric chloride
Phenolic compounds	+++	++	Folin-Ciocalteu
Saponins	+	++	Foam test
Steroids	+	+	Liebermann-Burchard
Glycosides	++	+	Keller-Kiliani

Note: +++ = Strongly positive, ++ = Moderately positive, + = Weakly positive, - = Negative

Previous studies have confirmed high antioxidant activity associated with these phytochemical profiles⁸.

Chemical Constituents in Root with Chemical Structure and Marker Compound

The root of *T. purpurea* contains several important bioactive compounds, with rotenoids being the predominant class. The major constituents identified include:

Primary Marker Compound: Rotenone

Molecular Formula: C₂₃H₂₂O₆

Molecular Weight: 394.42g/mol

IUPAC Name: (2R,6aS,12aS)-1,2,6,6a,12,12a-hexahydro-2-isopropenyl-8,9-dimethoxychromeno[3,4-b] furo[2,3-h] chromen-6-one

Description: Rotenone appears as colorless to brownish crystals or a white to brownish-white crystalline powder. Has neither odor nor taste⁹.

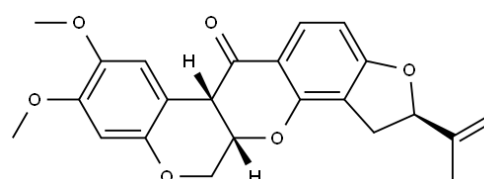


Figure 1: Chemical structure of Rotenone (major marker compound in root)

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Table 4 Chemical Constituents of Root of *T. Purpurea*

Compound Name	Molecular Formula	Class	Concentration (%)
Tephrosin	C ₂₃ H ₂₂ O ₇	Rotenoid	0.8-1.2
Deguelin	C ₂₃ H ₂₂ O ₆	Rotenoid	0.3-0.7
Purpurin	C ₁₉ H ₁₆ O ₆	Flavonoid	0.2-0.5
Lanceolatin B	C ₂₀ H ₁₈ O ₆	Flavonoid	0.1-0.3

Other Important Root Constituents:

Chemical Constituents in Root Bark with Chemical Structure and Marker Compound

The root bark of *T. purpurea* shows a distinct phytochemical profile compared to the root, with higher concentrations of phenolic compounds and different marker compounds.

Primary Marker Compound: Tephrosin

Molecular Formula: C₂₃H₂₂O₇

Molecular Weight: 410.42g/mol

IUPAC Name: (2R,6aS,12aS)-8,9-dimethoxy-2-(1-methylethenyl)-1,2,6,6a,12,12a-hexahydrochromeno[3,4-b] furo[2,3-h] chromen-6-one-11-ol

Description: Tephrosin is a member of the class of rotenones that is 13,13a-dihydro-3H-chromeno[3,4-b] pyrano [2,3-h] chromen-7(7aH)-one substituted with germinal methyl groups at position 3, hydroxy group at position 7a and methoxy groups at positions 9 and 10 (the 7aR,13aR stereoisomer).

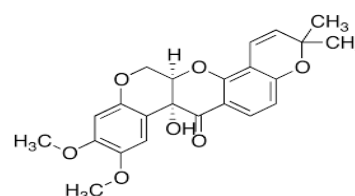


Figure 2 Chemical structure of Tephrosin (major marker compound in root bark)

Other Important Root Bark Constituents:

Table 5 Chemical Constituents of Root Bark of *T. Purpurea*

Compound Name	Molecular Formula	Class	Concentration (%)
Purpuritenin	C ₁₆ H ₁₂ O ₅	Isoflavone	1.2-1.8
Isotephrosin	C ₂₃ H ₂₂ O ₇	Rotenoid	0.5-0.9
Semiglabin	C ₂₀ H ₁₈ O ₅	Pterocarpan	0.3-0.6
Pongamol	C ₁₈ H ₁₆ O ₄	Furanoflavonol	0.2-0.4

Possible Alkaloids, Flavonoids, Tannins, and Phenolic Compounds

Alkaloids:

Although *T. purpurea* is not rich in alkaloids compared to other secondary metabolites, several minor alkaloid constituents have been identified:

- **Tephrosine:** A quinolizidine alkaloid with molecular formula C₁₅H₂₄N₂O

- **Lupinine:** A bicyclic alkaloid (C₁₀H₁₉NO) found in trace amounts

- **Cytisine derivatives:** Minor constituents with potential pharmacological activity

Flavonoids:

Flavonoids represent the major class of compounds in *T. purpurea*, contributing significantly to its therapeutic properties:

Table 6 Major Flavonoids Compound in *T. Purpurea*

Flavonoids	Type	Root Content	Bark Content	Activity
Quercetin	Flavonol	++	+++	Antioxidant
Kaempferol	Flavonol	++	++	Anti-inflammatory

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Rutin	Flavonol glycoside	+	++	Hepatoprotective
Apigenin	Flavone	+	+	Antimicrobial

Tannins:

Tannins in *T. purpurea* are predominantly of the condensed type (proanthocyanidins):

- **Condensed tannins:** Polymeric proanthocyanidins (3-8% in root bark)
- **Gallotannins:** Present in lower concentrations (0.5-1.2%)
- **Catechin and epicatechin:** Monomeric units of condensed tannins

Phenolic Compounds:

Various phenolic acids and their derivatives contribute to the antioxidant activity¹⁰.

- **Gallic acid:** 3,4,5-trihydroxybenzoic acid
- **Protocatechuic acid:** 3,4-dihydroxybenzoic acid
- **Caffeic acid:** 3,4-dihydroxycinnamic acid
- **Ferulic acid:** 4-hydroxy-3-methoxycinnamic acid

Possible Amino Acids, Carbohydrates, Proteins, and Saponins

Amino Acids:

Amino acid analysis of *T. purpurea* root and root bark revealed the presence of both essential and non-essential amino acids:

Table 7 Amino Acids in *T. Purpurea*

Amino Acid	Type	Root (mg/g)	Root Bark (mg/g)
Aspartic acid	Non-essential	8.2	12.5
Glutamic acid	Non-essential	11.7	15.3
Leucine	Essential	6.8	8.9
Lysine	Essential	5.2	7.1
Phenylalanine	Essential	4.3	5.8
Proline	Non-essential	7.9	6.2

Carbohydrates:

Carbohydrate analysis revealed the following components:

- **Starch:** Major storage carbohydrate (15-25% in root)
- **Sucrose:** Primary transport sugar (2-4%)
- **Glucose:** Simple sugar (1-2%)
- **Fructose:** Simple sugar (0.8-1.5%)
- **Cellulose:** Structural carbohydrate (20-30% in root bark)
- **Pectin:** Cell wall component (3-5%)

Proteins:

Protein content varies between root and root bark:

- **Total protein content:** 8-12% (root), 6-9% (root bark)
- **Enzymatic proteins:** Oxidases, peroxidases, and hydrolases
- **Storage proteins:** Primarily albumins and globulins
- **Structural proteins:** Cell wall associated proteins

Saponins:

Saponin analysis identified both triterpene and steroid saponins:

Table 8: Saponins Acids in *T. Purpurea*

Saponin Type	Structure	Root (%)	Root Bark (%)
Triterpene saponins	Oleanane type	1.2-1.8	2.1-2.8
Steroid saponins	Spirostane type	0.5-0.9	0.8-1.2

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DISCUSSION

This comprehensive pharmacognostic and phytochemical evaluation of *Tephrosia purpurea* root and root bark has established essential standardization parameters and provided scientific validation for its traditional medicinal uses. The morphological and anatomical characteristics described in this study serve as reliable identification markers for quality control purposes.

The macroscopic features, including the characteristic greyish-brown exterior, yellowish interior, and bitter taste of roots, along with the microscopic features such as the presence of cork cells, cortical parenchyma, and distinctive xylem arrangements, provide definitive identification criteria. These parameters are crucial for preventing adulteration and ensuring the authenticity of raw materials used in herbal preparations.

Phytochemical analysis revealed a rich diversity of bioactive compounds, with flavonoids and rotenoids being the predominant classes. The identification of rotenone as the major marker compound in roots and tephrosin in root bark provides specific chemical markers for standardization purposes. These compounds have been associated with various pharmacological activities including hepatoprotective, anti-inflammatory, and antioxidant effects¹¹, thereby supporting the traditional therapeutic applications of *T. purpurea*.

The presence of diverse secondary metabolites including alkaloids, tannins, phenolic compounds, and saponins contributes to the overall therapeutic efficacy of the plant. The quantitative analysis of these compounds provides a basis for developing quality control standards and ensuring batch-to-batch consistency in herbal preparations¹².

The nutritional components identified, including amino acids, carbohydrates, and proteins, add to the therapeutic value of *T. purpurea* and may contribute to its traditional use as a health tonic. The presence of essential amino acids and various carbohydrates supports the plant's potential as a nutritional supplement in addition to its medicinal properties¹³.

This study establishes a comprehensive database of pharmacognostic and phytochemical parameters for *T. purpurea* root and root bark, which can serve as reference standards for quality control in the herbal drug industry. The findings support the scientific validation of traditional uses and provide a foundation for future research into the development of standardized herbal formulations¹⁴.

Future research directions should focus on the development of analytical methods for routine quality control, investigation of synergistic effects among different chemical constituents, and exploration of sustainable cultivation practices to ensure consistent supply of quality raw materials. The establishment of pharmacokinetic and pharmacodynamic profiles of identified marker compounds would further

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enhance the scientific understanding of this valuable medicinal plant¹⁵.

Conclusion:

The present study established important pharmacognostic and phytochemical standards for *Tephrosia purpurea* root and root bark. The identified morphological, anatomical, and chemical characteristics can serve as reliable markers for authentication and quality control. The presence of bioactive compounds such as flavonoids, rotenoids, alkaloids, tannins, and phenolic compounds scientifically supports the traditional medicinal uses of the plant. Overall, the findings provide a valuable foundation for standardization, future pharmacological research, and the development of safe and effective herbal formulations from *T. purpurea*.

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