



**PHYTOCHEMICAL SCREENING AND TLC PROFILING OF *TEPHROSIA PURPUREA*
LEAVES AND STEM**

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ABSTRACT

Tephrosia purpurea is a Ayurvedic plant commonly known in Sanskrit as 'sharapunkha' belong to family Fabaceae. The whole plant and various parts of the plant are useful as ayurvedic medicines. The pharmacological studies have shown that *Tephrosia purpurea* possesses biological activities such as antibacterial, antimicrobial, antioxidant and wound healing properties. In the present study phytochemical analysis and thin layer chromatography profiles were performed on different solvent extractions like methanol, ethanol, petroleum ether, and chloroform extract of *Tephrosia purpurea*. The phytochemical analysis done in various extracts showed the presence of carbohydrates, flavonoids, steroids, protein, resident sugar, tannins, glycerols. The TLC profiling of leaves and stem extract on different solvent systems showed the presence of various phytochemicals.

KEYWORDS: *Tephrosia purpurea*, TLC, phytochemical, Plant extracts.

INTRODUCTION

In Ayurveda is mainly originated in India long back in prevedic period. Ayurveda means science of life and people are concerned more about their applications. Now people are more concerned about its various importance. *Tephrosia purpurea* is a genus which belongs to family Fabaceae. The genus name derives from the Greek word 'téphr' meaning 'ashes, ash coloured and grey' as the colour of the stem, leaves, and fruits of all the species are grey in colour. In Ayurvedic literature *Tephrosia purpurea* has been described as "sarva warnavishapak" means it has the property to heal all types of wounds. Traditionally, the drug is used as a liver tonic. *Tephrosia purpurea* is commonly known as wild indigo and in Tamil 'kolanji'. *Tephrosia purpurea* is also a perennial herb, found in dry, gravelly or rocky sandy soil. These plants have branched, suberect, slender, herbaceous perennial green climber. *Tephrosia purpurea* plant possesses antibacterial^[5] and antifungal properties. The whole plant and its roots are used for medicinal purposes. This plant part has been used for hydrophobia, asthma, cough, heart disease, and kidney problems. The plant has anti-inflammatory, depurative, styptic, alexiteric and antipyretic properties also. It is also used in thermogenic, anthaminitic, digestive, laxative, diuretic. The plant extracts or secondary metabolites have served as antioxidants^[8,9] in phytotherapeutic medicines to protect against various diseases for centuries. The plant is used in folk medicine as an anti-diabetic, antipyretic, anticancer,

and anti-ulcer agent in addition to its usefulness in the treatment of diseases related to oxidative stress and free radical activity. They play an important role in the progression of various diseases like ageing, respiratory diseases, cancer, and gout. The roots are bitter and the decoction is used as a nematicide for treatment. The whole plant may be used for its rich flavonoid^[1] and polyphenol content. Though a lot of research is going on in the plant. The infusion of seed is used as an anthelmintic oil. Leaves of the plant are used in dyspepsia, pectoral diseases, haemorrhoid, syphilis, gonorrhoea. Leaves are also used as fodder in India and South Africa. Seeds are used as a substitute for coffee and as an insect repellent.

MATERIALS AND METHODS

Preparation of plant extract: The whole plant of *Tephrosia purpurea* was collected from the college garden. The plant material was washed thoroughly 2-3 times with running tap water. All the plant materials were then air-dried under shade for 15-20 days. Powder samples of leaves and stems were carried out separately at room temperature for 72 hours in methanol, chloroform, petroleum-ether, ethanol and it was kept in a rotatory shaker for 72 hours. After that the different plant extracts of leaves and stems were filtered by using Whatman no-1 filter paper.

The phytochemical analysis was done on various extracts of *Tephrosia purpurea* as prescribed by

Harbone^[7], Siddiqui and Ali to detect the presence of the active chemical constituents like tannins, steroids, flavonoids, glycosides, terpenoid, saponins, quinone

Test for tannins: To the 0.5ml plant extract 1ml water and 1-2 drop ferric chloride. The blue colour is observed shows the presence of tannins.

Test for glycosides: 2ml plant extract 1ml glacial acetic acid and few drops of ferric chloride and conc sulphuric acid. Reddish brown colour is observed.

Test for flavonoids: To test the presence of flavonoids to the 2ml plant extract few drops of conc. HCL and magnesium ribbon was added. Pink tomato red color is observed.

Test for protein: For protein test 1ml extract few drops of nitric acid by the side of test tube yellow colour is observed shows the presence of protein.

Test for carbohydrates: 1 ml of Fehling of A and B Solution were added in diluted extract and heated. Brick red color is observed

Test for Quinone: To test the presence of quinone to the 1ml extract add 1ml con H₂SO₄. Red color is indicates the presence of quinone.

Test for steroids

1ml extract was treated with 10ml chloroform and Con. H₂SO₄ side of test tube. Upper layer turns red while sulphuric acid layer indicates yellowish green colour.

Test for terpenoids

To the 5ml extract 2ml chloroform was added. To this 3ml Con H₂SO₄ was mixed to form a layer. Appearance of reddish brown indicates the presence of terpenoids.

TLC Profiling of *Tephrosia purpurea* leaves and stem:

Isolation of components was done by thin layer chromatography. Thin layer chromatography (TLC) was carried on analytical plates over silica gel-G for each extract with different solvent mixtures used as developing systems. 1.5µl test sample was loaded in TLC plate with silica gel. The TLC plates were placed in TLC chamber which was saturated with mobile phase. In each case the TLC plates were exposed to iodine vapour and reported spots were visualized and noted down. The R_f Values for all studied TLC was calculated.

RESULT

Table 1: Phytochemical Evaluation of *Tephrosia purpurea* leaf extract

Test	Different solvents leaf extract			
	Methanol	Ethanol	Chloroform	Petroleum ether
Tannins	+	-	+	+
Steroids	+	+	+	+
Glycosides	+	-	+	-
Flavonoids	+	+	-	+
Quinone	+	-	+	-
Terpenoids	+	+	+	-
Carbohydrate	+	+	+	-
Protein	+	+	-	+

Table 2: Phytochemical Evaluation of *Tephrosia purpurea* stem extract

Test	Different solvents stem extract			
	Methanol	Ethanol	Chloroform	Petroleum ether
Tannins	+	-	+	+
Steroids	+	+	-	-
Glycosides	+	-	+	-
Flavonoids	+	+	+	+
Quinone	+	-	+	+
Terpenoids	+	+	+	-
Carbohydrate	+	+	+	+
Protein	+	+	+	+

Table 3: Different Solvents Used For TLC Analysis Of *Tephrosia purpurea*

DEVELOPING SYSTEM	DEVELOPING SYSTEM SOLVENT	RATIO
System - 1	Chloroform - hexane	7.5:2.5
System -2	Ethyl acetate; Methanol	1:1
System -3	Chloroform; Glacial acetic acid; methanol; water	60:32:12.8

Table 4: Calculated R_f Values for *Tephrosia purpurea* leaf extract TLC systems.

Plant extract	System 1	System 2	System 3
Methanol	0.88	0.98	0.94
Ethanol	0.74	0.83	0.94
Petroleum Ether	0.56	0.40	0.09
Chloroform	0.29	0.31	0.45

Table 5: Calculated R_f Values for *Tephrosia purpurea* stem extract TLC systems.

Plant extract	System 1	System 2	System 3
Methanol	0.90	0.82	0.84
Ethanol	0.66	0.57	0.65
Petroleum Ether	0.13	0.27	0.32
Chloroform	0.35	0.09	0.28

Qualitative phytochemical analysis is an important step towards discovery of new drug indicating the presence of primary or secondary metabolites.^[10] Phytochemical analysis performed on different solvent extracts of *Tephrosia purpurea* leaf shows the presence of Tannins, Steroids, Glycosides, Flavonoids, Quinone, and stem extracts showed the presence of Terpenoids, Carbohydrate, Protein. Flavonoids are nearly reported in all the extracts. Petroleum ether extracts of both the plant part showed lesser amount of phytochemicals. The results of phytochemical analysis are reported in table 1 and 2. The presence of these phytochemical compounds are further analyzed by thin layer chromatography in three different solvents for various plant extracts. The R_f values for *Tephrosia purpurea* leaf and stem extracts in different developing solvent are reported in table 4 and 5.



Petroleum Ether Extract

Figure 1: Phytochemical Evaluation of *Tephrosia purpurea* Leaf Extract

Methanol Extract



Methanol Extract



Ethanol Extract



Ethanol Extract



Chloroform Extract



Chloroform Extract



Petroleum Ether Extract

Figure 2: Phytochemical Evaluation of *Tephrosia purpurea* Stem Extract

DISCUSSION

Tephrosia purpurea plant parts screened for phytochemical constituents seemed to have potential as source of drugs and also to improve the health and disease of its users.^[3] All major tested phytochemicals are reported in various extracts of leaves and stem of *Tephrosia purpurea*. The presence of various phytochemical compounds that are vital have been found mostly in methanol and ethanol extract. The phytochemicals screening performed on the stem and leaves extracts of *Tephrosia purpurea* indicates the presence of most of phytochemicals. The presence of these significant bioactive natural product indicates the necessity of separation of the compounds from the mixture of compounds through thin layer chromatographic techniques. Separation technique was performed on different solvent system. R_f values for leaf and stem extracts in different solvent system was calculated. In the present study in methanol extract quinone, steroids flavonoids, glycosides and terpenoids have been found. In case of ethanol extract tannin, quinone, flavonoids, and terpenoids, are confirmed. The present studies analysis provides very useful information which may helpful in authenticating the plant along with the nature of phytoconstituents present in it.

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