RESEARCH ARTICLE

PHYTOCHEMICAL INVESTIGATION BY USING TENDER LEAF PART OF GREWIA TILIIFOLIA VAHL.

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ABSTRACT

This study was aimed to evaluate the phytochemical potential of different extracts of tender leaf part of *Grewia tiliifolia* Vahl which are commonly used in Ayurveda drug preparations. Tender leaf part of *G.tiliifolia* subjected to analyze the phytochemical constituents by using qualitative and quantitative methods. The results of the present investigation revealed that the presence of flavonoid, phenol, tannin glycoside, resin, steroids, terpenoids and triterpenoids in different solvent extract like petroleum ether, chloroform, ethyl acetate, methanol and water. Tender leaves of the species which exhibited well marked potential activity and rich in secondary metabolite contents (flavonoids and phenols).

Keywords: Grewia tiliifolia Vahl, Tender leaf part, Phytochemical screening,

1. INTRODUCTION

During last few decades there has been an increase in the study of medicinal plants and their traditional use in different parts of the world (1). A wide range of bioactive substances present in plants have traditional medicine that can be used for the treatment of infectious diseases (2). Recently, researchers all over the world focused on finding naturally occurring medicines from plants. The genus Grewia, (Family: Tiliaceae) is an important medicinal plant which comprises of shrubs and trees and is distributed in the warmer parts of the world. Nearly 40 species of this genus are found in India some of which are well known for their medicinal value (3, 4). Avurveda, the ancient Indian treatise on medicine, mentions the use of different plant parts of Grewia to cure inflammation, burning sensation, fever, blood disorders, wound healing, ulcerative colitis, heavy menstrual flow and diabetes etc. (5). Grewia tiliifolia Vahl is a medium sized tree up to 20 m in height, leaves simple, alternate, and ovate with oblique base, crenate-denate, acuminate, upper surface minutely stellately hairy. It is useful in vitiated conditions of kapha and pitta, burning sensation, hyperdipsia, rhinopathy, ulcers, skin diseases, haematemesis and general debility (6). Therefore, considering the traditional use of the plant, the present study has been designed to investigate the phytochemical constituent of different solvent extract of G. tiliifolia in tender leaf part.

2. MATERIALS AND METHODS

2.1. Preparation of extract

Tender leaf part of *G tiliifolia* was collected from the Western Ghats region of Malappuram district, Kerala India. Plant materials (tender leaf part of plant) was collected and washed with distilled water and shade dried for a week. The dried sample were manually ground to fine powder using pulverizer and passed through 40 mesh sieve and stored in air tight containers. The coarsely powdered plant material was weighed to 50g and Soxhlet extracted with petroleum ether, chloroform, ethyl acetate and methanol separately for 12 hours. The filtrate was evaporated to dryness under reduced pressure using rotary vacuum evaporator and the solid mass obtained was stored at 4°C until further use. The stored filtrate was used for the various phytochemical and pharmacological studies.

2.2. Phytochemical Screening of tender leaf parts of G. tiliifolia.

2.2.1. Alkaloids test: (Mayer's test)

The plant extract was evaporated to dryness and the residue was heated on a boiling water bath with 2% hydrochloric acid. After cooling, the mixture was filtered and treated with a few drops of Mayer's reagent. Formation of turbidity or yellow precipitation showed the presence of alkaloid.

2.2.2. Glycosides:

Glycosides are compounds which upon hydrolysis give rise to one or more sugars (glycone) and a compound which is not a sugar (glycone) to the solution of the extract, few drops of sodium hydroxide was added, and observed for yellow color which shows the presence of glycosides.

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2.2.3. Terpenoids and steroids: (Salkowski's test)

1 ml of extract was taken in a boiling tube and 2 ml of concentrated sulphuric acid was added slowly and red violet color was observed for terpenoid and green bluish color for steroids.

2.2.4. Flavonoids: (Ferric chloride test)

In a test tube containing 1ml of extract, 5-6 drops of dilute hydrochloric acid was added and small pieces of magnesium were added. Red color was observed for flavonoids and orange color for flavones.

3.1.1. Saponins: (Foam test)

1gm of extracts was added to 5ml of distilled water in a test tube. The solution was shaken vigorously and observed for a stable persistent froth which indicate the presence of saponin.

3.1.2. Phenols: (Ferric chloride test)

1ml of extract was taken in a test tube, to this few drops of neutral 5% ferric chloride solution are added. A dark green color indicates the presence of phenolic compounds.

3.1.3. Tannins: (Braemer's test)

1ml of extract solution 1-2 drops of lead acetate solution was added. Red precipitate was formed indicating the presence of tannins.

3.1.4. Cardiac glycosides: (Keller-killani's test)

5ml of extract was taken in a boiling tube to which 2ml of glacial acetic acid containing one drop of ferric chloride solution was added and 1ml of concentrated sulphuric acid was added slowly. Appearance of brown ring indicates the presence of cardiac glycosides.

2.2.9. Resin: (sulphuric acid test)

5ml of extract was taken in a boiling tube to which 2-3ml of acetic anhydride was added, dissolved by gentle heating and 0.5ml of sulphuric acid was added. Bright purple color was produced it indicates the presence of resin.

2.2.10. Triterpenoids

2ml of extract was added with 1 ml of acetic anhydride followed by the addition of 2ml concentrated sulphuric acid. Formation of reddish violet color indicates the presence of triterpenoids.

2.2.11. Reducing sugar

The crude extract was shaken with 5 ml of distilled water and filtered. The filtrate was boiled with drops of Fehling's solution A and B for 2 minutes. An orange red precipitate indicates the presence of reducing sugar.

3. RESULTS AND DISCUSSION

The present study investigates the phytochemical potential of tender leaf part of *G. tiliifolia*.

Table 1. Qualitative phytochemical screening in different extracts of tender leaf plant part of *G. tiliifolia*

Secondary Metabolites	Р	C	EA	М	A
Alkaloid	-	-	-	-	-
Flavonoid	+++	_	_	_	++
Phenol	+++	++	_	+++	_
Tannin	_	_	_	++	+
Glycoside	++	+	_	+++	++
Saponin	_	_	_	_	_
Resin	_	+++	_	+++	+
Steroids	_	+++	_	+++	++
Terpanoid	_	_	_	+++	++
Cardiac glycosides	_	_	_	-	_
Triterpenoids	_	+++	_	+++	++
Reducing sugar	_	_	_	_	_
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P- Petroleum Ether; C – Chloroform; EA – Ethyl Acetate; M – Methanol; A – Aquous.

Table 2. FT-IR Peak value and its functional groups in methanol extract of tender leaf Plant part of *G. tiliifolia*

S.No.	FT-IR Peak Values	Functional groups	
1.	995.27	C=C STRETCH (Benzene)	
2. 3.	1056.99 1288.45	C-O STRETCH (Alcohol) C-H STRETCH (Alcane)	
4.	1435.04	C=C STRETCH (Benzene)	
5.	1620.21	N-H STRETCH(Amine)	
6.	1697.36	C=O STRETCH (Carbonyl)	
7.	2476.60	N-H STRETCH(Amine)	
8.	2762.06	N-H STRETCH(Amine)	
9.	2862.36	O-H STRETCH(Carboxylic acid)	
10.	3086.11	O-H STRETCH(Carboxylic acid)	
11	3950.22	N-H STRETCH(Amine)	

Table 3. Total Phenol content of tender leaf plant part of G. tiliifolia with different solventExtracts.

Solvents	Sample (µl)	Total Phenol Content
Petroleum ether		2.4
Chloroform Ethyl acetate	20	0.88 0.21
Methanol		0.87

Table 4. Total flavanoid content of tender leaf plant part of *G. tiliifolia* with different Solvent extracts.

Solvents	Sample (ul)	Total Flavanoid		
	Sumple (µI)	Content		
Petroleum ether		38.17		
Chloroform	500	38.45		
Ethyl acetate		48.1		
Methanol		21.52		



Fig. 1. FT-IR analysis of tender leaf plant part of *G. tiliaefolia* in methanol extract.



Fig. 2. Images of the tender leaf part of *G. tiliaefolia.*

3.1. Phytochemical Analysis

G. tiliifolia (tender leaf powder 30g) were extracted with 4 solvents, viz; petroleum ether (1.0g), chloroform (1.2g), ethyl acetate (2.1g), methanol (3.1g) and water (3.5g). The extractive values were useful to evaluate the chemical constituents present in the crude extract (drug). The present study was undertaken to evaluate the presence of active principles in Grewia tiliifolia which is highly medicinal. The preliminary phytochemical qualitative test of extracts confirmed in the presence of flavonoid, phenol, tannin glycoside, resin, steroids, terpanoids and triterpanoids. The total phenolic content in the petroleum ether extract of tender leaf part of G. *tiliifolia* measured high (2.4µg). Ethyl acetate extract of tender leaf part *G. tiliifolia* measured very least value (0.21 µg). Highest content of total flavonoids (48.1 $\mu g)$ is found in ethyl acetate extract while methanol showed very low flavonoid (21.5 μg). The presence of the secondary metabolites in the investigated plants account that it can be used as medicinal plants.

3.2. Fourier Transform Infrared Spectrophotometer (FTIR)

FTIR spectroscopic analysis of the tender leaves of *G. tiliifolia* methanolic extract with infrared spectroscopy revealed the presence of C=C, C-O, C-H, C=C, N-H, C=O, N-H, O-H bonds stretching. The peaks revealed that the plant sample had the compounds like Benzene, Alcohol, Alcane, Amine, Carbonyl and Carboxylic acid.

4. CONCLUSION

Grewia tiliifolia is an important traditional folk medicine. Tender leaves of the species which exhibited well marked potential activity and rich in secondary metabolite contents. Thus the results obtained from this investigations indicate that tender leaf plant part extract of *G.tiliifolia*, rich in secondary metabolites and confirmed that the tender leaves have great importance as therapeutic agents in preventing diseases.

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