



Research Article

**PRELIMINARY PHYTOCHEMICAL SCREENING OF LEAF EXTRACT OF *ZIZIPHUS
GLABRATA* B. HEYNE EX ROTH**

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Abstract: Medicinal plants are an important source of phytochemicals that offer traditional medicinal treatment of various ailments. Medicinal plants are the local heritage with global importance. The present paper shows the therapeutic importance of *Ziziphus glabrata* leaves and features its medicinal character. The leaves of *Ziziphus glabrata* were collected, shadow dried and extracted with petroleum ether, chloroform, benzene, ethyl acetate, ethanol, hydroalcohol and water. The preliminary phytochemical screening of *Ziziphus glabrata* leaves was performed using standard procedures and the results revealed the presence of alkaloids, flavonoids, phenols, steroids and tannins in leaf extracts of *Ziziphus glabrata*.

Keywords: Phytochemical, *Ziziphus glabrata*, Alkaloids, Flavonoids.

INTRODUCTION

Medicinal herbs are an important source for the therapeutic remedies of various ailments¹. Medicinal herbs constitute effective sources of antimicrobial and antioxidant natural products². Natural phytochemicals derived from medicinal plants have gained significant recognition in the potential management of several human clinical conditions, including cancer³.

Many of these indigenous medicinal plants are used as spices and food plants. They are also sometimes added to foods meant for pregnant and nursing mothers for medicinal purposes⁴. Medicinal plants contain some organic compounds which provide definite physiological action on the human body and these bioactive substances include tannins, alkaloids, carbohydrates, terpenoids, steroids and flavonoids^{5, 6}. According to World Health Organization (WHO), medicinal plants would be the best source to obtain variety of drugs. About 80% of individuals from developed countries used traditional medicines, which have compounds derived from medicinal plants⁷.

Ziziphus glabrata Heyne (Syn: *Z. Trinervia* Roxb), is a small tree that grows up to 30 ft in height, having olive-brown wood and commonly found in the forests of Peninsular India and Bhutan. The leaves and aerial parts of the plant are traditionally used to treat inflammation, to relieve pain, convulsions and viral infections⁸. Plants belonging to the genus *Ziziphus* (Rhamnaceae) have been noted to produce a variety of characteristic secondary metabolites ranging from cyclopeptide alkaloids that possess antibacterial and antifungal activities⁹, and the dammarane class of triterpenes that are reported as sweetness inhibitor^{10, 11}.

MATERIALS AND METHODS

Collection and identification of plant materials

Ziziphus glabrata leaves were collected from the Metala Hills of Namakkal, Tamil Nadu and authenticated by Dr.A.Balasubramanian, Executive Director, ABS Botanical

Conservation, Research and Training Centre, Salem. Leaves were taken for the investigation of phytochemical analysis. The leaves were washed thoroughly 2-3 times with running tap water, leaf material was then air dried under shade. The plant material was grinded and powders were kept in small plastic bags.

Preparation of plant extracts

The grinded leaf powder of 25g/250ml was taken in a conical flask and it was immersed by the organic solvent (Petroleum ether, Chloroform, Benzene, Ethyl acetate, ethanol, Hydroalcohol (80% of aqueous ethanol) and water. The flask was then plugged with cotton wool and then kept at room temperature for 48 hrs. After two days, the supernatant was collected and the solvent was evaporated to make the final volume one-fourth of the original volume and stored at 4°C in air tight bottles.

Preliminary phytochemical screening

The various extracts of *Ziziphus glabrata* leaves were subjected to preliminary phytochemical screening¹².

Alkaloids:

Mayer's Test: A small quantity of the extract was treated with few drops of dilute hydrochloric acid and filtered. The filtrate was tested with alkaloid Mayer's reagent. Formation of cream precipitate indicated the presence of alkaloids.

Dragendroff's Test: To 2-3ml of the extract added few drops of Dragendroff's reagent. Formation of orange red (or) reddish brown precipitate indicated the presence of alkaloids.

Wagner's Test: To 2-3 ml of the extract added few drops of Wagner's reagent. Formation of reddish brown precipitate indicated the presence of alkaloids.

Flavonoids:

Sodium hydroxide Test: To 2-3 ml of the extract, few drops of sodium hydroxide solution were added in a test

tube. Formation of intense yellow colour that became colourless on addition of few drops of dilute HCl indicated the presence of flavonoids.

Phytosterols:

Salkowski Test: To 2 ml of extract, added 2ml chloroform and 2 ml concentrated sulphuric acid and was shaken well. Appearance of red Chloroform layer and greenish yellow fluorescence acid layer indicated the presence of sterols.

Glycosides:

5 ml of extract was dissolved in pyridine followed by the addition of the freshly prepared sodium nitroprusside solution. The formation of pink to red colour indicated the presence of glycosides.

Phenols:

0.5 ml of ferric chloride solution was added to 2 ml of test solution. Formation of blue-green or red colour indicated the presence of phenols.

Tannins:

Small quantity of extract was boiled in 20 ml of water in a test tube and then filtered. A few drop of 0.1% ferric chloride was added and observed for brownish green or blue-black coloration which indicated the presence of tannins.

Carbohydrates:

Benedict's test: The extract was treated with Benedict's reagent and heated for few minutes. Formation of brick red precipitate shows the presence of reducing sugar.

Fixed oils and fats (spot test):

Press a small quantity of powder between two filter papers. Oil stains on the filter paper indicated the presence of fixed oils.

RESULTS

In the present investigation, preliminary phytochemical screening has been done in the various extract of *Ziziphus glabrata* leaves and the results are presented in Table-1.

Table 1: Phytochemical analysis of leaf extract of *Ziziphus glabrata*

S.No.	Solvent/ Phytochemical constituents	Petroleum ether	Chloroform	Benzene	Ethyl acetate	Ethanol	Hydro alcohol	Water
1	Alkaloids	+	+	+	+	+	+	+
2	Flavonoids	+	+	+	+	+	+	+
3	Glycosides	+	+	+	+	+	+	+
4	Steroids	+	-	+	+	+	+	+
5	Phenol	-	-	-	-	+	+	+
6	Tannin	-	+	+	+	+	+	+
7	Carbohydrate	-	-	-	-	-	+	+
8	Fixed oil and Fat	+	+	+	+	+	+	+

The petroleum ether extract showed the presence of alkaloids, flavonoids, glycosides, steroids and fixed oil and fat. The chloroform extract showed the presence of alkaloids, flavonoids, glycosides, tannin and fixed oil and fat. The benzene and ethyl acetate extract showed the presence of alkaloids, flavonoids, glycosides, steroids, tannin and fixed oil and fat. The ethanol extract showed the presence of alkaloids, flavonoids, glycosides, steroids, phenol, tannin and fixed oil and fat. The results showed the presence of all the phytochemicals in the hydroalcoholic and aqueous extract of *Ziziphus glabrata* leaves.

The results of phytochemical analysis comprehensively validate the presence of therapeutically important and valuable secondary metabolites like alkaloids, flavonoids, phenols, tannins and steroids in *Ziziphus glabrata* leaves.

DISCUSSION

Alkaloids:

In this study a cream or reddish brown precipitate was observed in petroleum ether, chloroform, benzene, ethyl acetate, ethanol, hydroalcoholic, and aqueous extract of *Ziziphus glabrata* leaves.

The alkaloids have been associated with medicinal uses for centuries and one of their common biological properties is cytotoxicity¹³. Alkaloids and its derivatives played important role in analgesic, antispasmodic and

bactericidal activities¹⁴. The alkaloids have been investigated for many pharmacological properties including antiprotozoal, cytotoxic, antidiabetic and anti-inflammatory properties¹⁵.

Flavonoids:

In this study a yellow precipitate (or) cherry red colour was observed in petroleum ether, chloroform, benzene, ethyl acetate, ethanol, hydroalcoholic, and aqueous extract of *Ziziphus glabrata* leaves.

Flavonoids are hydroxylated phenolic substance known to be synthesized by plants in response to microbial infection and they have been found to be antimicrobial substances against wide array of microorganisms *invitro*¹⁶. Flavonoids have been reported to possess many useful properties, including anti-inflammatory, antiallergic, antioxidant, vascular and antitumour activity¹⁷.

Glycosides:

In this study a pink to red colour was observed in petroleum ether, chloroform, benzene, ethyl acetate, ethanol, hydroalcoholic and aqueous extract of *Ziziphus glabrata* leaves.

Glycosides are known to lower the blood pressure according to many reports¹⁸. Glycosides were reported to exhibit antidiabetic characteristics¹⁹.

Steroids:

The result indicated that a brown ring was formed at the junction of two layers in the petroleum ether, benzene,

ethyl acetate, ethanol, hydroalcoholic and aqueous extract of *Ziziphus glabrata* leaves.

Steroids have been reported to have antibacterial properties²⁰. Steroids have been reported to possess anti-inflammatory activities²¹. Plant steroids are known to be important for their cardiogenic activities, possession of insecticidal, anti-inflammatory, analgesic properties, central nervous system activities and antimicrobial properties²².

Phenols:

The results showed the presence of dark blue colour in ethanol, hydro alcoholic and aqueous extract of *Ziziphus glabrata* leaves.

The phenolic compounds are one of the largest and most ubiquitous groups of plant metabolites²³. Phenols and phenolic compounds are greatly used in skin infections and other wounds treatment and also for healing, when compared to other bactericides⁴. Primarily phenolic compound are of great importance as cellular part of cell wall structure by polymeric phenolics²⁴. They possess biological properties such as antiapoptosis, antiaging, anticarcinogen, antiinflammation, antiatherosclerosis, cardiovascular protection and improvement of endothelial function, as well as inhibition of angiogenesis and cell proliferation activities²⁵.

Tannins:

In this study a brownish green (or) bluish-black colour was observed in chloroform, benzene, ethyl acetate, ethanol, hydroalcoholic and aqueous extract of *Ziziphus glabrata* leaves.

Tannins bind to proline rich protein and interfere with protein synthesis²¹. Tannins decrease the bacterial proliferation by blocking key enzymes in microbial metabolism. Tannins act as potent antioxidant. Tannins are reported to be used for treating intestinal disorders such as diarrhoea and dysentery²⁶.

CONCLUSION

The present study attempts to assess the status of phytochemical properties in the leaves of *Ziziphus glabrata*. This study suggests that ethanol, hydroalcoholic and aqueous extract of *Ziziphus glabrata* leaves is a promising candidate which could be exploited further to develop as pharmacologically active agents due to their rich phytochemical constituents. Further work should be carried out to isolate, purify, and characterize the active constituents responsible for the activity of *Ziziphus glabrata*.

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