



International Journal of Pharmaceutical Research and Development (IJPRD)

Platform for Pharmaceutical Researches & Ideas

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PHYTOCHEMICAL SCREENING OF THREE MEDICINALLY IMPORTANT FICUS SP.

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ABSTRACT

Medicinal plants represent a rich source of antimicrobial agents. The traditional medicine involves the use of different plant extracts of bioactive constituents. Ficus is a huge tropical deciduous or evergreen tree with more than 800 species. Ficus benghalensis, Ficus religiosa, Ficus recemosa are important ingredients in many siddha, ayurveda and traditional formulations and used for the treatment of bacterial infections. An attempt was made to find out the phytoconstituents of Ficus sp., namely, Ficus benghalensis, Ficus religiosa, and Ficus recemosa. The aqueous and methanol extracts of the barks were obtained by standard methods. The extracts were undergone phytochemical analysis. The aqueous and methanolic extracts of Ficus benghalensis found to contain tannins, saponins and flavonoids. Ficus religiosa showed the presence of tannins, saponins, flavonoids and terpenoids. Ficus recemosa contain tannins, saponins, flavonoids and glycosides. The present study concludes that most of biologically active phytochemicals were present in the methanolic extract of three medicinally important Ficus sp.

Key words: Ficus benghalensis, Ficus religiosa, Ficus recemosa, phytoconstituents.

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INTRODUCTION

The present study was carried out to find out the phytoconstituents of three ficus sp., namely Ficus benghalensis, Ficus religiosa, Ficus recemosa. Medicinal plants have been used an exemplary source for centuries as an alternative remedy for treating human diseases because they contain numerous active constituents of therapeutic value

(Nostro et al., 2000). The potential of higher plants as source for new drugs is still largely unexplored. Thus, any phytochemical investigation of a given plant will reveal only a very narrow spectrum of its constituents. Plants are important source of potentially useful structures for the development of new chemotherapeutic agents. The genus *Ficus* includes some 750 species of woody plants

occurring in most tropical and subtropical forests throughout the world (Berg, 1989). The genus is remarkable for the large variation in the habits of its species (Jander and Machado 2008).

OBJECTIVE

Phytochemicals are naturally occurring, biologically active chemical compounds in plants. More than 4,000 of these compounds have been discovered to date and it is expected that scientists will discover more. The present study was aimed at investigating the phytoconstituents of *Ficus* sp.,

***Ficus benghalensis*:**

Ficus benghalensis belongs to the family Moraceae, which is commonly known as Banyan tree. *F. benghalensis* are fast growing, evergreen tree found in monsoon and rain forests, grow up to 3.0 meters, with spreading branches and many aerial roots. Leaves, stalked, ovate-cordate, 3-nerved entire, when young downy on both sides; petiole with a broad smooth greasy gland at the apex, compressed, downy; Fruit in auxiliary panicle, the size of a cherry, round and downy, Hardy, drought resistance and withstands mild frost.

Ficus religiosa

Ficus religiosa Linn (Moraceae) commonly known as 'Peepal tree' is a large widely branched tree with leathery, heart shaped, long tipped leaves on long slender petioles and purple fruits growing in pairs. The tree is regarded as a sacred tree to both Hindus as well as Buddhists. It has got mythological, religious and medicinal importance in Indian culture since ancient times (Singh, and Goel, 2009).

Ficus racemosa

Ficus racemosa synonymously also known as *Ficus glomerata*. An evergreen tree 50-60 ft. high; young shoot glabrous, pubescent or scaberrulous. Leaves 3-6 by 1.25-2.5 in. long, glabrous; stipules 0.75 in. long, ovate-lanceolate, scarious, pubescent. Peduncle shortly pendunculate, on short leafless warted branches often only a few inches long which issue from the stem and larger branches, much contracted at the base when young, subglobose, pyriform or

subturbinate, 1.5 in. across, smooth or pubescent, red when ripe.

Chemical constituents of the plants studied

Preliminary phytochemical investigation of root of *F. benghalensis* showed the presence of carbohydrates, flavonoids, amino acids/ proteins, steroids, saponins and Tannins, (Aswar et al., 2008). Leaves contain crude protein, crude fibers, calcium and phosphorus. It yields latex containing caoythowe (Mukherjee et al., 1998). The bark of the *Ficus benghalensis* contains leucopelargonidin-3-O-x-L rhamnoside and leuco cynidin. 3-O-x-D galactosyl cellobioside, glucoside beta glucoside, 20-tetrahydro-2H-pyran-2-one, 6-heptatriacontene-10-one, pentatricentan-5-one, beta sitosterol- α -D-glucose and mesoinositol (Subramaniam and Misra 1978). The preliminary phytochemical analysis of the methanol extract of *Ficus religiosa* bark studied by Uma et al., showed the presence of flavonoids, saponins and tannins. (2009). *F. racemosa* bark contain phytochemical constituents namely alkaloids, carbohydrates, flavonoids, glycosides, saponins, steroids, tannins, phenols and triterpenoids (Pongothai et al., 2011).

Ethnobotany of plants studied

Ficus plants are found throughout the world as moderate woody plants or trees. It has a vast traditional role in indigenous system of medicine like ayurveds, siddha, unani and homeopathy. *Ficus* species, namely, *F. benghalensis* and *F. racemosa*, *F. religiosa* are important ingredients in many ayurvedic and traditional formulations. The barks, leaves fruits and latex are considered to be very effective in various treatments, such as diabetes, skin diseases, ulcers, dysentery, diarrhea, stomach ache, piles and as carminative, astringent, anti-inflammatory antioxidant and anticancer agents.

Ficus benghalensis used in Ayurveda for treatment of diarrhea, piles, teeth and skin disorders. The plant parts are used in diseases of blood, vaginal uterus, and leucorrhea, burning sensation, gonorrhoea, diarrhea, dysentery, hemorrhoids, gastrohecosis (Warrier et al., 1995). The bark is used in inflammation, swelling at neck, gonorrhoea, scabies mouthwash for tooth ache, and

for strengthening gums, and steeped freshly burnt bark has been said to cure cases of obstinate hiccup. The latex (Rathish Nair and Sumitra, 2007) is used in inflammations and hemorrhages. Malpamaram is an important group of ayurvedic formulation that constitutes the barks of *Ficus recemosa*, *Ficus religiosa* and *Ficus benghalensis* widely used in the treatment of skin diseases and also used in various ailments (Sivarajan and Balachandran, 1994; Joy et al., 2001).

Pharmacological studies of the plants

The bark of *Ficus benghalensis* exhibited significant antibacterial activity against pathogenic bacterial like *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Klebsiella premoniae* (Gayathri and Kannabiran, 2009). *Ficus religiosa* aqueous extract showed high antimicrobial activity. High activity was found against *Bacillus subtilis* and *Pseudomonas aeruginosa*, (multi-drug resistant) by Preeti et al.(2010) Iqbal et al (2001) studied invitro antihelmintic activity of *Ficus religiosa*. *Ficus religiosa* bark methanolic extract was 100% lethal for *Haemonchus contortus* worms. Wound healing activity of the hydroalcoholic extract of *Ficus religiosa* leaves in rats studied by Nayeem et al. The leaf extract of *Ficus religiosa* (both 5% and 10%) applied topically possess dose dependent wound healing activity.

Ficus recemosa are medicinally important in traditional system of medicine in India, and have been used extensively in biliary disorders, jaundice dysentery, diabetes and diarrhea and in inflammatory conditions (Bhaskara rao, et al., 2002). Anthocyanins (ACN) are part of a large and widespread group of plant constituents known collectively as flavonoids. *F. recemosa*__fruit anthocyanides preparation demonstrated significant vaso-protective effect in rabbits (Sarpate et al., 2009). Ethnomedicinal study and anti bacterial activities of *F. recemosa* was studied by Mahato et al.,(2005) *F. recemosa*, country big tree paste of bark is applied twice a day for 2-3 days to cure swellings of foot and hands. Bark decoction is gargled to cure mouth ulcer paste of stem bark of *F. recemosa* was used to cure swelling of foot and hand showed activity against *Bacillus subtilis*.

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MATERIALS AND METHODS

Plant material collection:

The barks of *Ficus benghalensis*, *Ficus religiosa*, *Ficus recemosa* were collected from herbal garden of Gloris biomed Research center, Vadapalani. The plants authenticated identification done by Dr. S. Sankaranarayanan, Head, Department of Medicinal Botany, Sairam Siddha Medical College, Tambaram. The voucher specimens were submitted to Presidency College, Department of Botany. The voucher numbers are P.5123, P.5134, P.5137.

Preparation of Extracts:

Aqueous and methanolic extracts of bark of *Ficus benghalensis*, *Ficus religiosa*, *Ficus recemosa* were prepared in 20g/200ml. The solvent of organic extract was dried at 60°C protected from light. The dried bark powder stored at 40°C until use.

Phytochemical analysis of the plant extract

The aqueous extract is freshly prepared and divided into different test tubes and various chemical constituents were analysed according to methods described by Allen (1974) and Harbone (1976). The different chemical constituents tested for included tannins, saponin, glycosides, glucosides, alkaloids, triterpenes and flavonoids

Test for tannins: About 0.5 g of the dried powdered samples was boiled in 20 ml of water in a test tube and then filtered. A few drops of 0.1% ferric chloride was added and observed for brownish green or a blue-black colouration.

Test for saponin: About 2 g of the powdered sample was boiled in 20 ml of distilled water in a water bath and filtered. 10ml of the filtrate was mixed with 5 ml of distilled water and shaken vigorously for a stable persistent froth. The frothing was mixed with 3 drops of olive oil and shaken vigorously, then observed for the formation of emulsion.

Test for flavonoids: Three methods were used to determine the presence of flavonoids in the plant sample (Harbone, 1973). 5 ml of dilute ammonia solution were added to a portion of the aqueous filtrate of each plant extract followed by addition of concentrated H₂SO₄. A yellow colouration

observed in each extract indicated the presence of flavonoids. The yellow colouration disappeared on standing. Few drops of 1% aluminium solution were added to a portion of each filtrate. A yellow colouration was observed indicating the presence of flavonoids. A portion of the powdered plant sample was in each case heated with 10 ml of ethyl acetate over a steam bath for 3 min. The mixture was filtered and 4 ml of the filtrate was shaken with 1 ml of dilute ammonia solution. A yellow colouration was observed indicating a positive test for flavonoids

Test for terpenoids (Salkowski test): Five ml of each extract was mixed in 2 ml of chloroform, and concentrated H_2SO_4 (3 ml) was carefully added to form a layer. A reddish brown colouration of the inter face was formed to show positive results for the presence of terpenoids.

Test for glycosides (Keller-Killani test): Five ml of each extracts was treated with 2 ml of glacial acetic acid containing one drop of ferric chloride solution. This was underlaid with 1 ml of concentrated sulphuric acid. A brown ring of the interface indicates a deoxysugar characteristic of cardenolides. Green-blue colour indicated the presence of cardiac glycosides.

Test for Alkaloids: Dragendorffs reagent: 8g of bismuth nitrates $Bi(NO_3)_3 \cdot 5 H_2O$ was dissolve in 20ml of HNO_3 and 2.72g of Potassium iodide in 50ml of H_2O . These were mixed and allowed to stand for deposition of KNO_3 Crystals. The Supernatant was decanted off and made up to 100ml with distilled water. **Procedure:** To 0.5ml of bark extract 2ml of HCl was added. To this acidic medium 1ml of dragendorffs reagent was added on, orange or red precipitate produced immediately indicate the presence of alkaloids.

Mayers test: 1.36g of Mercuric chloride was dissolved in 60ml of distilled water and 5g of Potassium iodide in 10ml of water. These two solutions were mixed and diluted to 100 ml with distilled water. **Procedure:** 1.2 ml of plant extract was taken in a test tube and to this 0.2 ml of dilute HCl and 0.1 ml of Mayers reagent were added. Formation of yellowish Puff coloured precipitate indicates the presence of alkaloid.

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Test for Anthraquinones: Five ml of the extract was hydrolysed with diluted conc. H_2SO_4 extracted with benzene. 1 ml of dilute ammonia was added to it. Pink coloration suggested the positive response for anthraquinones.

RESULTS AND DISCUSSION

Herbal extracts contain different phytochemicals with biological activity that can be of valuable therapeutics Index. Plants have the ability to produce a large variety of secondary metabolites (Phytochemicals) such as terpenoids, phenylpropanoids, flavonoids and alkaloids which together account for over 2,00,000 compounds. (Dixon, 1998). In the present study, three Ficus species namely, Ficus Benghalensis Ficus religiosa and Ficus recemosa were analyzed for their phytochemical constituents. During the last three decades, the development of drug resistance and their undesirable side effects of certain antibiotics have led to the search of new antimicrobial agents mainly among plant extracts with the goal to discover new chemical structures.

In recent years, secondary plant metabolites (phytochemicals) previously with unknown pharmacological activities, have been extensively investigated as a source of medicinal agents (Baladrin et al., 1985). Phytochemicals with adequate, antibacterial effect will be used for the treatment of bacterial infections (Tanaka et al., 2002). It is time to examine more closely our natural resources, i.e., the plants, which contain compounds of potential medical use.

Phytochemistry:

Phytochemical screening of aqueous and methanolic extract of Ficus benghalensis showed the presence of Tannins, saponins, flavonoids (Table 1). Aqueous and methanolic extracts of Ficus religiosa found to contain Tannins, saponins, flavonoids and terpenoids (Table 2). Aqueous and methanolic extract of Ficus recemosa showed positive results for the tests of Tannins, saponins, flavonoids and glycoside (Table 3).

Table 1. Phytochemical screening of bark extract of *Ficus benghalensis*

S/No	Phytochemical Constituents		Observation	Methanol extract of <i>Ficus benghalensis</i>
1.	Alkaloids	Dragendorff's test	Orange / red precipitate	-
		Mayers test	Yellowish precipitation	
2.	Fla.vonoids		Intense yellow colour	+
	Alkali Reagent			
3.	Glycosides		Green-blue colour	-
	KellerKialni			
4.	Tannin		Bluish black colouration	+
	FeCl ₃ test			
5.	Saponins		Foam	+
	Frothing test			
6.	Terpenoids		Reddish brown at the interface	-
	Salkowski test			
7.	Anthraquinones Benzene Ammonia Test		Pink colour	-

Key : - → Negative (absent) + → Positive (present)

Table 2. Phytochemical screening of bark extract of *Ficus religiosa*

S/No.	Phytochemical Constituents		Observation	Methanol extract of <i>Ficus religiosa</i>
1.	Alkaloids	Dragendorff's test	Orange / red precipitate	-
		Mayers test	Yellowish precipitation	
2.	Flavonoids		Intense yellow colour	+
	Alkali Reagent			
3.	Glycosides		Green-blue colour	-
	KellerKilani test			
4.	Tannin		Violet colour	+
	FeCl ₃ test			
5.	Saponins		Foam	+
	Frothing test			
6.	Terpenoids		Reddish brown at the interface	+
	Salkowski test			
7.	Anthraquinones Benzene Ammonia Test		Pink colour	-

Key : - → Negative (absent) + → Positive (present)

Table 3. Phytochemical screening of bark extract of *Ficus recemosa*

S/No.	Phytochemical Constituents		Observation	Methanol extract of <i>Ficus recemosa</i>
1.	Alkaloids	Dragendorff's test	Orange / red precipitate	-
		Mayers test	Yellowish precipitation	
2.	Flavonoids		Intense yellow colour	+
	Alkali Reagent			
3.	Glycosides		Green –blue colour	+
	KellerKialni test			
4.	Tannin		Violet colour	+
	FeCl ₃ test			
5.	Saponins		Foam	+
	Frothing test			
6.	Terpenoids		Reddish brown at the interface	-
	Salkowski test			
7.	Anthraquinones Benzene Ammonia Test		Pink colour	-

Key : - → Negative (absent) + → Positive (present)

The previous studies on the phytochemical screening of *Ficus benghalensis* revealed the presence of saponins, tannins and flavonoids in aqueous and methanolic extract. (Ashwar et al., 2008). The preliminary phytochemical analysis of the methanol extract of *Ficus religiosa_bark* studied by Uma et al., showed the presence of flavonoids, saponins and tannins. (2009). The phytochemical screening of *Ficus recemosa* bark (various extracts) studied by Poongothai et al., showed the presence of alkaloids, flavonoids, glycosides, saponins, tannins and triterpenoids and the absence of anthraquinones (2011). Such screening of various natural organic compounds and identification of active agents is the need of the hour because successful prediction of lead molecule at the onset of drug discovery will payoff later in drug discovery.

CONCLUSION

From these findings, it is evident that aqueous and methanolic extracts of *Ficus* species contain

phytochemicals of therapeutic value. Since the study was conducted in a controlled manner, the

phytochemicals can be used for the formulation of compound drugs. Further investigations should be conducted to isolate and characterize the active components of these *Ficus* species.

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