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EFFECT OF DIFFERENT EXTRACTS OF STEM BARK OF *FICUS* SP. ON MULTIDRUG RESISTANT PATHOGENIC BACTERIA

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ABSTRACT

Keywords:

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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.

Purpose: *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 study the antibacterial effect of *Ficus* sp., by appropriate methods.

Method: The acetone, methanol, ethylacetate extracts of *Ficus benghalensis*, *Ficus religiosa*, *Ficus recemosa* were evaluated for antibacterial activity against medically important bacteria *Pseudomonas aeruginosa*, *Escherichia coli*, *Proteus vulgaris*, *Bacillus subtilis*, *Staphylococcus aureus*. Phytoconstituents of three plants were screened. The *in-vitro* antibacterial activity was performed by agar disc diffusion assay.

Results: The plant extract that showed higher active antibacterial activity against tested bacterial strain was chosen analyse beta-lactamase inhibitory activity. *Ficus* sp. Showed more antibacterial activity on *Escherichia coli*, *Staphylococcus aureus* and *Proteus vulgaris*.

Conclusion: Amongst the plant species screened, *Ficus benghalensis* extracts showed best antibacterial activity. These findings suggest a new pathway in elucidating a potent antimicrobial agent from *Ficus benghalensis*.

INTRODUCTION: 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 value¹. 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 first step towards, this goal is the *invitro* antibacterial assay².

Infectious diseases are the leading cause of death world-wide. Antibiotic resistance has become a global concern³. The clinical efficacy of many existing antibiotics is being threatened by the emergence of multi-drug resistant pathogens⁴. *Staphylococcus aureus* is one of the major pathogens found on the mucous membranes and the skin of around a third of

the population. It's extremely adaptable to antibiotic pressure. It was the first bacterium in which penicillin resistance was found. MRSA was responsible for 37% of fatal cases of blood poisoning in the UK in 1999; up from 4% in 1991, half of all *S. aureus* infections in the U.S are resistant to penicillin, methicillin, tetracycline and erythromycin. Considering the vast potentiality of plants as sources for antimicrobial drugs with reference to antibacterial agents, a systematic investigation was undertaken to screen the antibacterial activity of three different species from *Ficus* genus.

The genus *Ficus* includes some 750 species of woody plants occurring in most tropical and subtropical forests throughout the world ⁵. The genus is remarkable for the large variation in the habits of its species ⁶.

***Ficus benghalensis*:** *Ficus benghalensis* belongs to the family Moraceae, which is commonly known as Banyan tree. The tree is a fast growing, evergreen tree found in monsoon and rain forests grow up to 30 meters, with spreading branches and many aerial roots. **Figure 1** shows the fully grown banyan tree with aerial roots.



FIGURE 1: FICUS BENGHALENSIS

Kingdom : Plantae
 Division : Magnoliophyta
 Class : Magnoliopsida
 Order : Urticales
 Family : Moraceae
 Genus : Ficus
 Species : *F. benghalensis*
 Binomial name: *Ficus benghalensis*

***Ficus religiosa*:** *Ficus religiosa* Linn (Moraceae) commonly known as 'Peepal tree' is a large widely branched tree with leathery, heart-shaped, leaves on long slender petioles and purple fruits growing in pairs. The tree is regarded as a sacred tree to both Hindus and Buddhists. It has got mythological, religious and medicinal importance ⁷. **Figure 2** shows the branched peepal tree and **figure 3** represents the typical leaf structure of the same tree.

Kingdom : Plantae
 Division : Magnoliophyta
 Class : Magnoliopsida
 Order : Rosales
 Family : Moraceae
 Genus : Ficus
 Species : *F. religiosa*
 Binomial name: *Ficus religiosa*



FIGURE 2: FICUS RELIGIOSA



FIGURE 3: TYPICAL LEAF STRUCTURE FICUS RELIGIOSA

***Ficus racemosa*:** *Ficus racemosa* synonymously also known as *Ficus glomerata*. An evergreen tree 50-60 ft. high; young shoot glabrous, pubescent or scaberulous. Leaves 3-6 by 1.25-2.5 in. long, glabrous; stipules 0.75 in. long, ovate-lanceolate, scarious, pubescent. Receptacle 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, subglobes, pyriform or subturbinate, 1.5 in. across, smooth or pubescent, red when ripe, with depressed umbilicus (edible but usually full of worms); basal bracts 3, ovate triangular; male, female and gall flowers together in one receptacle, the male flower forming a zone near the mouth, the fertile female flowers forming a layer near the walls of the receptacle, and the gall flowers a more internal layer. Figure 4 shows the branched *Ficus recemosa* tree with figs.



FIGURE 4: *FICUS RACEMOSA*

Kingdom	:	Plantae
Division	:	Magnoliophyta
Class	:	Magnoliopsida
Order	:	Rosales
Family	:	Moraceae
Genus	:	<i>Ficus</i>
Species	:	<i>F. recemosa</i>
Binomial name:		<i>Ficus recemosa</i>
Synonyms	:	<i>F. glomerata</i> Roxb.

Ethnobotany of plants studied: *Ficus* plants are found throughout the world as moderate woody plants or trees. *Ficus* species, namely, *F. benghalensis* and *F. recemosa*, *F. religiosa* are important ingredients in many ayurvedic and traditional formulations. Roots of *Ficus benghalensis* shows antihelmintic activity. Fruit extracts exhibits anti-tumour activity⁸. The fruit extracts of *F. benghalensis* exhibit antitumor activity and antibacterial activity, but no antifungal activity⁹. In Ayurveda it is claimed that *Ficus religiosa* possesses anticonvulsant activity. Different parts of *Ficus religiosa* showed cholinesterase inhibitory activity and antianxiety activity. The methanolic extract of figs of *Ficus religiosa* had anticonvulsant activity¹⁰.

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^{11,12}.

Pharmacological studies of the plants: The bark of *Ficus benghalensis* exhibited significant antibacterial activity against pathogenic bacterial like *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Klebsiella pneumoniae*¹³. *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*¹⁴.

Ethnomedicinal study and anti bacterial activities of *F. recemosa* was studied by Mahato *et al.*,¹⁵ *F. recemosa*, country fig tree paste of bark is applied twice a day for 2-3 days to cure swellings of foot and hands. Stem bark of *F. recemosa* showed activity against *Bacillus subtilis*.

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.

Test organisms: The screening for antibacterial activity was carried out in vitro condition using *Pseudomonas aeruginosa* (MTCC 27853), *Escherichia coli* (MTCC 25922), *Proteus vulgaris* (MTCC 13315), *Bacillus subtilis* (MTCC12567), and *Staphylococcus aureus* (MTCC 29213). All bacterial strains were obtained from Microbial Culture Collection Centre, Chandigarh, India.

Preparation of Extracts: Aqueous and methanolic extracts of bark of *Ficus benghalensis* were prepared in 20g/200ml. The solvent of organic extract was dried at 60°C protected from light. The dried bark powder stored at 4°C until use.

Phytochemical analysis of the Plant Extract: The extracts were subjected to phytochemical tests for plant secondary metabolites, tannins, saponins, flavonoids, alkaloids and glycosides in accordance with Allan¹⁶ and Harborne¹⁷ with little modification (**table 1**).

1. **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.
2. **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.
3. **Test for Flavonoids:** Three methods were used to determine the presence of flavonoids in the plant sample^{18, 19}. 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.
4. **Test for Terpenoids (Salkowski test):** Five ml of each extract was mixed in 2 ml of chloroform, and concentrated H₂SO₄ (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.
5. **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. A violet ring may appear below the brown ring, while in the acetic acid layer, a greenish ring may form just gradually throughout thin layer.

6. Test for Alkaloids:

- (A) **Dragendorffs reagent:** 8g of bismuth nitrates Bi (NO₃)₃.5H₂O was dissolve in 20ml of HNO₃ and 2.72g of Potassium iodide in 50ml of H₂O. These were mixed and allowed to stand for deposition of KNO₃ Crystals. The Supernatant was decanted off and made up to 100ml with distilled water.

Procedure: To 0.5ml of leaf 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.

- (B) **Mayer's 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 Mayer's reagent was added. Formation of yellowish Puff coloured precipitate indicates the presence of alkaloid.

Staphylococcal Beta-lactamase Detection Methods: In order to perform iodometric slide method, one million units penicillin was dissolved in 1 ml sterile distilled water. The solution was divided into portions of 0.15 ml and stored at -20°C until use. On test day, iodine solution was added to penicillin solution and mixed. The mixture was dropped on the slide and bacteria were transferred to the solution. Solution and bacteria were mixed by loop and 4% sterile starch solution was dropped. If the purple color of the solution disappeared, bacteria were considered to be beta-lactamase positive²⁰. In iodometric tube method, benzyl penicillin was dissolved in phosphate buffer, which was adjusted to pH 6. 0.1 ml of the solution was taken to microtitration plate. The solution was made

cloudy with 3-4 colonies of bacteria. After 30-60 min, 20 μ l of sterile 1% starch solution and iodine solution were added.

If the color of iodine disappeared in 5 min, the isolate was considered beta-lactamase positive²¹.

TABLE 1: PRELIMINARY PHYTOCHEMICAL ANALYSIS OF MEDICINAL PLANTS

Tests Medicinal Plant Species	→						Alkaloids	
	Tannins	Saponins	Flavonoids	Terpenoids	Glycosides	Anthraquinones	Meyer's test	Dragendorff's Test
<i>Ficus Benghalensis</i>	+	+	+	-	-	-	-	-
<i>Ficus religiosa</i>	+	+	+	+	-	-	-	-
<i>Ficus recemosa</i>	+	+	+	-	+	-	-	-

Antibacterial activity of different Extracts: The antibacterial activity was studied using the disc-diffusion Method²². Bacteria were grown overnight on Mueller Hinton agar plates, five young colonies were suspended with 5ml of sterile saline (0.9%). The swab was used to inoculate the dried surface of MH agar plate by swabbing several times four times over the surface of the agar. The medium was allowed to dry for about 3 min before adding a sterile paper disc of 9mm diameter. Compounds (50 μ g) were weighed and dissolved in 1ml of 7% acetone. Twenty microlitres of the compounds were introduced on each disc (five replicates) and 7% acetone alone served as a normal control. The plates were incubated at 37 °C for 24 h; inhibition zones were measured and calculated.

RESULTS: 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 compounds²³.

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. Preliminary screening of antibacterial activity was conducted for the three *Ficus* sp; against both gram positive and gram negative type of bacterial and two pathogenic fungal species. Among, those *Ficus benghalensis* showed the highest inhibition of both Gram positive and Gram negative bacteria and fungi.

The antimicrobial activity was determined by measuring the diameter of the zone of inhibition, i.e. the mean of triplicates \pm S.D. In recent years, secondary plant metabolites (phytochemicals) previously with unknown pharmacological activities have been extensively investigated as a source of medicinal agents²⁴. Phytochemicals with adequate, antibacterial effect will be used for the treatment of bacterial infections²⁵. 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. Aqueous and methanolic extracts of *Ficus religiosa* showed the presence of Tannins, saponins, flavonoids and terpenoids. Aqueous and methanolic extract of *Ficus recemosa* showed the presence of Tannins, saponins, flavonoids and glycosides.

Antibacterial activity: The dried bark powder of three *Ficus* sp., namely *Ficus benghalensis*, *Ficus religiosa* and *Ficus recemosa* were extracted with three organic solvents acetone, methanol and ethyl acetate. The antibacterial activity of these extracts was performed by disc diffusion assay. The results for antibacterial activity of *Ficus benghalensis*, *Ficus religiosa* and *Ficus recemosa* by the agar disc diffusion assay are given in tables 2, 3, 4 respectively.

***Ficus benghalensis*:** Acetone extract of *F. benghalensis* showed good antibacterial activity against the entire tested gram positive and gram negative bacteria. The same extract was found to be very active against *P. vulgaris*, measuring 14.3 mm of zone of inhibition. Methanol extract of the plant found to be potentially good antibacterial activity against all the bacteria except *P. aeruginosa*. Ethyl acetate extract of *F.*

benghalensis was active against *E. coli*, *S. aureus* and *B. subtilis*.

***Ficus religiosa*:** The growth of *Bacillus subtilis* was significantly inhibited by acetone extract of *Ficus religiosa*. Higher concentrations of the same extract were required for the inhibition of *E. coli*. Methanol extract of the plant was very active against all the tested bacterial pathogens except *P. aeruginosa*. Ethyl acetate extract was not active against all the bacterial species.

***Ficus recemosa*:** Acetone extract of *Ficus recemosa* was found to be active against *E. coli*, *Proteus vulgaris* and *Bacillus subtilis*. It showed maximum activity against *Proteus vulgaris* measuring 14.0mm of zone inhibition at 100µg/ml. Methanol extract of plant was found to be active against *E. coli*, *P. vulgaris* and *B. subtilis* and *P. aeruginosa*. The ethyl acetate extract showed no anti bacterial activity against all the tested bacteria. The mean zone of inhibition on obtained using the agar disc diffusion assay ranged from 6mm (for *Pseudomonas aeruginosa*) to 14mm (*Proteus vulgaris*, *Bacillus subtilis*, *E. coli*).

TABLE 2: ANTIBACTERIAL ACTIVITY OF DIFFERENT BARK EXTRACTS TESTED AGAINST PATHOGENIC BACTERIA BY DISC DIFFUSION ASSAY (*FICUS BENGHALENSIS*)

Different extracts of <i>F. benghalensis</i>	<i>Escherichia coli</i>				<i>Proteus vulgaris</i>			
	25ug/ml	50ug/ml	75ug/ml	100ug/ml	25ug/ml	50ug/ml	75ug/ml	100ug/ml
Concentration Per disc	25ug/ml	50ug/ml	75ug/ml	100ug/ml	25ug/ml	50ug/ml	75ug/ml	100ug/ml
Acetone extract	7.1±1.5	10.4±1.09	12.2±0.98	13.2±0.15	9.2±0.95	11.36±0.81	11.8±1.3	14.26±0.91
Methanol Extract	7.5±1.35	10.3±1.05	11.9±1.42	12.5±1.23	8.4±0.65	11.3±0.95	12.4±1.4	13.1±1.35
Ethyl acetate Extract	8.53±1.02	10.0±1.47	11.4±0.92	12.3±1.38	-			
Different of extracts of <i>F. benghalensis</i>	<i>Staphylococcus aureus</i>				<i>Bacillus subtilis</i>			
	25ug/ml	50ug/ml	75ug/ml	100ug/ml	25ug/ml	50ug/ml	75ug/ml	100ug/ml
Concentration Per disc	25ug/ml	50ug/ml	75ug/ml	100ug/ml	25ug/ml	50ug/ml	75ug/ml	100ug/ml
Acetone extract	8.16±1.25	9.53±0.55	11.2±1.06	13.3±1.30	7.3±0.90	10.3±0.95	11.66±1.56	13.3±1.21
Methanol Extract	9.1±1.05	11.13±1.05	12.2±1.05	13.5±0.60	7.4±1.02	10.5±1.1	12.4±0.80	13.5±0.66
Ethyl acetate Extract	7.2±0.98	9.1±0.9	11.5±0.8	12.8±0.3	8.46±0.76	11.3±1.12	12.86±0.75	13.6±1.25
Different of extracts of <i>F. benghalensis</i>	<i>Pseudomonas aeruginosa</i>							
	Concentration Per disc	25ug/ml	50ug/ml	75ug/ml	100ug/ml			
Acetone extract		8.3±0.61	10.46±1.29	11.96±1.05	13.9±1.76			
Methanol Extract		-						
Ethylacetate Extract		-						

- No activity zone of inhibition in mm. The antimicrobial activity was determined by measuring the diameter of zone of inhibition that is the mean of triplicates ±SD.

TABLE 3: ANTIBACTERIAL ACTIVITY OF DIFFERENT EXTRACTS TESTED AGAINST PATHOGENIC BACTERIA BY DISC DIFFUSION ASSAY (*FICUS RELIGIOSA*)

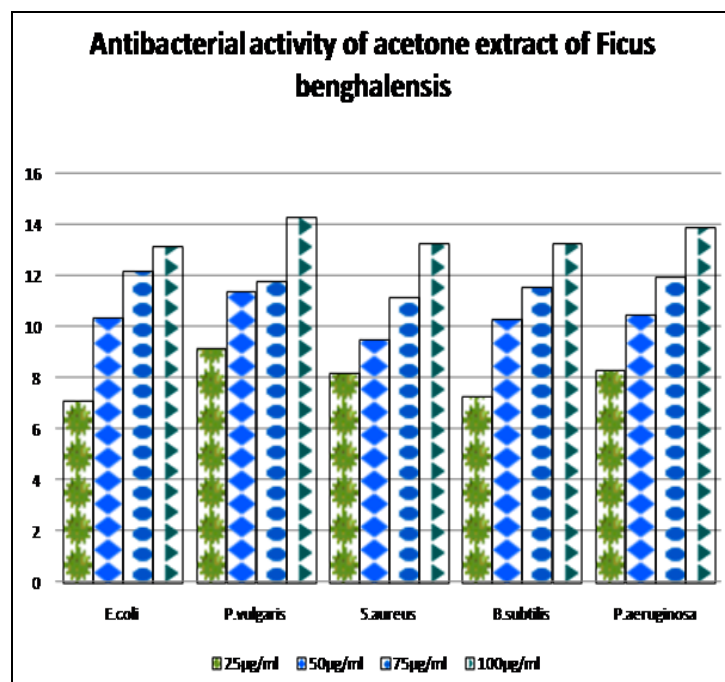
Different extracts of <i>F. Religiosa</i>	<i>Escherichia coli</i>				<i>Proteus vulgaris</i>			
	25ug/ml	50ug/ml	75ug/ml	100ug/ml	25ug/ml	50ug/ml	75ug/ml	100ug/ml
Concentration Per disc	25ug/ml	50ug/ml	75ug/ml	100ug/ml	25ug/ml	50ug/ml	75ug/ml	100ug/ml
Acetone Extract	-	-	8.03±1.62	10.43±0.92	-	-	-	9.36±0.94
Methanol Extract	8.63±1.1	11.4±0.83	13.4±1.40	14.1±1.01	-	8.5±0.83	10.4±0.79	
Ethyl acetate Extract	-				-			
Different of extracts of <i>F. religiosa</i>	<i>Staphylococcus aureus</i>				<i>Bacillus subtilis</i>			
	25ug/ml	50ug/ml	75ug/ml	100ug/ml	25ug/ml	50ug/ml	75ug/ml	100ug/ml
Concentration Per disc	25ug/ml	50ug/ml	75ug/ml	100ug/ml	25ug/ml	50ug/ml	75ug/ml	100ug/ml
Acetone Extract	-				7.9±1.32	10.6±0.56	11.4±1.23	13.6±0.91
Methanol Extract	12.8±1.05	13.13±1.15	9.16±1	12.6±1.15	13.9±0.37	14.6±0.92		
Ethyl acetate Extract	-				-			
Different of extracts of <i>F. religiosa</i>	<i>Pseudomonas aeruginosa</i>							
	Concentration Per disc	25ug/ml	50ug/ml	75ug/ml	100ug/ml			
Acetone extract		-						
Methanol Extract		-						
Ethyl acetate Extract		-						

- No activity zone of inhibition in mm. The antimicrobial activity was determined by measuring the diameter of zone of inhibition that is the mean of triplicates ±SD.

TABLE 4: ANTIBACTERIAL ACTIVITY OF DIFFERENT EXTRACTS OF *FICUS RECEMOSA* AGAINST BACTERIAL SPECIES TESTED BY DISC DIFFUSION ASSAY

Different extracts of <i>F. Recemosa</i>	<i>Escherichia Coli</i>				<i>Proteus Vulgaris</i>			
Concentration Per disc	25ug/ml	50ug/ml	75ug/ml	100ug/ml	25ug/ml	50ug/ml	75ug/ml	100ug/ml
Acetone extract	7.3±1.13	9.53±0.49	12.7±1.3	13.5±1.15	9.6±1.35	11.43±1.35	13.2±0.36	14.0±0.9
Methanol Extract	9.16±0.47	10.8±0.7	12.03±1	13.76±0.85	9.3±1.07	12.23±0.92	13.7±0.80	14.8±1.19
Ethyl acetate Extract	-				-			
Different of extracts of <i>F. Recemosa</i>	<i>Staphylococcus aureus</i>				<i>Bacillus subtilis</i>			
Concentration Per disc	25ug/ml	50ug/ml	75ug/ml	100ug/ml	25ug/ml	50ug/ml	75ug/ml	100ug/ml
Acetone extract	-				8±1.65	10.4±0.8	13.2±0.75	14.23±1.25
Methanol Extract	-				8.2±1.23	10.6±1.07	12.36±1.02	13.8±0.85
Ethylacetate Extract	-				-			
Different of extracts of <i>F. recemosa</i>	<i>Pseudomonas aeruginosa</i>							
Concentration Per disc	25ug/ml	50ug/ml	75ug/ml	100ug/ml				
Acetone extract	-							
Methanol Extract	6.8±0.70	8.43±0.61	10.7±0.97	12.73±0.40				
Ethyl acetate Extract <i>F. recemosa</i>	-							

- No activity zone of inhibition in mm. The antimicrobial activity was determined by measuring the diameter of zone of inhibition that is the mean of triplicates ±SD.

**FIGURE 5: ANTIBACTERIAL ACTIVITY OF ACETONE EXTRACT OF *FICUS BENGHALENSIS***

DISCUSSION: The previous studies on the phytochemical screening of *Ficus benghalensis* revealed the presence of saponins, tannins and flavonoids in aqueous and methanolic extract²⁶. The preliminary phytochemical analysis of the methanol extract of *Ficus religiosa* bark studied by Uma *et al.*, showed the presence of flavonoids, saponins and tannins²⁷.

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 anthraquinones²⁸.

The present study indicates strong antibacterial activity of bark extract of *Ficus benghalensis*. With the zone of inhibition more than 10mm against four of the five bacterial strains tested, the acetone extract of *Ficus benghalensis* clearly possesses a strong and broad spectrum of antibacterial activity. The antibacterial activity against both gram positive and gram negative bacteria was in the order of Acetone>Methanol>ethyl acetate extract of *F. benghalensis*.

The methanol extract of *Ficus religiosa* and *Ficus recemosa* showed moderately good antibacterial activity against all tested bacterial strains. The ethyl acetate extract of these plants were less potent against most of the pathogens tested.

CONCLUSION: These findings suggest a new pathway in elucidating a potent antimicrobial agent from *Ficus benghalensis*. The present study indicates that the plant contains antimicrobial compound which can be further developed as phytomedicine for the therapy of infection.

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.

Lastly to conclude, the extracts were found to inhibit the growth of Gram positive bacteria as well as the Gram negative bacteria and also and the methanolic extract of the three plants was comparably more effective to inhibit the growth of microbes than acetone and ethyl acetate extracts.

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