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EVALUATING THE EFFICIENCY OF *FICUS CARICA* FRUITS AGAINST A FEW DRUG RESISTANT BACTERIAL PATHOGENS

***Jasmine R, Manikandan K, Brinda, Niveditha, Kalaivani, Thirupathi, Manikandan G**

P.G.& Research Department of Biotechnology, Bishop Heber College, Tiruchirappalli-17, Tamil Nadu, India.

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***Correspondence for Author:**
Dr. R.Jasmine,
Assistant Professor, Dept of Biotechnology, Bishop Heber College, Tiruchirappalli 620 017, South India.

ABSTRACT

The emergence of antibiotic resistance of clinically important pathogens has led the scientists to screen for plants as alternate therapeutic sources. In this study, we have evaluated two different extracts of *Ficus carica* against drug resistant human pathogens. The ethanol extracts was found to be more effective than methanol extract. The MIC values fell in the range of 0.94 to 30 µg/ml and hence it can be concluded that *Ficus carica* can be a potential source of therapeutic drugs. Further investigations to discover natural compounds that may serve as lead compounds for drug development are underprogress.

Keywords: *Ficus carica*, Antibacterial, MIC, Drug resistant.

INTRODUCTION

Ficus carica is commonly referred as Fig. Various parts of the plant like bark,leaves, tender shoots, fruits, seeds, and latex are medicinally important. The fig is a very nourishing food and used in industrial products. It is rich in vitamins, mineral elements, water, and fats. Figs are one of the highest plant sources of calcium and fiber They are good source of flavonoids and polyphenols. and some bioactive compounds such as arabinose, β-amyrins, β- carotines, glycosides, β-setosterols and xanthotoxol [1-3]. The dried figs produced a significant increase in plasma antioxidant capacity and also used in various disorders such as gastrointestinal respiratory,inflammatory ,cardiovascular disorders, ulcerative diseases, and cancers [4-7]. In traditional medicine the roots are used in treatment of leucoderma and ringworms and its fruits which are sweet, have antipyretic, purgative, aphrodisiac properties and have shown to

be useful in inflammations and paralysis[8-9].*F. carica* has been reported to include antioxidant,antiviral, antibacterial, hypoglycemic, cancer suppressive, hypotriglyceridaemic, and anthelmintic effects [10-12]. Recent bacterial infections have become difficult to tract as several bacteria have drug resistant emergence such drug resistant bacteria within the community settings need to be controlled. Hence a search was made for alternate therapeutic sources including plants. Traditionally used medicinal plants have known to possess antibacterial activities. The selection of medicinal plants is based on their traditional uses. Very few reports of plants against MDR bacteria are available from India. Aqil *et al* [13] have evaluated the ethanolic extract against few medicinal plants. Jasmine *et al* [14] have reported few plants to be active against ESBLs. Though many plants are reported in the indigenous systems of medicine for the biological activities, yet, several are unknown in the medical community, since they remain to be scientifically established along with the active compounds. The present study was aimed at comparing the methanol and ethanol extracts of the *Ficus carica* against a few bacterial pathogens.

MATERIALS AND METHODS

Collection of plant materials

Ficus carica fruits were purchased from local supermarket at Tiruchirappalli District, Tamil Nadu, India during the month of March and authenticated by the botany department of our college and the voucher specimen was deposited. The fruits chosen for the study had been washed, macerated and lyophilised. About 500g of fruits yielded 37g powder. The procedure was repeated to collect the needed quantity.

Preparation of plant extract

The fresh fruits of *Ficus carica* were collected from different places. The collected fruits were washed with clean water immediately after collection. The collected fruits were chopped into small pieces, sun dried for about 5days and grinder into coarse powder with a mechanical grinder and stored in an air tight container. Then 25grams of powder was taken mixed with 100ml of ethanol and methanol respectively for 5 days at room temperature 25 ± 2° C with occasional stirring. After 5days the ethanol and methanol extract was filtered with Whatman.No. 1 filter paper. The extract was concentrated under reduced pressure below 50°C through rotary vaccum evaporator. The concentrated extract was collected in a petridish and allowed to a dry for complete evaporation of ethanol and methanol. The whole process was

repeated 3times and finally, collected the blackish green color, concentrated plant extract. It was stored at 4℃ for future use.

Bacterial strains used in the study

The test organisms used in this study include ***E.coli, Pseudomonas aeruginosa, Streptococcus sp., Enterobacter sp., Klebsiella pneumonia, S.typhi, S.paratyphi.*** All bacterial species were maintained in nutrient agar medium.

Antibacterial Assay

The crude extracts [ethanol, methanol] obtained from the fruits of *Ficus carica* was studied for its antibacterial activity against the test organisms using disc diffusion method. Each strain was inoculated in a conical flask containing 100ml of nutrient broth. The conical flasks were incubated at 37^0C for 24hours and were referred to as seeded broth. Nutrient agar was prepared and about 20ml of molten media was poured into the sterile petriplates. After solidification, the media in each plate were inoculated with the test organisms from the seeded broth using cotton swabs.

Disc Diffusion test [15]

All isolates were tested for susceptibility to the extracts and antimicrobial agents on Mueller Hinton agar (Hi-Media India) by the standard disc diffusion method recommended by the National Committee for Clinical Laboratory Standards. The diameter of the zone of inhibition of growth was recorded, and interpreted by the criteria of CLSI[16]. Triplicates were maintained in each extract and average values were calculated for the eventual antibacterial activity. The diameters zone of inhibition in the triplicates was measured by calculating the difference between diameter of the disc and the diameter of the inhibition.

Micro-Well dilution essay

Minimum inhibitory concentrations (MIC) were performed by a serial dilution technique using 96 Well microliter plates[17]. Bacterial species were cultured from 12-hour broth cultures and suspensions were adjusted to 0.5 McFarland standard turbidity[18]. All tests were performed in Muller Hinton broth (MHB). The fractions were first diluted in ethanol 10% to the highest concentration 10mg/ml. Serial two-fold dilutions were made in the eight consecutive wells in a concentration ranging from 500µg/ml to 40µg. 45µl of MHB and 5µl of the inoculums were then added in each well containing 50µl of compound. The last well containing 95µl of MHB without compound and 5µl of the inoculums on each strip was used

as negative control. The final volume in each well was 100µl. Plates were incubated for 24 hours at 37°C then examined by a binocular microscope. The MIC was defined as the lowest concentration of the compounds to inhibit the growth of micro-organisms. The tested extract in this study was screened in three replicates against each organism. The minimum inhibitory concentration (MIC) was determined as the lowest concentration of test samples that resulted in a complete inhibition of visible growth in the broth.

RESULTS AND DISCUSSION

The alarming increase in microbial resistance to antibiotics has created a growing interest in herbal remedies in order to discover and develop new antimicrobial compounds[19]. There is also a call for a change in direction from the use of synthetic products, which can be more harmful than useful, to the use of more natural products[20]. Table 1 shows the antibacterial activity of fruit extracts of *Ficus carica* against the test organisms using disc diffusion method.

Table 1 : Inhibitory effect of the methanol and ethanol extracts of *Ficus carica* fruits against a few human pathogens by disc diffusion method (1000µg/10µl/disc)

Bacterial isolates	Zones of Inhibition (dia in mm)	
	Ethanol extract	Methanol extract
E.coli	3	6
Pseudomonas sp	15	8
Streptococcus sp.	7	5
Enterobacter sp.	8	7
Klebsiella pneumoniae	10	7
Salmonella typhi	11	10
Salmonella paratyphi A	8	6

Both the extracts of plant showed considerable antimicrobial activity at four different concentrations [10µl,20µl,30µl,40µl]. Among the test organisms used in the study, the ethanol extract of *Ficus carica* showed good antimicrobial effect against the organisms under study than the methanol extract. The zones of inhibition ranged from 3-11mm, where the maximum activity of 15mm was recorded against *Pseudomonas sp*. and 11mm against *Salmonella sp.*, suggesting the efficiency of the plant against drug resistant bacteria. The minimal inhibitory concentration varied from 0.94 to 30.0 µg/ml.

Table 2: Minimum Inhibitory Concentration of the methanol and ethanol extracts of *Ficus carica* fruits

Bacterial isolates	Minimum Inhibitory Concentration(µg/ml)	
	Methanol extract	Ethanol extract
E.coli	30	1.87
Pseudomonas sp	3.75	1.87
Streptococcus sp.	30	0.94
Enterobacter sp.	7.5	1.87
Klebsiella pneumoniae	7.5	7.5
Salmonella typhi	7.5	1.87
Salmonella paratyphi A	7.5	1.87

Comparing the two extracts, the ethanol extract demonstrated significant activity than its counterpart. The minimal inhibitory concentration of the ethanol extract was 0.94 µg/ml against *Pseudomonas sp.*, which was a significant result. The lower the MIC, the greater is the effect. From the obtained data, it can be suggested that *Ficus carica* is a potential candidate against bacterial infections. Further, isolation of the active principle may pave way for designing a drug with broad spectral activity. Our results have shown both the plants to have equal effect against both groups of bacteria, suggesting that our plants could have easily broken the effective permeability barrier[21]. It is possible that these bacteria, both gram positive and gram negative responded well to the plants as they had not been exposed to the plants before, and therefore had not had the opportunity to develop resistance yet as they have to antibiotics over the years.

CONCLUSION

This study has provided insight towards the possibility of using plants effectively as a possible alternative treatment for these bacterial lung infections. supporting the traditional use of these plants. Further fractionation and spectral analysis are under progress to identify an effective and safer drug for bacterial infections.

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