



ANTIBACTERIAL EFFECT OF FRUIT EXTRACTS OF *FICUS AURICULATA* LOUR

*¹S. Senthil Kumar, ²R. Rekka, ¹Treesa Santhosh and ¹Jils Thankachan

¹PG and Research Department of Botany, Vivekanandha College of Arts and Science for Women (Autonomous), Tiruchengode. 637205.

²Department of Botany, ERK Arts and Science college, Dharmapuri - 636905.

***Corresponding Author: S. Senthil Kumar**

PG and Research Department of Botany, Vivekanandha College of Arts and Science for Women (Autonomous), Tiruchengode. 637205.

Article Received on 07/12/2017

Article Revised on 28/12/2017

Article Accepted on 18/01/2018

ABSTRACT

Fruit of *Ficus auriculata* Lour. have been used to treat many diseases like diarrhea, dysentery, mumps, cholera, cuts and wounds. The present study was designed to evaluate the *in vitro* antibacterial activities of the methanolic fruit extracts of *Ficus auriculata* Lour. Bacterial strains were subjected to antibiotic sensitivity testing by Kirby–Bauer’s disc diffusion method. The antibacterial activity was calculated based on the zone of inhibition and activity index using Muller–Hinton broth in a spread plate method. The best antibacterial activity calculated as zone of inhibition and activity index. Methanolic fruit extracts of *Ficus auriculata* Lour. possessed highest activity against *Mycoplasma genitalium* and *Staphylococcus epidermidies*. Methanolic fruit extract of *Ficus auriculata* Lour. has the potential to be developed as antibacterial agents against some bacteria.

KEYWORDS: *Ficus auriculata* Lour., Antibacterial activity, disc diffusion method.

INTRODUCTION

Medicinally important plants have been identified as a part of the evolution of human healthcare for thousands of years. Medicinal components from plants play an important role in traditional as well as modern medicine. Bacterial infections are of particular concern mainly due to the development of antibiotic resistance. Plants are known to produce certain bioactive molecules which react with other organisms in the environment and inhibiting bacterial growth. The antibacterial property was claimed to be conferred by phytochemicals present in the plant. The use of the phytochemicals as natural antibacterial agent commonly called “biocides” is gaining popularity. There has been growing interest in the investigation of the natural products from plants for the discovery of new antibacterial agent. It has been reported that the higher plants have shown to be a potential source for the new antimicrobial agent^[1]

Ficus auriculata Lour. is a huge tropical and evergreen tree with more than 800 species. The whole parts of this plant are frequently used for medicinal properties in Ayurvedic and traditional chinese medicine to treat several common ailments. Phytochemical screening of *Ficus auriculata* Lour. revealed the presence of tannins, flavonoids, saponins, phenolic compounds etc. Tannins have been reported to prevent the development of microorganisms by precipitating microbial protein. Flavonoids displayed a remarkable array of biochemical and pharmacological viz. anti-inflammatory, antioxidant,

antiallergic, hepatoprotective, antithrombotic, antiviral and anticarcinogenic activities. Flavonoids are also shown to inhibit microbes which are resistant to antibiotics. Saponins are a special class of glycosides which have soapy characteristics. It has also been shown that saponins are active antifungal agents. Phenolic compounds like phenolic acids, phenols, flavonoids, phenyl propanoids, and phenolic quinones acted as antiseptic and anti-inflammatory.^[2] So the main aim of this research is to explore the antibacterial efficacy of methanolic fruit extracts of *Ficus auriculata* Lour.

MATERIALS AND METHODS

Preparation of Extraction

The dried fruit powder of *Ficus auriculata* Lour. was subjected to methanolic extraction adopting Soxhlet method. The extract was concentrated under reduced pressure to yield semisolid mass which was dried in a desiccators and stored properly for further study.

Antibacterial Assay^[3]

The antibacterial potential of methanolic fruit extract of *Ficus auriculata* Lour. was estimated by disc diffusion method. The disc diffusion is a simple and reliable test to find out the effect of a particular substance on a specific bacterium.

Source of Microbial Strains

The strains of common pathogenic microorganisms were used in this study such as *Proteus vulgaris*,

Staphylococcus epidermidies, *E. coli*, *Klebsiella pneumoniae*, *Neisseria gonorrhoeae*, *Mycoplasma genitalium* and *Pseudomonas aeruginosa*. All the bacterial cultures were obtained from Microbial Type Culture Collection (MTCC), Institute of Microbial Technology, Chandigarh, India. The young bacterial broth cultures were prepared before the screening procedure.

Preparation of Muller Agar Media:

38 g of Muller Hinton agar was dissolved in 1000 ml of glass water. The pH was adjusted to 7 and autoclaved for 30 minutes in 15 lb pressure.

Preparation of Culture Plates

20 ml of sterile Muller Hinton agar medium was poured into petriplates under sterile condition and kept in laminar air flow chamber for solidification. After solidification the plates were dried for 30 minutes in an oven to remove excess of moisture from the surface.

Preparation of Inoculums

Nutrient agar - 1gm
Bacteriological peptone - 0.5gm
Sodium chloride - 0.25gm
Distilled water - 100 ml

The above components were dissolved one by one in 100 ml of glass distilled water and the pH was adjusted to 7. 10 ml of medium was poured into test tube and the mouth of the tube was covered with sterile cotton. The test tubes were autoclaved for 30 minutes in 15 lb pressure. After autoclaving the test tubes were cooled in laminar air flow chamber and selected microorganisms were inoculated into the medium separately. The tubes were incubated overnight in 37°C and used for inoculation.

Inoculation

The test microorganisms were inoculated in nutrient agar medium by spread plate method. About 10 µl (106 cells/ml) of nutrient broth of overnight bacterial cultural was spread evenly on the solidification medium. Sterile cotton swabs were dipped separately into inoculums of organisms and swabbed inside the wall of the tubes. The agar surface of the plates was streaked in three directions by turning the plates to 60° angle between each streaking. The lid of the petriplates was on and kept at room temperature for 5-10 minutes to get confluent growth for accurate results.

Preparation and Application of Disc

Sterile discs (Hi Media) of 6 mm were used to load the plant extract. Various concentration of extract such as 30, 40, 50, 60 mg were dissolved in Dimethyl Sulfoxide (DMSO) and loaded in the discs. The standard antibiotic generation was used as a control due to its broad spectrum of activity against various organisms.

The impregnated discs were incubated at 37°C for an hour. The dried discs were placed over the surface of swabbed medium with equal distance to avoid the overlapping of the zones of inhibition. The discs were gently pressed on the surface of the medium and they were placed at least 25 mm away from the edge.

Incubation

The plates were incubated at 37°C for 16-18 hours in an incubator.

Measurement of Zone Inhibition

The diameter of the zone of inhibition was measured in mm at the end of incubation period of 18 hours and recorded. Each experiment was done in triplicate.

Determination of Activity Index (AI)

The activity index of the crude plant extract was calculated by comparing the mean value of the extracts with the mean value of zone of inhibition of standard antibiotic, using the following formula,

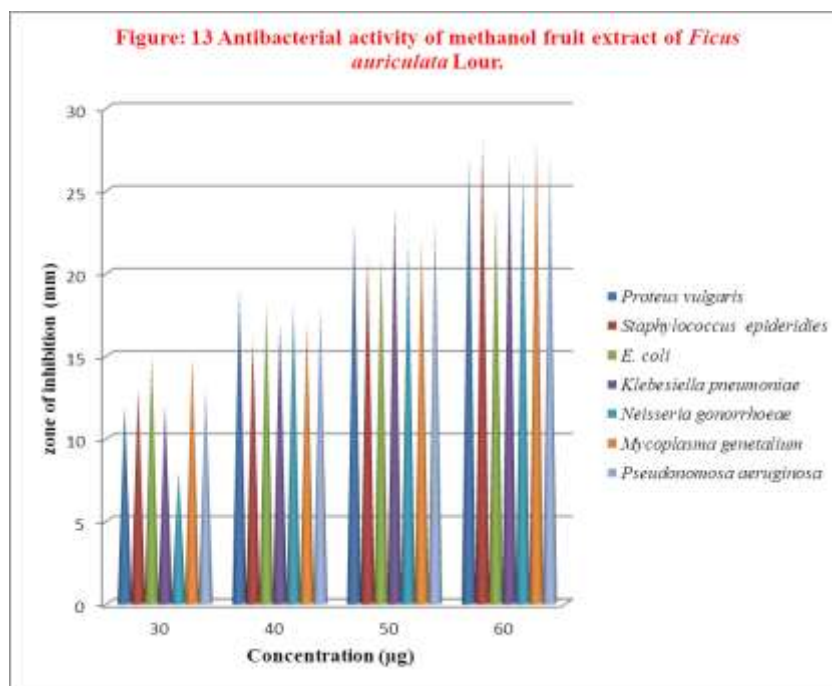
$$\text{Activity index (AI)} = \frac{\text{Zone of inhibition of extract}}{\text{Zone of inhibition of Standard antibiotic drug}}$$

RESULT AND DISCUSSION

The antibacterial activity of the methanolic fruit extract of *Ficus auriculata*. Lour was established by disc diffusion method. The methanolic fruit extracts were active against seven kinds of pathogenic bacteria. Four concentrations of the extract were used (30, 40, 50 and 60 µl). The methanolic fruit extract showed a clear zone of inhibition against *P. vulgaris*, *S. epidermidies*, *E. coli*, *K. pneumoniae*, *N. gonorrhoeae*, *M. genitalium* and *P. aeruginosa*. The methanolic fruit extract showed significant antibacterial activity as compared to standard antibiotics (amoxicillin). The zone of inhibition increased with the increase in concentration (table 1; figure 1 and Plate 1). Among the various microorganisms, the methanolic fruit extract of *F. auriculata* was more active against *Mycoplasma genitalium* (28 mm) and *Staphylococcus epidermidies* (28 mm) in concentration 60 µg and lowest effect in *E. coli* (24 mm) in concentration 60 µg.

Table 1: Antibacterial activity of methanol fruit extract of *Ficus auriculata* Lour.

S. no.	Organism Name	Control (Amoxicillin)	Concentration(μ g)			
			30	40	50	60
1.	<i>Proteus vulgaris</i> (MTCC No: 1771)	17	12	19	23	27
2.	<i>Staphylococcus epidermidies</i> (MTCC No: 435)	16	13	16	21	28
3.	<i>E.coli</i> (MTCC No: 443)	17	15	18	21	24
4.	<i>Klebsiella pneumoniae</i> (MTCC No: 109)	21	12	17	24	27
5.	<i>Neisseria gonorrhoeae</i> (MTCC No: 19424)	17	08	18	22	26
6.	<i>Mycoplasma genitalium</i> (MTCC No: 2288)	16	15	17	22	28
7.	<i>Pseudomonosa aeruginosa</i> (MTCC.No: 2488)	14	13	18	23	27

**Antibacterial activities of Methanolic Fruit extract of *F. auriculata***

a). *Proteus vulgaris* b). *Staphylococcus epidermidies* c). *E. coli*



d). *Klebsiella pneumonia* e). *Neisseria gonorrhoea* f). *Mycoplasma genitalium*

g). *Pseudomonosa aeruginosa*

There is an influence of certain microbial infection on male infertility. Several investigators have reported difference types of microorganisms in seminal fluid.^[4] Oligospermia and azoospermia are most common causes of male infertility which has been reported due to bacterial infections.^[5] Based on this information, the above microorganisms were selected for this study and also Ali Hussein Al-Marzoqi^[6] identified *P. vulgaris*, *S. epidermidies*, *E. coli*, *K. pneumoniae*, *N. gonorrhoeae*, *M. genitalium* and *P. aeruginosa* in seminal fluid and associated with male infertility. Antibacterial assay revealed, the methanolic fruit extract of *F. auriculata* having capacity to control these bacterial infections. The antibacterial property was claimed to be conferred by phytochemicals present in the plant. Tannins and flavonoids have been reported to inhibit the growth of many fungi, yeast, bacteria and viruses, alkaloids widely well known to have anti diabetic and antimicrobial activity,^[7] terpenoids, steroids and saponins may also responsible for the antibacterial activity.^[8] Based on earlier reports and present study, antibacterial assay confirmed that the methanolic fruit extract of *Ficus auriculata* possess antibacterial property.

CONCLUSION

The results of the present investigation suggest that the methanolic fruit extract of *Ficus auriculata* can be used as potential leads to discover new drugs to control some bacterial infections. The antibacterial activity of the extracts was established by disc diffusion method and the extract showed a clear zone of inhibition against *Proteus vulgaris*, *Staphylococcus epidermidies*, *E. coli*, *Klebsiella pneumoniae*, *Neisseria gonorrhoeae*, *Mycoplasma genitalium* and *Pseudomonosa aeruginosa*. Highest activity was seen in *Mycoplasma genitalium* and *Staphylococcus epidermidies* in a concentration of 60 µg.

ACKNOWLEDGMENT

We extend our sincere thanks to Management of Vivekanandha College of Arts and Science for Women (Autonomous), Tiruchengode.637205

REFERENCES

1. Lalitha, V., Kiran, B. and. Raveesha, K.A. *In vitro* evaluation of *Mimusops elengi* L. plant extract for antibacterial activity and phytochemical analysis. *Pharmacophore*, 2011; 2(1): 78 - 85.
2. Rekha Ravindran, P. Lavanya, A. Jamuna Devi, R. Krithika and Johanna Rajkumar. Phytochemical screening, GC-MS analysis and antimicrobial evaluation of Ambrex (an herbal formulation). *Journal of Chemical and Pharmaceutical Sciences*, 2014; 2: 31 - 35.
3. Bauer, A.W., Kirby, W.M.M. Sherris, J. C. and Turck, M. Antibiotic susceptibility testing by a standardized disc method. *Amer. J. Clin. Path.* 1966; 45:493-496.
4. Ajabor, L., Ezimokhai, M. and Kadiri, A. Male contribution to subfertility in Benin city, Nigeria, *Trop J Obst Gynaecol*, 1999; 2: 53.
5. Megafu U. Seminal fluid infection and oligospermia. *Trop J Obst. Gynaecol.* 2007; 9(2): 10-12.
6. Ali Hussein Al-Marzoqi, Mohammad Aboud, M. and Mohammad Sabari, A. Study of bacterial infection associated with male infertility in Hillah city-Iraq. *Journal of Biology, Agriculture and Healthcare*, 2012; 2(9): 10 - 16.
7. Revathi, P., Jayaseelan senthinath, T. and Thirumalai kolundhu subramanian, P. Preliminary phytochemical screening and GC-MS analysis of ethnolic extracts of mangrove plant-*Bruguiera cylindrica* (Rhizho) L. *International journal of Pharmacognosy and Phytochemical Research*, 2014; 6 (4): 729 - 740.
8. Akhilesh Bhat and Koteswar Anandarao Raveesh. Antibacterial activity and phytochemical analysis of the extracts of *Melastomamalabathricum* and *Holigarnaarnottiana*, important medicinal plants of Western Ghats of Karnataka, India. *Research Journal of Pharmaceutical, Biological and Chemical Sciences*, 2014; 5(6): 696 - 701.