WORLD JOURNAL OF PHARMACEUTICAL RESEARCH
SJIF Impact Factor 8.074
Volume 7, Issue 12, 691-697. Research Article ISSN 2277– 7105

ANTIMICROBIAL ACTIVITY AND MINIMUM INHIBITORY CONCENTRATION OF EHRETIA LAEVIS LEAVES EXTRACTS AGAINST DIFFERENT MICROORGANISMS

Kshama Murarkar[1]* Ph.D, Diksha Bande[2] and Anita Chandak[3] Ph.D

[1,3]Assistant Professor Kamla Nehru Mahavidyalaya.

Article Received on
22 April 2018,

Revised on 13 May 2018,
Accepted on 03 June 2018,

DOI: 10.20959/wjpr201812-12608

***Corresponding Author**
Kshama Murarkar Ph.D
Assistant Professor Kamla Nehru Mahavidyalaya.

ABSTRACT

Ehretia laevis is a medicinal plants belonging to the Boraginaceous or Borage family. The present study aimed to evaluating the in vitro antimicrobial activity of acetone and isopropanol extracts of *Ehretia laevis* plants dried leaves against *Escherichia coli*, *Staphylococcus aureus* and *Pseudomonas aeruginosa*. The extract was effective against *Escherichia coli* and *Staphylococcus aureus*. The minimum inhibitory concentration (MIC) of the all the extracts were found out by broth dilution method by taking extracts volume 1 ml and 5 ml respectively. The minimum inhibitory concentration of isopropanol extract was 1 ml and 2.5 ml, and for acetone extract was 2.5 ml and 1.25 ml respectively for *S. aureus* and *E. coli*.

KEYWORDS: *Ehretia laevis*, *Escherichia coli*, *Staphylococcus aureus* and *Pseudomonas aeruginosa*, minimum inhibitory concentration.

INTRODUCTION

Many medicinal plants are already explored for medicinal purpose, but more and greater still remains, which has great medicinal potential for social benefit. *Ehretia laevis* is a small tree belonging to the Boraginaceous or Borage family.

It is native tree of Asia, and found in China, Bhutan, India, Pakistan, Laos, Burma, and Vietnam. This plant is mainly used for wound healing, joint pain and minor fractures by local people with promising results and commonly known as khandu chakka in Maharashtra. The plants of this genus have significant medicinal importance and find uses in traditional medicine as a remedy for the treatment of diarrhea, cough, cachexia, syphilis, toothache,

stomach and venereal diseases and as an antidote to vegetable poison (The wealth of India raw materials 1952). All parts of the *E. laevis* plant are used for different medicinal purposes. Decoction of the fresh root is used in the treatment of syphilis and that of the stem bark for the treatment of diphtheria. The paste of tender leaves is used externally to cure eczema and the powder of flowers with milk is employed as an aphrodisiac (http://www.indianetzone.com/38ehretia -laevis-roxb-plant.htm). It has many uses such as ornaments, pot herbs, dye for wood, stone, medicines, wines, cosmetics etc. (S. J. Ali). The inner bark of the tree and the insipid fruit of *E. laevis* are eaten in times of scarcity. The leaves are used as cattle fodder (The Wealth of India Raw materials 1952).Literature survey revealed wide biological activity of family Boraginaceae.The leaves of *E. laevis* is used to treat ulcers and headaches (Joshi S. 2000).

The main objective of this study is to evaluate the antimicrobial and minimum inhibitory concentration (MIC) of *E. laevis* plant leaves against microorganisms.

METHODS AND MATERIAL

Collection and pretreatment of *E. laevis* leaves

Leaves of *E. laevis* plant were collected, rinsed thoroughly and dried in shed to make powder form.

Extraction by Soxhlet apparatus using acetone and isopropanol

Dried powdered leaves were with acetone and isopropanol (1:10, leaves: solvent) respectively using soxhlet apparatus. The extract was stored at 10 ^{0}C for further use.

Determination of antimicrobial activity

Antimicrobial activity was detected by Kirby- Bauer well diffusion method and minimum inhibitory concentration by using Muller Hington agar (MHA).

Well diffusion method

The pure bacterium cultures used for this study were *Escherichia coli, Staphylococcus aureus, and Pseudomonas aeruginosa.*

The broth cultures of these organisms were in 50 ml sterilized nutrient agar with one loopful of culture. It was incubated overnight at 37 ^{0}C. After incubation 0.1 ml culture was of respective organisms was spread on separate sterilized and solidified MHA plates with sterilized spreader. Then 0.1 ml extracts of leaves in each solvent was added into 2 mm well

with sterilized pipette. Control was set with 0.1 ml acetone and isopropanol in separate wells. The plates were incubated for 24 hrs at 37 ^{0}C.

Minimum Inhibitory Concentration method (MIC)

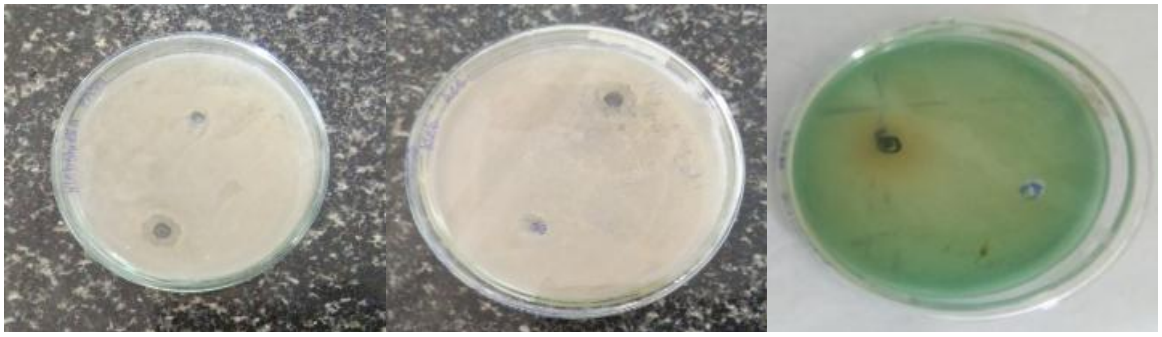
Minimum inhibitory concentration was carried out for *Escherichia coli*, *Staphylococcus aureus* by agar dilution method. Five milliliter nutrient broth was dispensed in 5 different test tubes accept first in which 9 ml broth was taken. One milliliter of extracts (in acetone / isopropanol) was added in the first tube having 9 ml of broth and mixed thoroughly. Five milliliter broth from first test tube upto fifth test tube was transferred by serial dilution method. After serial dilution, 0.1 ml broth culture (incubated overnight) was added in all the tubes, and incubated at 37 ^{0}C for 24 hours.

This 0.1 ml of broth from respective tubes was spread on respective sterilized nutrient agar plates and incubated at 37 ^{0}C for 24 hours. The concentration of extracts for MIC will be 1 ml, 0.5 ml, 0.25 ml, 0.125 ml, and 0.0625 ml.

For complete inhibition of growth of organism by Minimum Inhibitory Concentration of extract was taken 5 ml instead of 1 ml**.** The concentration of extracts for MIC will be 5 ml, 2.5 ml, 1.25 ml, 0.625 ml, and 0.3125 ml.

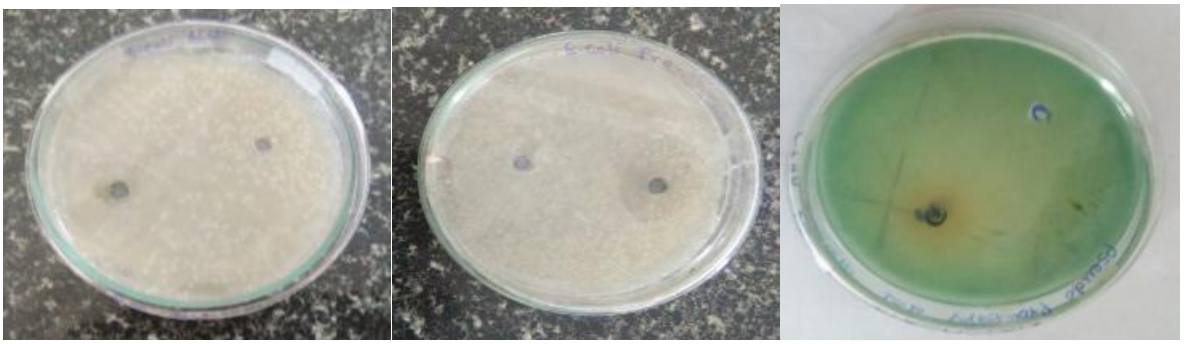
RESULT

The Antimicrobial activity was shown by all extracts against *E.coli* and *S.aureus.*



E. coli **S. aureus** **P. aeruginosa**

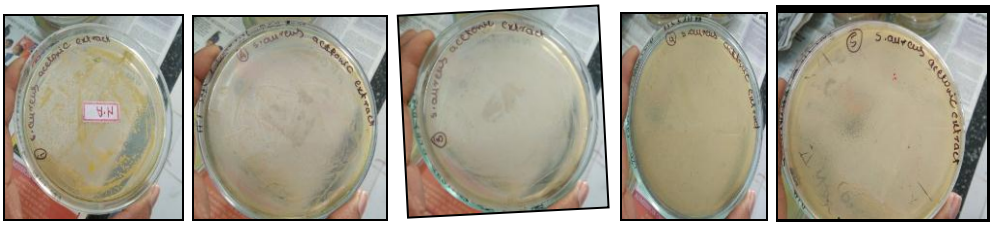
(A) With Acetone



E. coli *S. aureus* *P. aeruginosa*

(B) With isopropanaol

Photograph 1: Antimicrobial activity of *Ehretia laevis* plant dried leaves extracts against test organisms.



S. aureus

E.coli

(A) Acetone extract

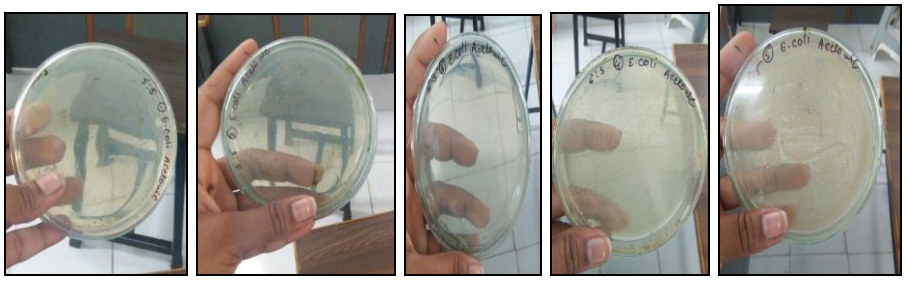
S.aureus

E.coli

(B) Isopropanol extract

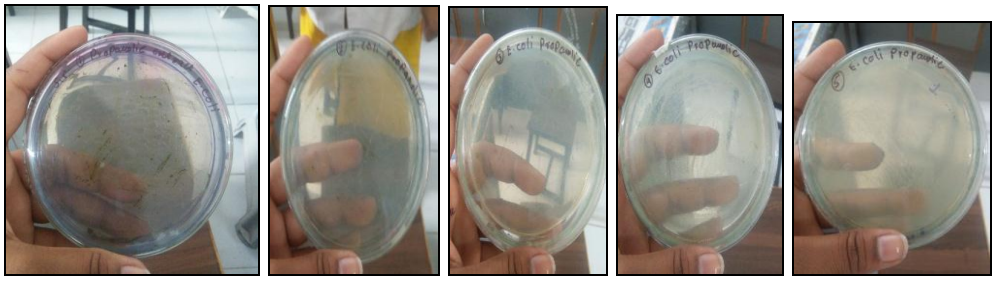
Photograph 2: Minimum inhibitory concentration of *E. laevis* leaves extract (Concentration of Extract 1 ml).

S.aureus



E.coli

(A) Acetone extract



E.coli

(B) Isopropanol extract

Photographs 3: Minimum Inhibitory Concentration of *E. laevis* leaves extract (Concentration of *E. laevis* leaves extract 5 ml).

DISCUSSION

The Antimicrobial activity was shown by all extracts against *E.coli* and *S.aureus*. This may be due to the presence antimicrobial components in these plant. There were no activity against *Pseudomonas aerugenosa.*

E. laevis (kandu chakka) plant contains tannic acid (Sivasankari Velappan 2014) which inhibits the growth of many fungi, yeasts, bacteria, and viruses (Chung KT et al 1998).

But the zone of inhibition was not clear (photographs 1). Therefore antimicrobial activity was further confirmed by MIC.

In MIC, with the acetone extract growth of *S. aureus* and *E. coli* was decreased with the increasing in serial dilution of extracts.

With the isopropanol extract growth of *S. aureus* was completely inhibited in the first petriplate with 1 ml of extract. The growth was increased as the concentration of extract was decreased. The growth of *E. coli* was decreased with the increase in concentration of extract (photograph 2).

By increasing the concentration of *E. laevis* plant extracts (5ml instead of 1ml) growth of *S. aureus* and *E. coli* was completely inhibited at concentration of 2.5 ml (second petriplate) and 1.25 ml (third petriplate) respectively for acetone extract. The growth of *E. coli* was completely inhibited at concentration 2.5 ml (second petriplate) in isopropanol extract (Photograph 3). The further increase in serial dilution growth of microorganisms was not inhibited completely.

Thakre R. et al (2016) reported the application of khandu chakka fresh leaves paste on for infected and non infected wounds, without giving any antibiotics, healed wounds completely from minimum seven days and maximum sixty six days.ion of khandu chakka.

Dhenge S.et. al (2016) did a case study on local application of (*Ehretia laevis roxb*) Ghrita in Dushtavrana.

CONCLUSION

From this study we concluded that *Ehretia laevis* plant is a very useful medicinal plant which have antimicrobial activity and can be used against infections of wound and can be used in pharmacological formulations.

ACKNOWLEDGMENT

The authors are thankful the management of Kamla Nehru Mahavidyalaya for providing facilities to complete this work.

REFERENCES

1. Ali S.J., Nasir J., Flora of Pakistan No. 191.
2. Chung KT, Wong TY, Wei CI, Huang YW, Lin Y. Tannins and human health: a review, Crit Rev Food Sci Nutr., 1998; 38(6): 421-64.
3. Joshi S.G. Medicinal Plants and IBH publishing Co. Pvt. Ltd. New Delhi, 2000; 102.

4. Salish Dhenge and Kiran Khandare. Efficacy of local application of Khandu chakka (Ehretia laevis roxb) Ghrita in Dushtavrana - A Case Report. International Ayurvedic Medical Journal. 2016; ISSN: 2320 5091. http://www.iamj.in/posts/images/upload/3726_3731.pd
5. Sivasankari Velappan, and Parimelazhagan Thangaraj. Photochemical Constituents and Antiarthritic Activity of *Ehretia laevis* Roxb. J. of Food Biochemistry, 2014; 38(4): 433–443.
6. Thakre Rushikesh, Bhutada Shyam, Chouragade Bharat, Khobragde Pramod, Harne Ketaki Ravindra. Ethano botanical properties of unexplored plant khandu chakka (*Ehretia laevis roxb*.), used on wound healing, joint pain. Int. J. Res. Ayurveda Pharm.2016 (supply 7).
7. The Wealth of India Raw materials D. E. Council of Scientific and Industrial Research, New Delhi, 1952; 111: 128.
8. http://www.indianetzone.com/38ehretia -laevis-roxb-plant.htm