

## EVALUATION OF ANTIBACTERIAL ACTIVITY OF SOME MEDICINAL PLANTS

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### ABSTRACT

The objective of the present study was to determine the antibacterial activity of ethanolic and aqueous extracts of *Erythrina indica* leaves, *Bergenia ciliata* rhizome and *Cissampelos pareira* stem. Antibacterial activities of above extracts were evaluated against five pathogenic bacterial strains i.e. *Staphylococcus aureus*, *Bacillus cereus*, *Bacillus subtilis*, *Escherichia coli* and *Salmonella typhi*. Testing was done by agar cup plate method using sterile nutrient agar. Zone of inhibition of ethanolic and aqueous extracts of above three plants was compared with that of standard Ampicillin prepared in DMSO. Ethanolic extracts of *Erythrina indica*, *Bergenia ciliata* and *Cissampelos pareira* shows greater antibacterial activity as compared to their aqueous extracts.

**KEYWORDS:** Antibacterial activity, *Bacillus cereus*, *Erythrina indica*, Agar cup plate method.

### INTRODUCTION

Infectious diseases are the most important causes as far as mortality rate is concerned<sup>1</sup>. Now a days due to increase in pollution, increased population and changed environmental conditions the bacterial and fungal infections are commonly observing everywhere. This factor causes less immunogenicity in human beings. Prolonged treatment with several antimicrobial drugs develops resistance in microorganism to the allopathic drugs & also increased toxicity in human being as well as animals<sup>2</sup>. The above data inspired us to discover a new herbal medicine, which can act as an excellent antimicrobial molecule to treat various microbial infections<sup>3</sup>. The literature serve revealed that *Bergenia ciliata*, *Cissampelos pareira*

and *Erythrina indica* possess antimicrobial activity<sup>4-6</sup>. But there is no any scientific data available till now. So it was our aim to study the antimicrobial profile of *Bergenia ciliata*, *Cissampelos pareira* and *Erythrina indica* by using different microbial species.

## MATERIALS AND METHODES

### Plant Collection and Authentication

All **crude drugs** (*Erythrina indica* leaves, *Bergenia ciliata* rhizomes and *Cissampelos pareira* stems) were provided by **S.G. Phyto Pharma Pvt. Ltd. Kolhapur, Maharashtra.** which were authenticated as per Ayurvedic standards as well as our Pharmacognostic authentication was also included to establish proper selection.

### Material

1. Nutrient agar (Hi Media)
2. Nutrient broth
3. Culture plates
4. Sterile cork borer
5. Ampicillin (Merck)
6. 70% ethanol (Merck)
7. Autoclave
8. Incubators
9. Wire loop
10. Bacterial Test Cultures

### Preparation of inoculums

The suspension of all organisms were prepared by inoculating single colony of the strain in 20 ml of nutrient broth in conical flask and incubated at 37°C for 24 hours to activate the strain. The suspension is adjusted such that it contained approximately  $1 \times 10^6$  cells/ml. It was obtained by calculating the cells by Neubers chamber.

### Culture medium

The medium was prepared by dissolving 13 gm of nutrient broth in 1000ml of distilled water adjusting the pH to  $(7.3 \pm 0.2)$  and finally subjecting it to sterilization in an autoclave at 121°C for 15 min.

### Microorganisms

Standard cultures of following microorganisms were obtained from **SG. Phyto Pharma Pvt. Ltd. Kolhapur, Maharashtra**. The microorganisms were identified by staining techniques. The organisms were maintained by sub culturing at regular intervals in nutrient agar medium.

Gram + Ve Bacteria: *Staphylococcus aureus*

*Bacillus cereus*

*Bacillus subtilis*

Gram - Ve Bacteria: *Escherichia coli*

*Salmonella typhi*

### Determination of zone of inhibition by Agar cup plate method

The antibacterial activity of ethanolic and aqueous extract of *Erythrina indica* leaves, *Bergenia ciliata* rhizome and *Cissampelos pareira* stem was performed using Agar cup-plate method. 20ml of sterile nutrient agar medium was poured into sterile Petri Plates (10x10 cm) , added 0.1 ml of the above diluted culture in to each plate and the plates were dried for 30 minutes at 37°C and allowed to solidify. Bores of 6 mm (approximate) diameter were made with sterile cork borer in the inoculated agar. The bores were filled with the plant extracts ( 2.5 mg/ml & 5mg/ml). Ampicillin (1 mg/ ml) and 70% ethanol were used as standard and control. Then the Petri Plates were kept in refrigerator for 2 hrs to allow uniform diffusion of plant extracts into agar medium. Finally all the plates were incubated for 48 hours at 37°C. At the end of incubation period, the clear zone of inhibition around the bores was measured in millimeter (mm).

### Methods

#### Preparation of extracts

Air dried coarsely powdered plant materials of (*Erythrina indica*, *Bergenia ciliata* & *Cissampelos pareira*) were extracted with water for 48 hrs by maceration and with ethanol (95%) using soxhlet apparatus for 4-5 hrs. Both the extracts were concentrated at low pressure by rotary flash evaporator and finally air-dried.

### RESULT AND DISCUSSION

Plants and their extracts have immense potential for the management and in the treatment of bacterial infections. The phyto-medicines as a antibacterial agents are not only cheap and affordable but are also safe as hyper sensitive reactions are rarely encountered with the use of

these agents. However, the discovery and use of allopathic drugs led to a remarkable decline in the popularity of herbal medicines used in the therapy.

#### Antibacterial Activity of *Erythrina indica*, *Bergenia ciliata*, *Cissampelos pareira* Extract

The results of zone of inhibition of the ethanolic and aqueous extracts of *Erythrina indica*, *Bergenia ciliata*, *Cissampelos pareira* and comparison with standard antibiotic Ampicillin were recorded in Table 1,2,3,4,5 & 6 respectively. The result shows that the ethanolic extract of *Erythrina indica*, *Bergenia ciliata*, *Cissampelos pareira* at concentration 5mg/ml shows excellent antibacterial activities in comparison to concentration 2.5 mg/ml. It also indicates that all ethanolic extracts shows antibacterial activity towards all the five investigated pathogenic bacteria. In case of *Bergenia ciliata* ethanolic extract the highest antibacterial activity was found towards *Escherichia coli* and *Bacillus cereus* than the remaining bacteria. While in case of ethanolic extracts of *Erythrina indica* and *Cissampelos pareira* the highest antibacterial activity was found towards *Staphylococcus aureus*, *Salmonella typhi* & *Bacillus subtilis*, *Escherichia coli*. The extract shows potential antibacterial properties comparable with that of standard Ampicillin against the organisms tested.

**Table 1. Antibacterial Activity Of Aqueous Extract of *Bergenia ciliata* Rhizome.**

Bacteria	Diameter of Zone of Inhibition in mm		
	Aqueous Extract of <i>Bergenia ciliata</i>		Ampicillin
	2.5mg/ml	5mg/ml	1mg/ml
<i>Staphylococcus aureus</i>	08	12	32
<i>Bacillus cereus</i>	12	14	26
<i>Bacillus subtilis</i>	07	14	30
<i>Escherichia coli</i>	08	10	24
<i>Salmonella typhi</i>	11	15	20

**Table 2. Antibacterial Activity Of Ethanolic Extract of *Bergenia ciliata* Rhizome.**

Bacteria	Diameter of Zone of Inhibition in mm		
	Ethanolic Extract of <i>Bergenia ciliata</i>		Ampicillin
	2.5mg/ml	5mg/ml	1mg/ml
<i>Staphylococcus aureus</i>	15	30	32
<i>Bacillus cereus</i>	13	28	26
<i>Bacillus subtilis</i>	17	27	30
<i>Escherichia coli</i>	15	26	24
<i>Salmonella typhi</i>	11	19	20

**Table 3. Antibacterial Activity Of Aqueous Extract of *Erythrina indica* Leaves.**

Bacteria	Diameter of Zone of Inhibition in mm		
	Aqueous Extract of <i>Erythrina indica</i>		Ampicillin
	2.5mg/ml	5mg/ml	1mg/ml
<i>Staphylococcus aureus</i>	15	20	32
<i>Bacillus cereus</i>	10	18	26
<i>Bacillus subtilis</i>	11	19	30
<i>Escherichia coli</i>	11	20	24
<i>Salmonella typhi</i>	14	16	20

**Table 4. Antibacterial Activity Of Ethanolic Extract of *Erythrina indica* Leaves.**

Bacteria	Diameter of Zone of Inhibition in mm		
	Ethanolic Extract of <i>Erythrina indica</i>		Ampicillin
	2.5mg/ml	5mg/ml	1mg/ml
<i>Staphylococcus aureus</i>	20	35	32
<i>Bacillus cereus</i>	16	22	26
<i>Bacillus subtilis</i>	16	28	30
<i>Escherichia coli</i>	19	21	24
<i>Salmonella typhi</i>	17	22	20

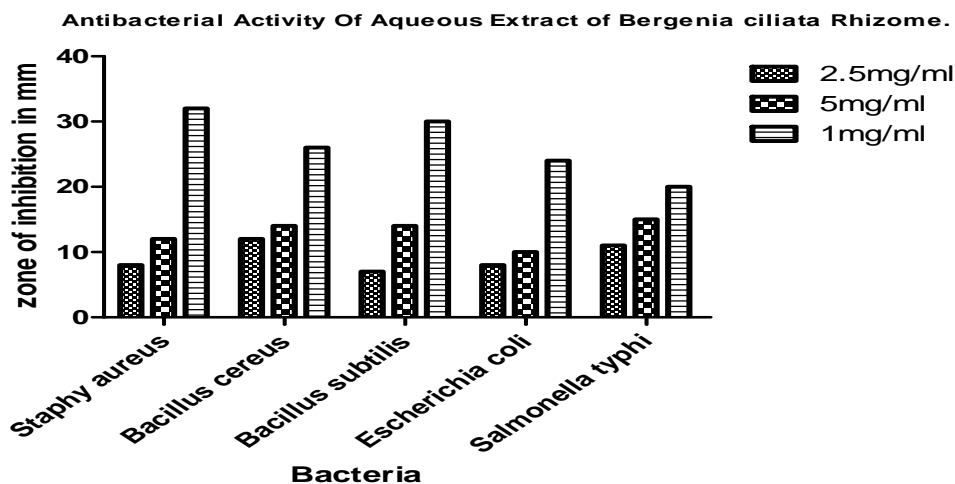
**Table 5. Antibacterial Activity Of Aqueous Extract of *Cissampelos pareira* Stem.**

Bacteria	Diameter of Zone of Inhibition in mm		
	Aqueous Extract of <i>Cissampelos pareira</i>		Ampicillin
	2.5mg/ml	5mg/ml	1mg/ml
<i>Staphylococcus aureus</i>	09	15	32
<i>Bacillus cereus</i>	09	10	26
<i>Bacillus subtilis</i>	10	14	30
<i>Escherichia coli</i>	07	13	24
<i>Salmonella typhi</i>	08	12	20

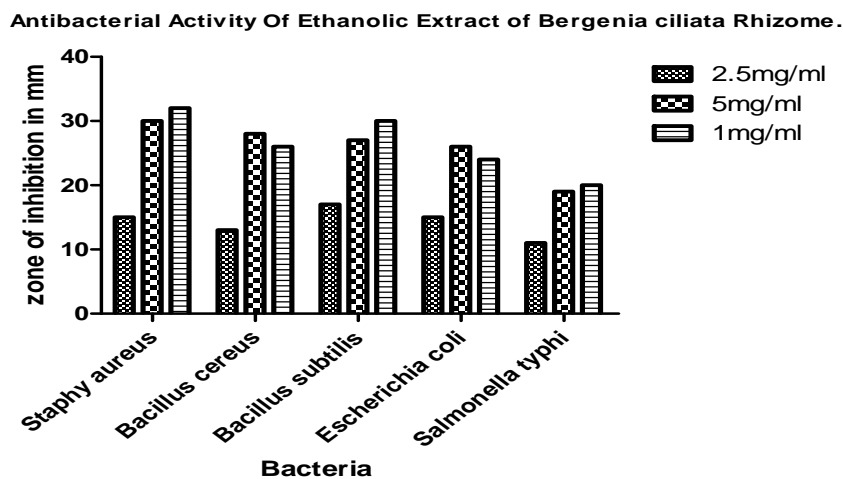
**Table 6. Antibacterial Activity Of Ethanolic Extract of *Cissampelos pareira* Stem.**

Bacteria	Diameter of Zone of Inhibition in mm		
	Ethanolic Extract of <i>Cissampelos pareira</i>		Ampicillin
	2.5mg/ml	5mg/ml	1mg/ml
<i>Staphylococcus aureus</i>	15	26	32
<i>Bacillus cereus</i>	14	29	26
<i>Bacillus subtilis</i>	12	31	30
<i>Escherichia coli</i>	17	28	24
<i>Salmonella typhi</i>	15	18	20

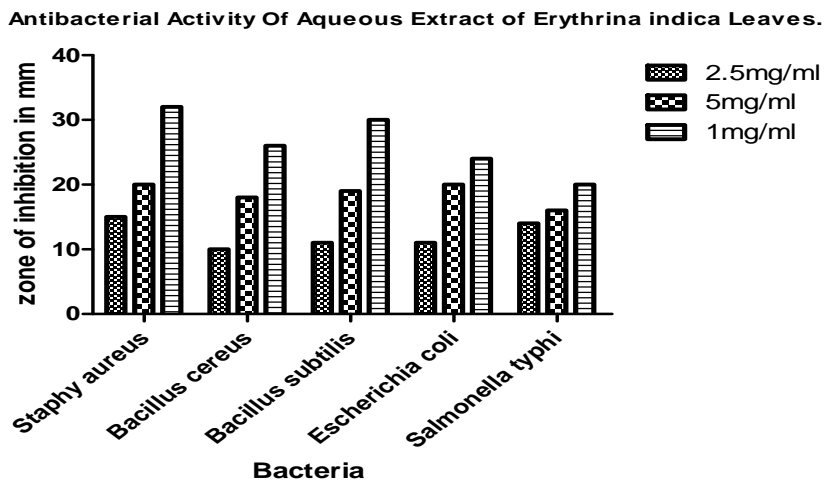
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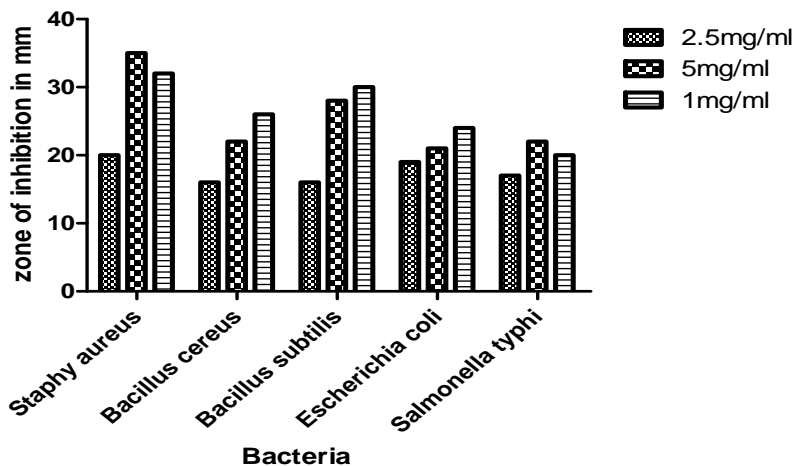


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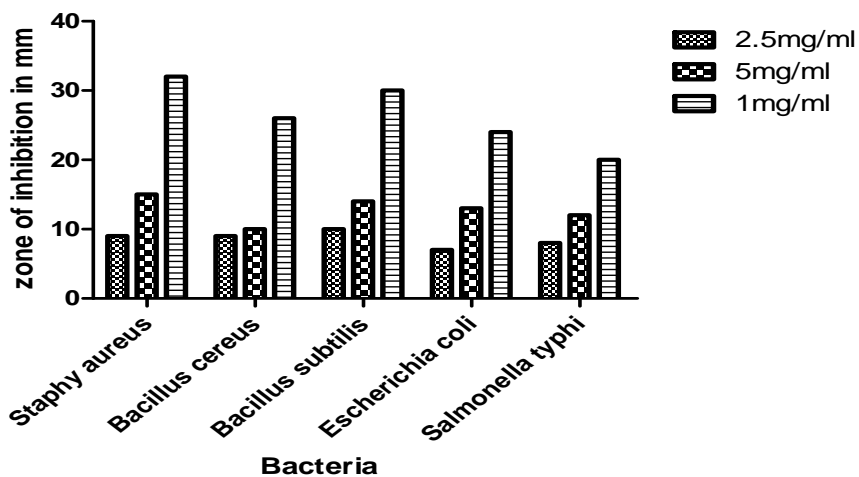
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Antibacterial Activity Of Ethanolic Extract of *Erythrina indica* Leaves.



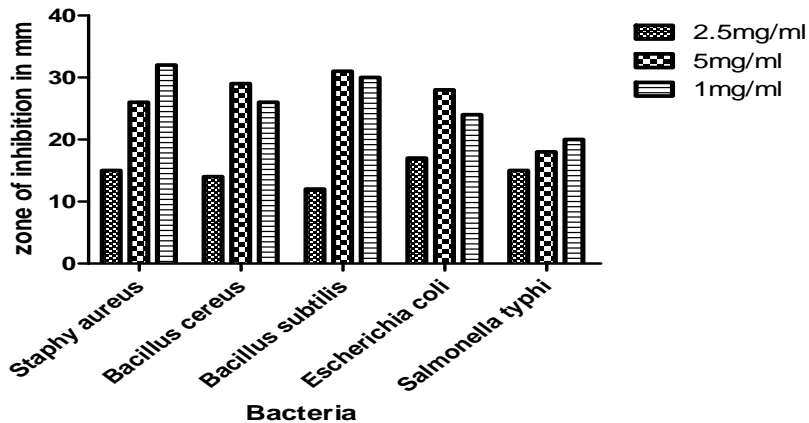
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Antibacterial Activity Of Aqueous Extract of *Cissampelos pareira* Stem.



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Antibacterial Activity Of Ethanolic Extract of *Cissampelos pareira* Stem.



## CONCLUSION

The antibacterial activity of ethanolic extracts of all the three medicinal plants ( *Erythrina indica*, *Bergenia ciliata* & *Cissampelos pareira* ) were promising against *Staphylococcus aureus*, *Bacillus cereus*, *Bacillus subtilis*, *Escherichia coli* and *Salmonella typhi* . It is inhibiting both gram positive and gram negative bacteria. The antibacterial activity of the extracts was done on some standard and wild pathogenic bacterial strains such as *Staphylococcus aureus*, *Bacillus cereus*, *Bacillus subtilis* *Escherichia coli* and *Salmonella typhi*. The testing was done by the agar cup plate method using sterile top agar. Zone of inhibition of extract ( 2.5mg/ml and 5mg/ml) was compared with that of standard Ampicillin (1 mg/ml) prepared in DMSO. Ethanolic extracts of *Erythrina indica*, *Bergenia ciliata* & *Cissampelos pareira* shows greater antibacterial activity as compared to their water extracts and could be the budding source to develop new antimicrobial agents.

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