



**IN VITRO ANALYSIS OF ANTIMICROBIAL ACTIVITY OF STEM EXTRACTS OF  
*ERYTHRINA VARIEGATA* L: A USEFUL MEDICINAL PLANT**

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**ABSTRACT**

Antimicrobial activity of methanolic, chloroform, hexane and aqueous stem extracts of *Erythrina variegata* L. were analyzed against nine bacterial pathogens such as *Salmonella typhi*, *Pseudomonas putida*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Klebsiella* sp., *Proteus* sp., *Serratia marcescens*, *Bacillus circulans*, *Staphylococcus aureus* and five fungal pathogens viz. *Aspergillus niger*, *Aspergillus fumigatus*, *Sclerotium* sp., *Rhizopus* sp., *Candida* sp. Among all the strains, few strains like *Serratia marcescens*, *E.coli*, *Pseudomonas putida*, *Bacillus circulans* was found to be more susceptible strains and among all the solvent extracts methanol showed promising antimicrobial activity. The results obtained in this study suggested that the stem extract of *Erythrina variegata* L. have high potential for use in the treatment of diseases caused by these test organisms.

**KEY WORDS:** Antimicrobial activity, disc diffusion, *Erythrina variegata* L., methanolic extracts



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## INTRODUCTION

Infectious diseases are the leading cause of death worldwide. Antibiotic resistance has become a global concern<sup>1</sup>. Resistance to antimicrobial agents is emerging in a wide variety of pathogens and multiple drug resistance is becoming common in diverse organisms<sup>2</sup>. The microbial resistance is growing day by day and the outlook for the use of antimicrobial drugs in the future is still uncertain. Therefore, measures were taken to reduce this problem, for example, to control the use of antibiotic, develop research to better understand the genetic mechanisms of resistance, and to continue studies to develop new drugs either synthetic or natural. The ultimate goal is to offer appropriate and efficient antimicrobial drugs to the patient<sup>3</sup>. And use of herbal products as antimicrobial agents may provide the best alternative to the wide and injudicious use of synthetic antibiotics. The demand on plant based therapeutics is increasing in both developing and developed countries due to growing recognition that they are natural products, non narcotic, easily biodegradable producing minimum environmental hazards, having no adverse side effects and easily available at affordable prices. Therefore researchers are progressively turning their attention to natural products, looking for new leads to develop better drugs against microbial infections and screening of several medicinal plants for their potential antimicrobial activities<sup>4</sup>.

Hence the aim of this present study is to evaluate the antimicrobial potential of various solvent extracts of *Erythrina variegata* L. for antimicrobial susceptibility. *Erythrina variegata* (Family, Fabaceae) is a medium-sized deciduous small tree with prickly stems and branches, leaves with triangular leaflets and large coral red flowers. Different parts of this plant have used in traditional medicine as nervine sedative, febrifuge, anti-asthmatic and antiepileptic<sup>5</sup>. The bark and leaves are used in many traditional medicines, including paribhadra, an Indian preparation said to destroy pathogenic parasites and relieve joint pain. Juice from the leaves is mixed with honey and ingested to kill tapeworm, roundworm and threadworm<sup>6</sup>. It is also useful in ophthalmia and skin diseases. The leaves are used in fever, inflammation and joint pain. The juice of the leaves is used to relieve earache and toothache<sup>7</sup>. It has the reputation to stimulate lactation and menstruation and is used as laxative, diuretic and expectorant<sup>8</sup>.

## MATERIALS AND METHODS

### *Collection of sample:*

The plant samples (Figure 1) were collected from Mandargiri, Angul district of Odisha during month of May. After collection, the samples were deposited at the herbarium of Institute of Mineral and Material Technology (IMMT), Bhubaneswar, Odisha and were identified following Flora of Orissa Volume-I<sup>9</sup>.



**Figure- 1**  
**Flowering twig of *Erythrina variegata* L.**

#### **Preparation of Plant extracts**

The plant test samples (stem) were first cut into smaller pieces and then air dried under shade around 15-20 days. After shade drying plant samples were pulverized by grinder. The dried powdered material was successively extracted using soxhlet extractor using various organic solvents such as methanol, hexane, chloroform and water. After the extraction, extracts were filtered by Whatmann filter paper and filtrate was concentrated in rotary evaporator to obtain the crude extracts. The dried extracts were collected in a sterile bottle and stored at 4°C for the antimicrobial study.

#### **Collection of test organism**

On the basis of pathogenic importance, nine pathogenic bacterial strains (*Salmonella typhi*, *Pseudomonas putida*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Klebsiella* sp., *Proteus* sp., *Serratia marcescens*, *Bacillus circulans*, *Staphylococcus aureus*) and five fungal strains (*Aspergillus niger*, *Aspergillus fumigatus*, *Sclerotium* sp., *Rhizopus* sp., *Candida* sp.) were collected from Department of Microbiology, SCB medical college, Cuttack. All strains were maintained at 4°C prior to further use.

#### **Antimicrobial assay**

Antimicrobial susceptibility test was carried out by disc diffusion method<sup>10</sup>. Bacterial and fungal cultures were used to lawn in nutrient agar and potato dextrose agar plates respectively by using a sterile swab. The plates were dried for 5 minutes and then used for sensitivity test. The plant extracts were freshly reconstituted with 1 ml DMSO and 1 ml solvent such as methanol, hexane, chloroform and aqueous, respectively. After swabbing the plates, sterile 5 mm disc were placed equidistantly in the plates. Discs were impregnated with 50µl of plant extracts and control respectively. The plates were incubated over night in the incubator at 37°C. The diameter of zone of inhibition surrounding the disc was recorded.

## **RESULTS**

The results of antibacterial activity of *E. variegata* L. stem extract were presented in the Table 1. Methanolic extract was found to have highest activity against all pathogens and highest zone of inhibition was observed against *S. marcescens* (20mm) as shown in figure 2. In chloroform extracts, highest activity was shown by *E.coli* (figure 3) followed by *Klebsiella* sp.,

*Proteus* sp., *P. putida*, *S. marcescens*. The aqueous extract showed positive effect against few bacterial pathogens such as *P. putida*, *S.*

*aureus* and *B. circulans*. Whereas a negative effect was observed in all the strain against hexane extract.

**Table-1**  
**Antibacterial activity of plant extract of *Erythrina variegata* L.**

Sl/No	Name of the extract	Zone of Inhibition (mm)								
		Gram negative bacteria							Gram positive bacteria	
		<i>EC</i>	<i>K</i>	<i>P</i>	<i>PP</i>	<i>PA</i>	<i>SM</i>	<i>ST</i>	<i>SA</i>	<i>BC</i>
1	Methanol	18	15	14	18	17	20	12	10	15
2	Chloroform	19	14	18	11	-ve	10	-ve	-ve	-ve
3	Aqueous	-ve	-ve	-ve	10	-ve	-ve	-ve	7	10
4	Hexane	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve

(*EC- E.coli*, *K- Klebsiella sp*, *P- Proteus sp.*, *PP- Pseudomonas putida*, *PA- P. aeruginosa*, *SM- Serratia marcescens*, *ST- Salmonella typhi*, *SA- Staphylococcus aureus*, *BC- Bacillus circulans*, -ve- negative activity)



**Figure-2**

**Antibacterial activity of methanol extract against *Serratia marcescens***



**Figure-3**

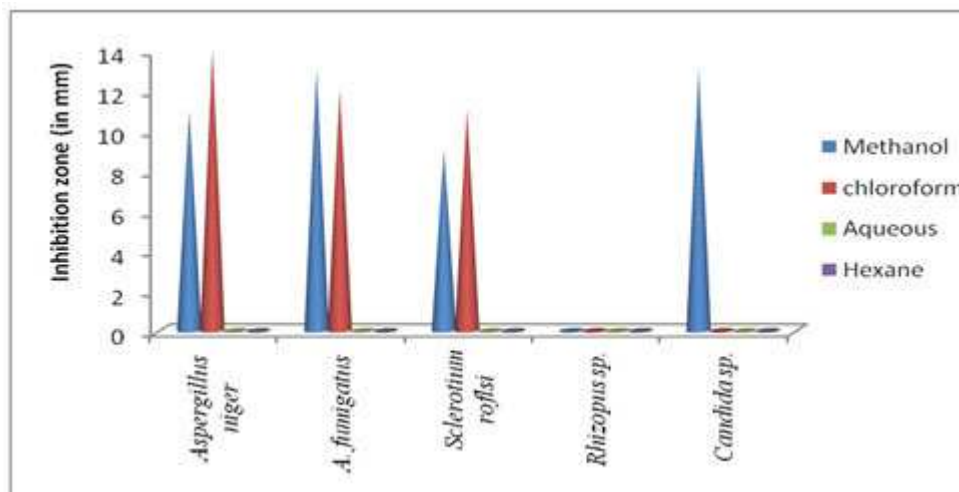
**Antibacterial activity of chloroform extract of *Erythrina variegata* L. against *E.coli***

In case of antifungal activity, the methanol extracts has shown inhibition zone against

*Aspergillus niger*, *A. fumigatus*, *S. roflsi* and *Candida* sp. whereas negative activity was

found against *Rhizopus* sp. (Figure 4) and maximum zone of inhibition was observed in *Aspergillus fumigatus* as shown in figure 5. The chloroform extracts of *E. variegata* L. stem has positive effect against *A. niger*, *A.*

*fumigatus*, *S. roffsi* and negative effect against *Rhizopus* sp. and *Candida* sp. But in case of hexane and aqueous extracts, a negative effect was observed against all the test fungus.



**Figure-4**

**Antifungal activity of plant extract of *Erythrina variegata* L. against different solvents**



**Figure- 5**

**Antifungal activity of methanol extract of stem against *A. fumigatus***

Standard antibiotic activity was also carried against various test strains using ampicillin as the antibiotic. *E.coli* did not have any inhibition but inhibition zone was observed against *Klebsiella* sp. (11mm), *S. typhi* (16mm), *Proteus* sp. (13mm), *B. circulans* (14mm), *P. putida* (15mm), *S. aureus* (13mm), *S.*

*marcescens* (12mm). In all the extracts, the MIC value was observed to be 7mg/ml against most of the extracts. Hence, it has been observed that *Erythrina variegata* L. posses a potential antibacterial property that is comparable to the standard antibiotics used.

## DISCUSSION

In recent times ethno medicinal and traditional pharmacological approaches are achieving great appreciation in modern medicine, because the search for new potential medicinal plants is often based on an ethno medicinal origin<sup>11</sup>. Successful prediction of botanical compounds from plant material is largely dependent on the type of solvent used in the extraction procedure. The traditional healers or practitioners make use of water primarily as a solvent, but the present study revealed that methanol extracts of these plants were certainly much better and powerful as compared to other extracts. This may be due to the better solubility of the active components in organic solvent. The antimicrobial activity have been screened because of its great medicinal relevance with the recent years, infections have increased to a great extent and resistant against antibiotics, becomes an ever increasing therapeutic problem<sup>12, 13</sup>. Erycristagallin and orientanol B isolated from *E. variegata* showed the highest antibacterial activity against *Staphylococcus aureus*<sup>14</sup>. But according to the present study methanol extract of *Erythrina variegata* L showed maximum antibacterial activity against *Serratia marcescens*. From the present study it can be concluded that the methanol extracts of *Erythrina variegata* L. stem has the maximum

antibacterial as well as antifungal activity and can be a useful tool for drug production.

## CONCLUSION

The increasing antibiotic resistance of some pathogens that caused infectious diseases as well as the appearance of undesirable side effects of antibiotics has increased the interest of new nontoxic and more effective and antimicrobial agents among herbal plants. The use of plant compounds to treat infections is an age old practice in a large part of countries. The present study reports the antibacterial and antifungal activity exhibited by extracts of *Erythrina variegata* L. stem used in traditional Indian medicine for the treatment of various ailments. Hence it can be used as a therapeutic agent for a variety of diseases. These active plant extracts may be further subjected to biological and pharmacological investigation for isolation of antibacterial, antifungal and therapeutic compounds.

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## REFERENCES

1. Westh H, Zinn C S, Rosdahl V T. An International multicenter study of antimicrobial consumption and resistance in *Staphylococcus aureus* isolates from 15 hospitals in 14 countries. *Microb. Drug Resist*, 10:169- 176, (2004).
2. Ahmad I and Beg A Z, Antimicrobial and phytochemical studies on 45 Indian medicinal plants against multi-drug resistant human pathogens. *J Ethnopharmacol*, 74, 113 - 123, (2001).
3. Nascimento G G F, Locatelli J, Freitas P C and Silva G L. Antimicrobial activity of plant extracts and phytochemicals on antibiotic resistant bacteria. *Braz. J. Microbiol.*, 31, 247 - 256, (2000).
4. Ghosh A, Das B K, Chatterjee S, Chandra G. Antibacterial potentiality and phytochemical analysis of mature leaves of *Polyalthia longifolia*. *The South pacific J. of Natural Science*, 26: 68-72, (2008).
5. Anwar M. *Ph D. Thesis*. Karachi, Pakistan: Faculty of Pharmacy, University of Karachi;

- The pharmacognostic and pharmacological studies on medicinal valued herbal drugs, *Erythrina variegata* Var. *Orientalis*, *Matricaria chamommilla*, *Psoralea corylifolia* and *Chenopodium album*, (2006). 15.
6. Hegde N. Cultivation and uses of *Erythrina variegata* in Western India. In S.B. Westley and M.H. Powell, eds. *Erythrina in the New and Old Worlds*. Paia, HI (USA): NFRA, 77-84, (1993).
  7. Ghani A. Medicinal Plants of Bangladesh: Chemical edition. Constituents and Uses, Asiatic Society of Bangladesh, 1<sup>st</sup> edition, (1998).
  8. Whistler W A, Elevitch C R. *Erythrina variegata* (coral tree), ver. 3.1. In: Elevitch CR (ed.). Species Profiles for Pacific Island Agroforestry. Permanent Agriculture Resources (PAR), Holualoa, Hawaii. <http://www.traditionaltree.org>, (2006).
  9. Saxena H O and Brahman M. The flora of Orissa, 1<sup>st</sup> Edn, vol.1, OFDC Ltd. : 523-524,(1994).
  10. Bauer A W, Kirby W M M, Sherris J C and Tuck M. Antibiotic testing by a standardized disc diffusion method. Am J.Clin. Pathol., 45: 493-496, (1966).
  11. Muthu C, Ayyanar M, Raja N, and Ignacimuthu S. Medicinal plants used by the traditional healers in Kancheepuram District of Tamil Nadu, India. Journal of Ethnobiology and Ethnomedicine, 2: 43, (2006).
  12. Austin D J, Kristinsson K G and Anderson R M. The relationship between the volume of antimicrobial consumption in human communities and the frequency of resistance. Proc Natl Acad Sci, USA, 96: 1152-6.18, (1999).
  13. Venkatesan D and Karrunakaran C M. Antimicrobial activity of selected Indian medicinal plants. J of Phytology, 2(2): 44-48, (2010).
  14. Kumar A, Lingadurai S, Jain A, and Barman N R. *Erythrina variegata* Linn: A review on morphology, phytochemistry, and pharmacological aspects. Pharmacogn Rev., 4(8): 147-152, (2010).