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Ex vivo Screening of Stem Extracts of *Callicarpa macrophylla* Vahl. for Antifungal Activity

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ABSTRACT: *Callicarpa macrophylla* Vahl., species of Beautyberry, is used as both internally as well as externally for its several beneficial health effects. Our objective was to determine the antifungal activity of ethanolic(**SEE**) and aqueous(**SAE**) extracts of the stems of *C. macrophylla* Vahl. Agar disc diffusion method was adopted for the antifungal screening against seven fungal strains. The overall results provide promising baseline information for the potential use of the crude antifungal extracts from *C. macrophylla* in the treatment of fungal infection. Further isolation of the responsible phytoconstituents may lead this plant to reach the bed side.

KEYWORDS: *Callicarpa macrophylla*; Stem Extracts; Antifungal Activity; Agar Disc Diffusion Method; Medicinal Plants.

INTRODUCTION

In the recent years, it has been observed that life threatening systemic fungal infections have become increasingly common, especially in the immune-compromised host suffering from tuberculosis, cancer or AIDS and in organ transplant cases[1-3]. The use of natural products with therapeutic properties is as ancient as human civilization because medicinal plants are capable of synthesizing an overwhelming variety of low molecular weight organic compounds called secondary metabolites, usually with unique and complex structures[4-8]. At present, approximately 25% of drugs in modern pharmacopoeia were derived from plants(phytomedicines) and many others were synthetic analogues built on the prototype compounds isolated from plants. Indian folk medicine comprises of numerous prescriptions for therapeutic purposes such a healing of wounds, inflammation, skin infections, leprosy, diarrhea, scabies, venereal diseases, ulcers, snake bite etc[9-12].

Callicarpa macrophylla Vahl. (fam-Verbenaceae) is an erect shrub which is globally distributed across India, Nepal, Bhutan, Myanmar, South East Asia, and China. In India it is distributed in Jammu & Kashmir, Himachal Pradesh, Uttar Pradesh, Bihar, Sikkim, West Bengal, Arunachal Pradesh, Assam, Meghalaya, Nagaland, Manipur, Mizoram, Tripura, and Andhra Pradesh, up to an

altitude of 1800 meters. Leaves are 12.5-23 cm long, ovate or ovate-lanceolate, acuminate, base cuneate or rounded. Upper surface wrinkled, glabrate when mature, white-tomentose beneath with compound stellate hairs, Petiole 6-13mm long. It is flowering in August-November and fruiting October-December[13-15].

Scientific reports suggested that leaves of *C. macrophylla* have anti-inflammatory, analgesic, antipyretic activities[4, 13], while its roots have significant role in the treatment of inflammation and pain[9]. Previously we had reported the presence of glycosides, saponins, flavanoids, tannins and carbohydrates in the aqueous extract of stems of *C. macrophylla* Vahl. while its alcoholic extract have significant glycoside, flavanoid, tannins, carbohydrates and steroid content[16]. In the current research work, we are trying to provide base line information about the antifungal properties of stem extracts of *Callicarpa macrophylla* Vahl.

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MATERIALS & METHODS

Collection and Authentification of Plant Material

The drugs were collected from Banaras Hindu University campus, Varanasi and authenticated by Dr. V.K. Joshi, Dean of Faculty of Ayurveda, Institute of Medical Science, B.H.U., Varanasi and also through National Botanical Research Institute (NBRI), Lucknow. A Voucher specimen of all the plants has been preserved in the Department of Pharmacognosy, College of Pharmacy, IFTM, Moradabad, for further references. The collected leaves were shade dried 15 days and size reduced by laboratory grinder in to coarse powder. The air dried coarse powder is used for preparation of extract.

Preparation of Extracts

The ethanolic and aqueous extracts were prepared according to the standard procedure[5, 17]. The filtered, extracts were dried in a vacuum evaporator and aqueous & alcoholic extracts were kept in desiccators until further use.

Fungal Strains Used

G. fujikoroi, *C. neoformans*, *C. albicans*, *M. verrucaria*, *A. niger*, *N. crassa* and *R. oligosporus* strains were used as test fungal strains for current study.

Agar Disc Diffusion Method for Antifungal Activity

Extract was evaluated for its antifungal activity against fungi mention above using fluconazole as standard drug by disc diffusion method. Same method was adopted as described above except the culture medium. Sabouraud dextrose agar was used as culture medium. The standard and extract was treated at a concentration of 10 and 200, 400µg/disc respectively[18-20].

RESULTS & DISCUSSION

Table 1 Zone of inhibition in mm of ethanolic, aqueous extract of C. macrophylla Vahl. stems in fungal strains(SEE- Stem ethanolic extract; SAE- Stem aqueous extract; S- Standard drug).

Fungal Strains	Zone of Inhibition(mm)				
	Ethanolic Extract (SEE)		Aqueous Extract(SAE)		Standard Drug
	200 µg/disc	400 µg/disc	200 µg/disc	400 µg/disc	
G. fujikoroi	13	17	0	0	23
C. neoformans	12	15	0	0	21
C. albicans	13	16	0	0	20
M. verrucaria	12	16	0	0	22
A. niger	14	16	0	0	18
N. crassa	13	14	0	0	21
R. oligosporus	0	0	0	0	24

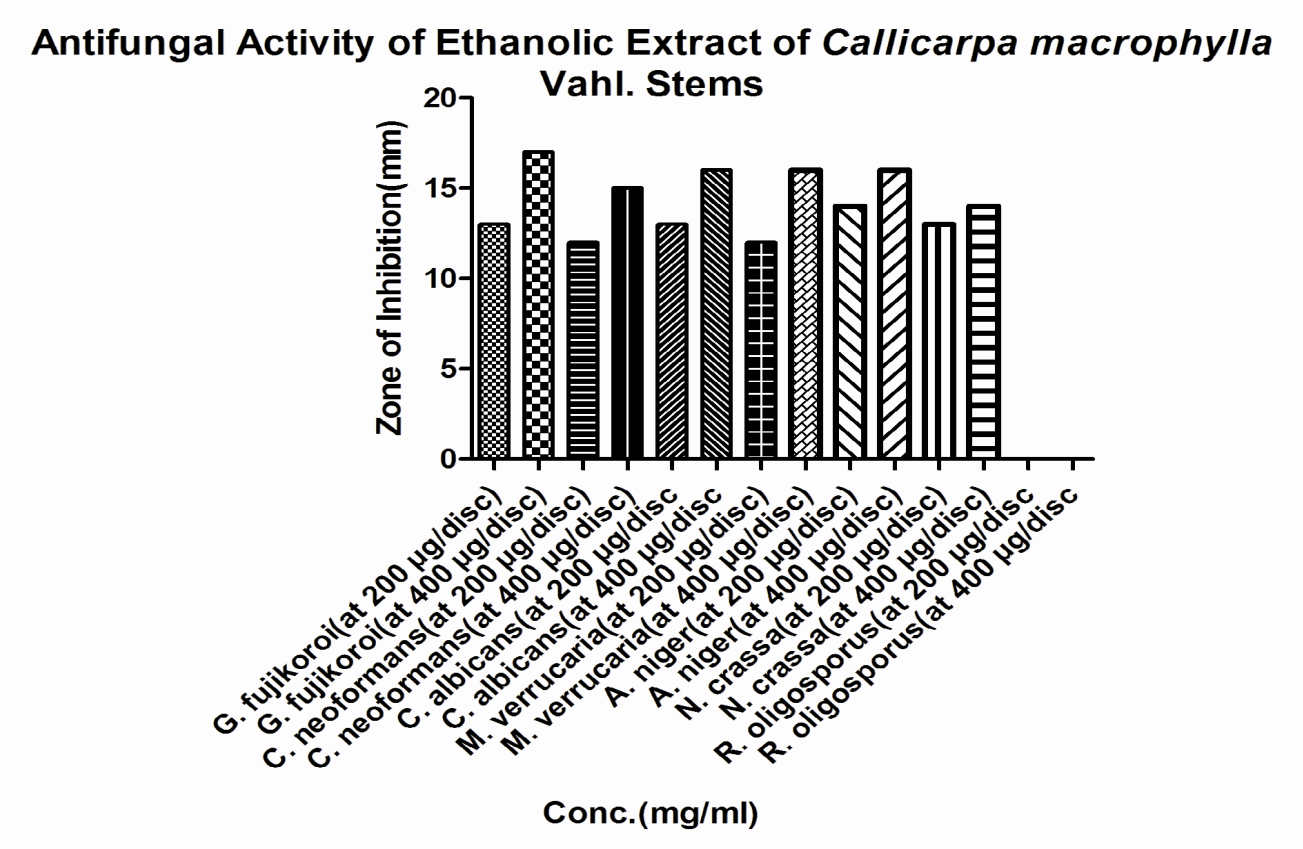

Figure 1

The ethanolic, aqueous extract of stem of *C. macrophylla* Vahl. was tested by disc diffusion method exhibited antifungal activity presented in **Table 1**. The ethanolic extract of stem exhibited antifungal activity against six strains in both 200 µg/disc and 400 µg/disc concentration. The largest zone of inhibition (17 mm in diameter) was recorded against G. fujikoroi but aqueous extract not exhibited antifungal activity. Results of ethanolic extract were also presented and explained by **Figure 1.** The benefit of local application of *C. macrophylla* Vahl. stems as antiseptic to cut/wound by Indian people could be attributed to their antimicrobial activity as observed in the present study.

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