

## Research Article



## Evaluation of Garhwal Himalaya Medicinal Plant *Eria alba*

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### ABSTRACT

In the present study, we investigated *in vitro* antibacterial, antifungal and antioxidant activity of *Eria alba* crude extract and its various fractions. The plant material was separated into its selected parts air dried ground to moderately fine powder and soxhlet extracted with increasing polarity solvent. *Eria alba* crude extract and methanolic extract showed significant activity *E. coli*, *P. aeruginosa* and *B. aureus* against human pathogens, the order of the species based on total antibacterial activity is as follows: *E. coli* > *P. aeruginosa* > *B. aureus*. The results showed that the minimum inhibitory concentration (MIC) of *Eria alba* methanolic extract was 80µg/ml against *E. coli* > *P.* In present work these whole plant crude extract and methanolic extract showed potent antimicrobial and potent antioxidant activity.

**Keywords:** *Eria alba*, Orchidaceae, Antibacterial, Antioxidant and Antifungal activity.

### INTRODUCTION

The antimicrobial plants are most potential for providing novel drug with new mechanism of action. They kill or inhibit the growth of micro-organism such as bacteria, fungi or protozoan's. The antioxidant supplements or foods rich in antioxidants may be used to help the human body in reducing oxidative damage by free radicals and active oxygen. The role of active oxygen and free radicals in tissue damage in such diseases, are becoming increasingly recognized<sup>1</sup>. The genus *Eria* (Orchidaceae) belongs to the tribe coelogyneae, It contain about 375 species in tropical Asia, Polynesia and Australia. 50 species are found in india. Commonly found throughout the Himalayan region at the altitude of 2200-3000m<sup>2</sup>. Most plants of the genus *Eria* found in India grow as epiphytes. Some are also found growing on moist, moss covered rock structures on large, hilly slopes<sup>3</sup>. On the earth, out of 4, 22,127 plant species, about 35,000 to 70,000 species are used as medicinal plants<sup>4</sup>. In the third world countries, 20,000 plants species are believed to be used medicinally<sup>5</sup>. At present, the pharmaceutical sector in India is making use of 280 medicinal plant species, of which 175 are found in the IHR<sup>6</sup>. The plants of this genus have been studied extensively because of the traditional medicinal uses associated with them. The leaves, stems and flowers are used mostly in folk medicine for the treatment of dysentery, treatment of asthma, coughs, bronchitis, eczema and wound healing. The plant leaves used as remedy for skin diseases to reduce swelling and pain<sup>2</sup>.

### MATERIALS AND METHODS

#### Collection and Identification

The fresh parts of whole plant of *Eria alba* were collected from adjoining area of Guptakashi village (Dist-

Rudraprayag Uttarakhand) in the month of September October 2013. The plant was authenticated by botanist Dr. C. S. Rana, Department of Botany and the voucher specimen number is GUH 8592. H. N. B. Garhwal (A Central University) Srinagar Garhwal, Uttarakhand India.

#### Preparation of Plant Extract

The plant material was separated into its whole plant part air dried ground to moderately fine powder and soxhlet extracted with increasing polarity solvent (petroleum ether, chloroform, ethyl acetate, acetone, methanolic, ethanolic and water)<sup>7</sup>. Each extract was evaporated to dryness under reduced pressure using rotary evaporator. The coarse powder of whole plant was subjected to successive hot continuous extraction with various solvent each time before extracting with next solvent the powdered material will be air dried (weight of crude extract 500gm). The various concentrated extracts were stored in air tight container for further studies.

#### Media

Nutrient broth, Nutrient agar, Muller Hinton agar, Malt extract broth and Sabouraud dextrose agar, Alcohol, Hydrochloric acid, alcohol, and sulphuric acid, Distilled water etc all product of Himedia Laboratories Mumbai (India) were used in this study.

#### Antibacterial assay

The disc diffusion assay methods were used to determine the growth inhibition of bacteria by plant extracts<sup>8,9</sup>. Diluted bacterial culture (100µl) was spread over nutrient agar plates with a sterile glass L-rod. 10mg/ml and 50mg/ml of the each extracts were applied to each filter paper disc (Whatman No. 1, 5 mm diam.) and allowed to dry before being placed on the agar plate. Each extract was tested in triplicate (3 discs/plate) and the plates were



inoculated at 37°C for 24 h. After incubation, the diameter of inhibition zones was measured with a caliper.

### Antifungal assay

The antifungal activity was tested by disc diffusion method<sup>10,11</sup>. The Sabouraud dextrose agar plates were each similarly seeded with each fungal strain. The 24 hrs. both culture of each bacterium and 7 days inoculated fungus culture were used to seed sterile Sabouraud dextrose agar at 45°C respectively, and fungal plates were incubated at 25-28°C for 7 days after which diameter of zones of inhibition were measured. Each disc filled with extract.

### Antioxidant Activity

The radical-scavenging capacity of *E. alba* crude extract and its various fractions of *E. alba* were determined using the DPPH radical method. A 2 ml aliquot of test solutions was added to 2 ml of  $2 \times 10^{-4}$  mol L<sup>-1</sup> ethanolic DPPH solution. The mixture was shaken vigorously and the absorbance was measured at 517 nm immediately. All the tests were performed in triplicate and mean values calculated. The antioxidant activity was expressed according to the ability of an extract to scavenge DPPH free radicals and was determined using the following equation:

$$\% \text{ Inhibition} = [1 - (A1 - A2) / A0] \times 100$$

Where A0 is the absorbance of negative control (original DPPH sample without sample), A1 is the absorbance of test sample (DPPH sample in presence of sample) and A2 is the absorbance of sample without DPPH<sup>12</sup>. The IC50 value is the concentration ( $\mu\text{g mL}^{-1}$ ) of extract/standard necessary to reduce the absorbance of DPPH by 50%

compared to the negative control. The IC50 was determined by interpolation from linear regression analysis of the antioxidant activity (% Inhibition) against sample concentration ( $\mu\text{g mL}^{-1}$ ) and the IC50 value decreases as a function of increasing antioxidant activity of samples.

## RESULTS AND DISCUSSION

Plants are important source of potentially bioactive constituents for the development of new chemotherapeutic agents. The results of antibacterial, antifungal and antioxidant activity, table 1, 2 & 3 and also figure 1, 2 & 3 reveals that antibacterial, antifungal and antioxidant activity of whole plant of *Eria alba* was evaluated against four bacterial and one fungal human pathogenic strains.

### Antibacterial and antifungal activity

*Eria alba* methanolic whole plant extract significant activity ( $23 \pm 1\text{mm}$ ,  $22 \pm 1\text{mm}$  and  $20 \pm 1\text{mm}$ ) *B. aureus* > *E. coli* > *P. vulgaris* against human pathogenic bacteria, the order of the species based on total antibacterial activity is as follows: *B. aureus* > *E. coli* > *P. vulgaris*.

### Antioxidant activity

The methanolic and ethyl acetate extracts of *Eria alba* whole plant showed significant antioxidant capacity which was found to be 85.65% and 90.03% respectively. When compared to BHT as standard, while the concentration of crude extract and petroleum ether extract was 50.452% and 25.125% (figure 3).

**Table 1:** Antibacterial activity of four bacterial strains against *Eria alba* plant whole extract, Disc size, 5 Mm, Inhibitory zone size  $\pm 1$  Mm, Mm means (millimetres) and – indicate (NIZ) No inhibitory zone.

Bacterial Name	Streptomycin	Ampicillin	Crude Extract	Petroleum ether Extract	Ethyl acetate Extract	Methanol Extract
Genus /Species /Subspecies	1 Mg/ml	1 Mg/ml	10 Mg/ml	10 Mg/ml	10 Mg/ml	10 Mg/ml
<i>E. coli</i>	19.3	15.1	22	14	16	19
<i>P. vulgaris</i>	16.6	20.6	20	6	7	17
<i>P. aeruginosa</i>	-	13.8	6	-	-	8
<i>B. aureus</i>	-	18.5	19	12	18	23

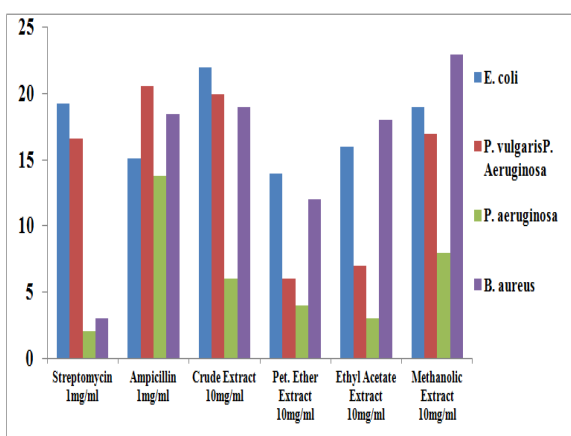
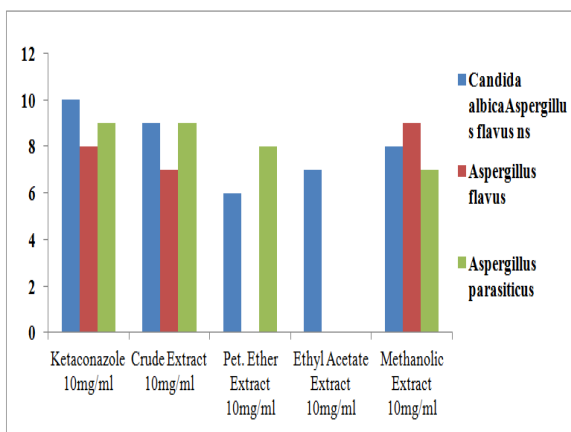
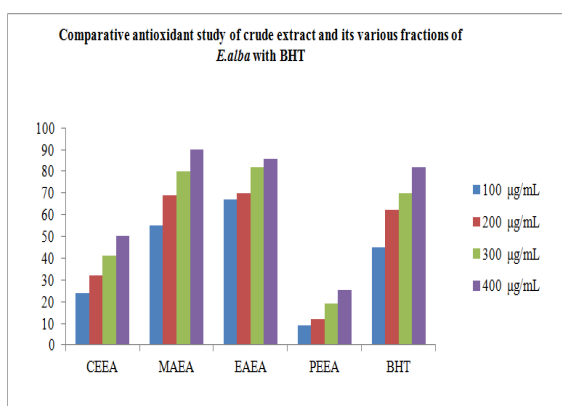
**Table 2:** Fungal activities of three fungal strains against *Eria alba* plant whole extract, Disc size, 5 Mm, Inhibitory zone size  $\pm 1$  Mm, Mm means (millimetres) and – indicate (NIZ) No inhibitory zone.

Fungal Name	Ketoconazole	Crude Extract	Petroleum ether Extract	Ethyl acetate Extract	Methanol Extract
Genus /Species /Subspecies	10 Mg/ml	10 Mg/ml	10 Mg/ml	10 Mg/ml	10 Mg/ml
<i>Candida albicans</i>	10	9	6	7	8
<i>Aspergillus flavus</i>	8	7	-	-	9
<i>Aspergillus parasiticus</i>	9	9	8	-	7



**Table 3:** Inhibitory concentration (IC<sub>50</sub>) of *E. alba* crude extract and its various fraction

Concentration (400µg/mL)	CEEA	MEEA	EAEA	PE EA	BHT
IC <sub>50</sub>	408.047	68.725	54.135	838.645	171.054

**Figure 1:** Antibacterial activity of *E. alba* different fractions activity.**Figure 2:** Antifungal activity of *E. alba* different fractions activity.

**Abbreviation:** - CEEA (Crude extract *E. alba*), MAEA (Methyl alcohol *E. alba*), EAEA (Ethyl acetate *E. alba*), PEEA (Pet. ether *E. alba*) and BHT (Butylated hydroxyl toluene)

**Figure 3:** Antioxidant activity of *E. alba* different fractions activity.

## CONCLUSION

In conclusion, all tested extracts showed significant antimicrobial and antioxidant effects to different fractions. *E. alba* was screened for the first time for antioxidant activity. Now our research will be directed to develop a broad spectrum antimicrobial herbal formulation with this plant. Even at low concentrations, these plant species contained potent antioxidant value and also showed high antibacterial and antifungal activity nearly equal to that of the commercial fungicide used as a positive control.

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