In vitro Antimicrobial Activity of Vernonia Cinerea (L) Less.

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Summary

The present study was designed to evaluate antimicrobial activities of pet ether extract and ethanol extract of whole plant of *Vernonia cinerea (L) Less* (family: Compositae). We carried out the antimicrobial screening of the extracts of *Vernonia cinerea (L) Less*. against most prevalent microbes like *S. aureus*, *P. aeruginosa, S. aureus, B. cereus*, *K. pneumonia*, *E. coli*, *A. niger* and *C. albicans* by disc diffusion method. Both the petroleum ether and alcoholic extracts *Vernonia cinerea* (PEVC and EEVC) of at various concentrations produced significant antibacterial and antifungal activities against the selected microorganisms when compared to the standard drugs Ciprofloxacin and Ketoconazole for antibacterial and antifungal activity, respectively.

Key words: Antibacterial, Antifungal, *Vernonia cinerea*.

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Introduction

The plant *Vernonia cinerea (L) Less*. belongs to Compositae family commonly known as Sahadevi is an annual herb found in upland crop area, waste places and roadsides (1). The leaves are eaten as a potherb. Fresh juice of the leaves is given in amoebiasis. A poultice of the leaves is used against humid herpes, eczema, ringworm and for the extraction of Guinea worm (2). Root is biter and used as an anthelmintic and given in diarrhea and stomachache. The flowers are used in conjunctivitis and fever (3). The seeds are commonly used as an anthelmintic and effective against roundworms and threadworms. They are useful for leucoderma, psoriasis and other chronic skin diseases (4). The use of different parts of *Vernonia cinerea (L) Less* against different worms and microorganisms suggest that the whole plant can produce significant antimicrobial activity. Thus we have selected this plant for screening antibacterial and antifungal activities against various gram (+), gram (-) and fugal microorganisms.

Materials and methods

Identification and collection of plant materials

The whole plant of *Vernonia cinerea (L) Less* was collected from potheri, chennai, identified and authentified by Dr. P. Jayaraman, Plant Anatomy Research Centre, Chennai and the plant specimen was placed in the herbarium of our college for future reference. The whole plant was collected, dried and pulverized using a mechanical grinder to coarse powder, sifted and stored in airtight containers.

Preparation of the extracts

100g of the powdered material was extracted with petroleum ether (500ml) using continuous hot percolation (Soxhlet apparatus). The product was evaporated to get a semisolid mass of pet ether extract (PEVC). Similarly ethanol extract was also prepared by using 90% ethanol. The extracts were stored in the refrigerator for future use.

Phytochemical screening

The extracts were treated with various reagents for the identification of different constituents present in the extracts (5,6).

Test microorganisms and growth media

The Gram-positive and gram negative organisms like *Staphylococcus aureus*, *Bacillus subtilis, Pseudomonas aeruginosa, Bacillus cereus, Escherichia coli* and *Klebsiella pneumonia* and the fungal organisms like *Aspergillus niger* and *Candida albicans* were used in the present study. The bacteria (nutrient agar slants), fungi including the yeasts (Sabouraud dextrose agar slants) were maintained (short term storage) at 4°C.

Antimicrobial activity

The prepared extracts were screened for antibacterial and antifungal activities. The microorganisms used are gram positive organisms like *Staphylococcus aureus*, *Bacillus cereus*, gram negative organisms like *Escherichia coli* and *Klebsiella pneumonia* and the fungal organisms like *Aspergillus niger* and *Candida albicans*. The PEVC and EEVC were used at 25, 50, 75 and 100 µg/ml concentration. The screening was carried out using disc diffusion method in nutrient agar media (7-8). The standard discs of Ciprofloxacin (100 µg/ml) and Ketoconazole (100 µg/ml) were used for antibacterial and antifungal activities, respectively. The zone of inhibitions produced by the extracts and the standard drug were tabulated and compared.

Determination of minimum inhibitory concentration (MIC)

MIC was determined by both broth dilution method (10). The extracts were dissolved 2% dimethyl sulfoxide (DMSO). Twofold serial dilutions (1.275 to 200.0 mg/ml) of the extracts were prepared in Mueller-Hinton broth for bacteria and Saboraud glucose broth for fungi. Subsequently, 0.1ml of standardized suspension of bacteria (10^6 CFU/ml) and fungal cell or spores ($5x10^5$ CFU/ml) was added to each tube (containing five extracts at a final concentration of 1.275 to 200.0 mg/ml) and incubated at 37°C for bacteria for 18 hours or at 28°C for fungi for 48 hours. The control tube contained only organisms and not the plant extract. MICs were taken as the average of the lowest concentration showing no growth of the organism and the highest concentration showing visible growth by macroscopic evaluation (11). Each assay was performed in triplicate.

Results

The percentage yields of the extracts were 4% and 3.5% for petroleum ether and ethanol extracts, respectively. The phytochemical screening suggested for the presence of fixed oils, fat, phytosterols and alkaloids in the petroleum ether extract and carbohydrates, alkaloids and glycosides in the ethanol extract. All the species were found to be active in at least one of the microbial strains compared to standard drugs. The results of the antibacterial and antifungal activity of the extracts were shown in Table 1 and 2, respectively.

Table.1. Antibacterial activity of *Vernonia cinerea* extracts against various gram (+) and (-) bacteria

Organisms	Zone of Inhibition (mm)								
	PEVC (µg/ml)				EEVC (µg/ml)				Ciprofloxacin (µg/ml)
	25	50	75	100	25	50	75	100	100
S. aureus	-	10	18	23	8	16	20	26	32
B. Subtilis	-	9	15	21	9	13	19	23	30
P. aeruginosa	5	10	15	20	-	11	16	21	28
B. cereus	-	12	18	21	-	9	14	19	31
E. coli	8	11	19	24	5	9	12	21	32
K. pneumonia	8	13	19	24	9	15	21	26	33

Table. 2. Antifungal activity of *Vernonia cinerea* extracts against various fungal strains

Organisms	Zone of Inhibition (mm)								
	PEVC (µg/ml)				EEVC (µg/ml)				Ketoconazole (µg/ml)
	25	50	75	100	25	50	75	100	100
A. niger	-	7	11	14	-	6	9	13	19
C. albicans	-	8	10	15	9	13	19	26	30

Discussion

The plant *Vernonia cinerea* has been used in the ethnomedicine for treating various infections including worm infestations. Thus we have selected this plant to prove its antimicrobial activity against commonly infecting microorganisms. For this we have prepared pet ether and alcoholic extracts and carried out the screening of antimicrobial activities against *S. aureus, B. cereus, K. pneumonia, E. coli, A. niger and C. albicans*. The extracts at 50mg/ml concentration produced comparable antibacterial and antifungal activities against selected microorganisms. Based upon the phytochemical screening both the extracts showed the presence of akaloid as a common constituent. Thus there may be some relation with the alkaloids of *Vernonia cinerea* and antimicrobial activity. Further confirmation is needed for the alkaloids by their isolation from the extracts and screening of the isolates against microorganisms. Finally structural elucidation of the isolated compounds will reveal a potential compound that will be useful as a potential agent.

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