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ANTI-FUNGAL POTENTIAL OF LEAVE EXTRACTS OF *MURRAYA KOENIGII*

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Received: 18-10-2010; Revised: 14-11-2010; Accepted: 26-11-2010

ABSTRACT

Shade dried leaves of *Murraya koenigii* Linn. (Rutaceae) was extracted successfully using soxhlet apparatus using petroleum ether (PE), benzene (BZ), chloroform (CF), acetone (AT), ethanol 95% (EN) and water (AQ). Essential oil was also isolated from the fresh leaves. Qualitative phytochemical screening showed presence of essential oil, phenolic compounds, glycogides, amino acids, resins and alkaloids. The extracts and essential oil were tested against four fungi. Zone of Inhibition was measured using the Disc Diffusion Plate Method. DW extract has no antifungal activity. AT extract was most active against Aspergillus niger, BZ extract was most active against Alternaria solani and Helminthosporium solani. EN extract was most active against Penicillium notatum. The essential oil also possesses moderate antifungal activity.

KEYWORDS: Antifungal, Disc Diffusion Plate Method, Zone of Inhibition, *Murraya koenigii*

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INTRODUCTION

Fungal diseases are a major cause of morbidity and mortality worldwide[1]. Fungi are the fifth most common pathogens, after Enterobacteriaceac, Staphylococcus aureus, Pseudomonas aeruginose and coagulase-negative Staphylococci[2]. The number of multi-drug resistant microbial strains and the appearance of strains with reduced susceptibility to antibiotics are continuously increasing. This increase has been attributed to indiscriminate use of broad spectrum antibiotics and immunosuppressive agents[3,4]. This situation provided the impetus to the search for new antifungal substances from various sources like medicinal plants. Synthetic drugs are not only expensive and inadequate for the treatment of diseases but are also often with serious side effects[5]. Traditional medicine has made use of many different plant extracts for treatment of fungal infections and some of these have been tested for in vitro antifungal activity[6].

Murraya koenigii Linn. (Rutaceae) is commonly known as curry leaf. It is an aromatic, more or less deciduous shrub or a small tree, upto 6m in height and 15-40cm in diameter, found almost throughout India and Andaman Islands. The leaves of the plant are extensively employed for flavoring in curries. The leaves are reported to contain essential oil, amino acids, glycosides and resins[7,8].

The literature review revealed that anticancer, anti-inflammatory, hypoglycemic, immunomodulatory and anthelmintic activity of *Murraya koenigii* extracts[9-12].

The present study was intended to screen different solvent extracts of the leaves as well as its essential oil against selected fungus species. Zone of inhibition of different extracts and its essential oil against selected pathogens is determined and is compared with the standard antifungal drug, Clotrimazole.

MATERIALS AND METHODS

Plant Material

Fresh plant materials (leaves) were collected from Botanical Garden of Regional Plant Resource Centre (RPRC), Bhubaneswar, Orissa after confirmation of its identity by comparing with the voucher specimen (RPRC/BGPL/2010/7/1008) present in their herbarium. After due authentication, fresh leaves were collected in bulk, cleaned thoroughly with distilled water and subsequently dried under shade. The shade dried leaves were pulverized in a mechanical grinder to obtain coarse powder.

Preparation of Extracts

The coarse leave powder (300g) was then extracted successively in Soxhlet apparatus using petroleum ether (PE), benzene (BZ), chloroform (CF), acetone (AT), ethanol 95% (EN) and distilled water (DW) for 72hr at each stage of extraction. The extracts were filtered and concentrated to dryness under vaccum. The essential oil was isolated from the fresh leaves using Clavenger's apparatus.

Drug Used

Cotrimazole was used as reference standard for antifungal studies.

Fungus Used

For the present study, Aspergillus niger, Penicillium notatum, Alternaria solani and Helminthosporium solani were used. Suitable strains of these micro-organisms were procured from Department of Microbiology, Orissa University of Agriculture and Technology (OUAT), Bhubaneswar, Orissa.

ANTIFUNGAL ACTIVITY STUDIES

Potato dextrose agars (PDA), its corresponding Broths (PDB) for fungus (HiMedia, Mumbai) were used in the study to maintain the fungus culture. The antifungal activity was carried out by disc diffusion plate method[13-15]. A comparison of the extracts was done with the standards in terms of zone of inhibition. 100mg/ml solutions of respective extracts were prepared using 6% dimethyl formamide. Clotrimazole 1mg/ml was used as standard for antifungal activity. Diameter of zone of inhibition was measured using Zone Reader and given in **Table 1**.

All above tests were carried out in aseptic environment and in triplicate and average values of overall observations were recorded.

RESULT AND DISCUSSION

The result of the present study indicates the presence of antifungal properties of various extracts as well as essential oil of leaves of *Murraya koenigii* Linn. The aqueous extract has no antifungal activity.

Aspergillus niger were inhibited by petroleum ether, benzene and acetone extracts and the essential oils. The acetone extract shows maximum zone of inhibition (10mm).

Penicillium notatum were inhibited by petroleum ether, benzene, chloroform and ethanol extracts. The ethanol extract shows maximum zone of inhibition (10mm).

Helminthosporium solani were inhibited by petroleum ether, benzene extract and essential oil. The benzene extract shows maximum zone of inhibition (10mm).

Alternaria solani were inhibited by petroleum ether, benzene, chloroform and acetone extracts and the essential oil. The benzene extract shows maximum zone of inhibition (8.33mm).

So, AT extract was most active against Aspergillus niger, BZ extract was most active against Alternaria solani and Helminthosporium solani. EN extract was most active against Penicillium notatum. The essential oil also possesses moderate antifungal activity.

Qualitative phytochemical screening showed presence of essential oil. Phenolic compounds, glycosides, amino acids, resins and alkaloids etc. Presence of constituents like flavonoids, tannins, triterpenoids in the extracts are likely to be responsible for the antimicrobial activity[16]. So the antifungal activity of the leave extracts might be due to presence of some active secondary metabolites in the plant. Our results indicates the potential usefulness of *Murraya koenigii* in the treatment of various pathogenic diseases as it may help in the discovery of new chemical classes of antibiotics that could serve as selective agents for the maintenance of human health and may provide biochemical tools for the study of fungal infectious diseases. The discovery of a potent remedy from plant origin will be a great advancement in fungal infection therapies.

CONCLUSION

Various extracts of *Murraya koenigii* Linn. Possess significant antifungal activity against selected pathogens. Further studies aimed at isolation and purification of active phyto constituents. There is a need to test the in vivo activity of the extract apart from the effect on many other fungi. This plant is an ideal candidate in the search for new bioactive phyto compounds, suggesting that a more extensive biological and chemical bioassay guided fractionation is required in order to isolate and characterize such bioactive compounds.

ACKNOWLEDGEMENT

The author is thankful to Botanical Garden of Regional Plant Resource Centre (RPRC), Bhubaneswar, Orissa for identification of the plant.

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Table 1: Antifungal activity of *Murraya koenigii* Linn

Test Organism	Diameter of zone of inhibition in mm							
	Petroleum ether	Benzene	Chloroform	Acetone	Ethanol	Aqueous	Essential oil	Clotrimazole
Aspergillus niger	8	8.66	No Zone	10	No Zone	No Zone	8.66	20.33
Pencillium notatum	6.66	9	9	No Zone	10	No Zone	No Zone	18.66
Alternaria solani	6.66	8.33	7	6.33	No Zone	No Zone	7.33	27.66
Helminthosporium solani	9.33	10	No Zone	No Zone	No Zone	No Zone	7.66	15.33

*Values are mean of three readings

Source of support: Nil, Conflict of interest: None Declared