

## ACTIVITY OF INDIAN TOPICAL SKIN PHARMACEUTICALS AGAINST SOME FUNGAL PATHOGENS

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Received: 16 January 2015, Revised and Accepted: 03 February 2015

### ABSTRACT

**Objective:** The aim was to assess the *in vitro* antifungal activity of some locally available pharmaceutical preparations used to cure skin diseases. Most commonly used preparations were evaluated for dermatophytic and non-dermatophytic fungi viz., *Aspergillus niger*, *Trichophyton rubrum*, *Candida albicans*, rhizopus microspores and *Cryptococcus neoformis*.

**Methods:** Disc diffusion method was used for inhibition zone of different preparations.

**Results:** When compared with different preparations used against skin diseases, Zalim lotion were found to be highly active against *T. rubrum*, *C. albicans*, *Rhizosporus microsporus*.

**Conclusion:** The result obtained from the experiment suggests broad spectrum activity of Zalim lotion against dermatophytic and non-dermatophytic fungi.

**Keywords:** Antifungal, Tinea, Dermatophytic, Rubrum, Aspergillus.

### INTRODUCTION

Due to frequent use of some marketed available antifungal drugs, suffer from various drawbacks due to lack of fungicidal efficacy, emergence of resistant strains drug-drug interactions and also in term of toxicity. There are still limited antifungal drugs that are available in the market. Need of novel antifungal agents that are more effective and toxicity than those already in use [1,2].

Ethnomedicinal plants preparations have been used to treat against fungal diseases particularly in central India of Madhya Pradesh. Plant preparations decorations paste of various plant extracts are also used to cure fungal diseases. Most of the antifungal drugs preparations and their mechanism of action are still unknown. A limited information exists about the use of plants and preparations toward human pathogenic yeasts dermatophytic fungi which could therapeutic benefit mainly skin mucosal and respiratory tract [3-5].

Ethnomedicinal preparations of topical use are under clinical trial for the treatment of dermatophytosis and other superficial cutaneous fungal infections in central India. Although these preparations are chemically different and has been clinically used for the topical treatment of dermatophytosis as well as of cutaneous candidiasis in central India.

To learn the antifungal potency and spectrum of various antifungal preparations, we tested *in vitro* activity against dermatophytes and a variety of causative agents of superficial cutaneous and/or subcutaneous fungal infections in comparison with fluconazole, which are widely used for the topical treatment of superficial dermatomycoses in central India.

Both oral and topical formulations used for the treatment of dermatophytosis mainly from two antifungal drug families: Azoles and allylamines [6]. Superficial mycosis (e.g. tinea pedis, tinea mannum, tinea corporis and tinea cruris) usually respond to topical antifungals e.g. (tinea pedis, tinea mannum, tinea corporis and tinea cruris) [6-8]. Most commonly used antifungal agents are azoles, allylamines, morpholine derivatives and butenafine have been also used [6]. With topical medication, primarily mild skin reactions may occur at the site of application [9]. For severe and persistent infection caused by tinea unguis and tinea capitis oral antifungal viz., terbinafine itraconazole

ketoconazole fluconazole are most commonly used [6,8]. Major side-effects of the oral formulation include neurotoxicity hematological reaction in the body and Steven-Johnson syndrome [6,10].

### METHODS

#### Collection of samples

Clinical isolates and collection strains of *Candida*, *Aspergillus*, *Cryptococcus neoformis*, *Trichophyton rubrum* and *Rhizopus microsporus* were used. One candidal species isolated from interdigital mycosis. *C. neoformis* isolated from patient cerebrospinal fluid was collected aseptically in a sterile bottle by puncturing subarachnoid space between third and fifth lumbar vertebrae. *R. microsporus* was isolated from the eye of the infected patient, and *T. rubrum* was isolated from skin of the patient. The fungal and yeast isolates were identified at society of research diagnostics and treatment of fungal diseases Society for the Indian Human Animal Mycologists Centre, Jabalpur (India) and stored in sabour dextrose broth with glycerol at 70°C. Prior to antifungal susceptibility, each isolate was inoculated on sabour dextrose agar to ensure optimum growth and purity.

#### Preparation of the inocula

The isolates were transferred from slants and then subcultured to sabour dextrose agar (SRL India) to enhance sporulation. 7-day-old cultures of *T. rubrum* and 2 day old culture of yeasts were covered with 1 ml DW and the colonies were probed with the tip of a sterile pasteur pipette to obtain a mixture of mycelium and conidia.

#### Antifungal agents

All antifungal preparations were purchased from the local market of Jabalpur Madhya Pradesh. Each antifungal cream was dissolved in dimethyl sulfoxide and stored at -80°C.

#### Disk diffusion assay

The tests were performed according to Esteban *et al.* [11]. The fungal and yeast inoculum was evenly spread on the surface of 10 cm Petri dishes containing sabouraud dextrose agar medium (SRL, India). About 50 µl of each preparation was poured in separate discs with the help of micropipette. Tween 20 (0.1%) was used as control. All the operations were carried out aseptically in the laminar chamber. Then, the disks loaded with various preparations were applied to the plates separately for both

Table 1

	Inhibition zone (mm)				
	<i>T. rubrum</i>	<i>C. albicans</i>	<i>C. neoformis</i>	<i>A. niger</i>	<i>R. microsporus</i>
Zalim lotion	40 mm±2	30 mm±3	25 mm±4	-	10 mm±3
Ring zone	4±1	3±1	-	-	-
Razal	2±2	7±3	-	-	-
B-tex	-	4±1	-	-	-
Rincolotion	4±1	19 mm±2	-	-	3±
Zerolin	-	-	-	-	-
Divya kalakep oil	-	-	-	-	-
Kanji oil	-	4±2	-	-	-

*T. rubrum*: *Trichophyton rubrum*, *C. albicans*: *Candida albicans*, *C. neoformis*: *Cryptococcus neoformis*, *A. niger*: *Aspergillus niger*, *R. microsporus*: *Rhizosporus microsporus*

yeast and fungal species after which the plates were incubated at 25°C for 5-10 days for *T. rubrum* and 2 days for yeast. After the colonies had grown, the zones of inhibition around the disks were measured and recorded.

## RESULTS

The anti-dermatophytic activities of the Zalim lotion by disc diffusion technique are given in Table 1. Zalim lotion exhibited the maximum activity with a halo of 30 mm against *T. rubrum*, *Candida albicans* followed by *C. neoformis* and *Aspergillus niger*. Other available marked available lotions viz., Rinco lotion B-Tex, Ring zone, D, K oil, Kanji oil, Razal cream, Kanji oil was inactive against these pathogenic fungal species. None of the creams was found to be protected enough against these fungal strains.

## DISCUSSION

In the present study, the antifungal and anti-yeast activity of marketed available medicinal preparations, which are most commonly used by the people were evaluated.

The present study reveals that one out of eight antifungal agents used in this study Zalim lotion had high value against fungal pathogens.

Our results related to antifungal activity of different synthetic, herbal shampoos and natural products used in traditional medicine on clinical isolates of fungi like, *Trichophyton* sps and *Aspergillus* sps can be very well visualized from the reports of Rao *et al.* [12]. They compared the effects of test compounds on seven different fungal strains, in which synthetic shampoos showed excellent inhibitory activity on all of them. Of the fungi used, *Trichophyton* was most inhibited by all test compounds and shown to be highly sensitive to them, *Aspergillus* was least inhibited. Recently many medicinal plants extracts and essential oils have been investigated for their antifungal properties against dermatophytic and yeast species.

However, no reports on the antifungal activity of the selected preparations from India have been so far come across the literature. The results of the present study indicated the presence of active antimicrobial agents in Zalim lotion and support the folk are usage of studied preparations. Development and standardization of medicinal preparations for antifungal susceptibility tests have also shown remarkable progress in the field of medical mycology. The disk diffusion method was simple, reliable, inexpensive and easily adaptable assay, which was more practical, simple and easier test in comparison with other reference methods. Modifying Zalim lotion formulation would improve its efficacy and thus provide a true value-addition to ethno medicinal based preparations with greater clinical benefits in medical mycology. However, as these preparations are commonly used by the peoples against a large number of skin diseases, it is expected to have a very less toxicity and one might conclude that these use of preparations would probably produce less side-effects and toxicity compared with conventional chemotherapeutic agents.

## ACKNOWLEDGMENT

The author is grateful to Professor Ravi Prakash Mishra Environmental Biotech Lab Department of Post-graduate Studies

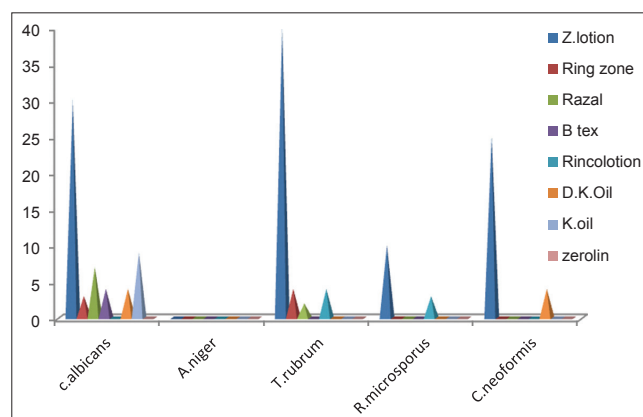


Fig. 1: Minimum inhibitory concentration ranges of topical skin pharmaceuticals

and Research in Biological Sciences Rani Durgavati University Jabalpur for giving me facility to carry out research work. Thanks are due to University Grant Commission for providing me Rajiv Gandhi National Fellowship.

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