



ORIGINAL RESEARCH PAPER

Dermatology

A CLINICOMYCOLOGICAL STUDY ON DERMATOPHYTOSIS AND ANTIFUNGAL DRUG SENSITIVITY BY DISC DIFFUSION METHOD

KEY WORDS: Dermatophytes , Culture , Antifungal Drugs, Sensitivity, Disc Diffusion Method.

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ABSTRACT

Background: Dermatophytosis is the most common superficial fungal infection across India and worldwide. Dermatophytes refer to superficial fungal infection of keratinized tissues caused by keratinophilic dermatophytes. Recently there is a shift in epidemiology from *T.rubrum* to *T.mentagrophytes* and antifungal resistance is increasing alarmingly. **Objective:** To assess the clinical profile of dermatophytosis and to identify the prevalent species and antifungal sensitivity pattern. **Methods:** 100 newly clinically diagnosed cases of dermatophytosis who were not under any topical treatment for the past 1 month were enrolled. Direct microscopy and culture followed by antifungal susceptibility was done in each case. **Results:** The most common clinical diagnosis was *Tinea corporis et cruris* (35%) followed by *Tinea cruris* (18%). Direct Microscopy (KOH) and culture showed positivity in 61% and 64% of samples respectively. *Trichophyton mentagrophytes* was the predominant dermatophyte isolate in 42% followed by *T.rubrum* in 12%. Voriconazole was the most sensitive drug with sensitivity of 75% -100% followed by Itraconazole 66.7%-100%. Fluconazole was least sensitive drug 0%-66.7% followed by Terbinafine 25%-33.3%. **Conclusion:** This study concluded that the causative agent for the dermatophytosis was changing in India and in our area *Trichophyton mentagrophytes* has replaced *Trichophyton rubrum* as the predominant species causing dermatophytosis. Voriconazole and Itraconazole were the most effective drugs and Fluconazole and Terbinafine were least effective.

INTRODUCTION

Dermatophytes are superficial fungal infections caused by dermatophytes affecting the skin, hair and/or nails¹. They are also termed as ringworm or tinea infections. Dermatophytes are ubiquitous filamentous fungi that invade and feed on keratinized tissue like skin, hair and nails of humans and other animals to produce an infection, causing an infection². Dermatophytes are classified in Three anamorphic (asexual or imperfect) genera, *Epidermophyton*, *Microsporum*, and *Trichophyton* which can be further divided into anthropophilic, zoophilic and geophilic based on their natural habitat³.

Superficial dermatophytosis affects 20-25% of the world population and is a common infective dermatoses in clinical practice. The current reported prevalence in India falls in a very wide range (6.09%-61.5%). A prevalence of 6.09% to 27.6% has been reported in studies from south India, 68 while a high prevalence of 61.5% has been recorded in north India. Tyre pile fires have been an even greater environmental problem.

Environmental factors which increases chances of infection include high humidity, high temperature, increased urbanization, use of tight-fitting clothes and occlusive footwear. It is more prevalent in developing, particularly in tropical and subtropical countries like India, evidently due to the hot and humid climatic conditions.¹⁹ In addition to climatic factors, geographic location, health-care system, overcrowding, urbanization, population migration, environmental and personal hygiene, cultural practice, the prevalence of virulent species, socioeconomic conditions, individual immune system, etc., may also affect the epidemiology and incidence of dermatophyte infections.^{19,20}

MATERIALS AND METHODS

Study Design

This hospital-based, prospective study was conducted in the Department of Dermatology, venereology and leprosy at

Jawahar Lal Nehru hospital, Ajmer, over a period of 1 year among 100 patients attending outpatient department. Informed consent was obtained from all patients to be a part of the study.

Inclusion Criteria - Newly clinically diagnosed cases of both gender, irrespective of age, of dermatophytosis who were not under any topical treatment for the past 1 month who were willing to involve in the study were included.

Exclusion criteria - Patients with associated secondary infections, pyoderma, infectious diseases like HIV, Hepatitis B, Hepatitis C, on immunosuppressive therapy were.

Specimen collection Skin scrapings, hair and nail samples were collected from the edge of the lesion with a sterile scalpel after decontaminating the affected areas with 70% ethyl alcohol. Samples were collected in a sterile paper folds and labelled with details of the patient. They were then sent to the Department of Microbiology for KOH Mount, fungal culture, species identification, and antifungal sensitivity determination.

Direct microscopic examination: KOH MOUNT: Direct microscopic examination of all samples was undertaken in KOH mount using 10% for skin and 40% for hair and Nail samples. One drop of 10% KOH was placed over a clean grease free slide with skin scraping samples placed on that and covered with a cover slip. Slides were examined in light microscopy and thin hyaline branched septate hyphae and arthrospores were identified.

Fungal cultivation: All samples were cultured on Sabouraud's dextrose agar (SDA) with chloramphenicol and cycloheximide slant tubes, and dermatophyte test media.

Incubation were done aerobically at 30°C for all dermatophytes. Cultures were observed, every alternate day, to check for the appearance of any fungal growth or

production of any pigment over the reverse side of the slant. Species were identified on the basis of real macro and microscopic features with lactophenol cotton blue staining and Urease Test and Hair perforation test.

Drug Sensitivity:

The cultured dermatophytes were subjected to drug sensitivity to the common anti fungals by Disc Diffusion Method. Sensitivity to Itraconazole, Fluconazole, Terbinafine, Griseofulvin, Voriconazole were tested. Among topical antifungals sensitivity to clotrimazole, miconazole and ketoconazole were determined. The Agar based disc diffusion method was performed as described by Nweze et al., (2010) ³² Inhibition zone diameter was measured in millimeters and recorded as sensitive, intermediate or resistant as described by Agarwal et al., 2015

RESULTS:

100 cases of clinical dermatophytosis were included in this study. The most commonly affected age group was 21–30 years in 29% of patients, followed by 31–40 years, in 27%. We depicted a male preponderance, 56 patients were male and 44 were female with the male to female ratio being 1.27:1. 84% of patients were hindu and 16 % were muslim. We observed 60% patients belong to urban area and 40% belong to rural area. majority of patients were home maker (31%) followed by students (24%). 25% of patients had a disease duration of 3–6 months, followed by 24% for 6–9 months. Positive family

Table 1- Sensitivity of antifungal drugs

Anti-Fungal Drug	T. mentagrophyte (n=42)	T. rubrum (n=12)	T. verrucosum (n=4)	T. violaceum (n=3)	Total (n=100)	Epidermophyton floccosum (n=2)	Microsporum audouinii (n=1)
Griseofulvin	61.9%	66.7%	75.0%	66.7%	42.0%	100.0%	100.0%
Fluconazole	19.0%	33.3%	50.0%	66.7%	17.0%	50.0%	0.0%
Itraconazole	97.6%	91.7%	75.0%	66.7%	60.0%	100.0%	100.0%
Terbinafine	26.2%	33.3%	25.0%	33.3%	20.0%	100.0%	100.0%
Voriconazole	97.6%	91.7%	75.0%	100.0%	61.0%	100.0%	100.0%
Ketoconazole	88.1%	33.3%	50.0%	100.0%	49.0%	100.0%	100.0%
Miconazole	81.0%	58.3%	25.0%	66.7%	47.0%	100.0%	100.0%
Clotrimazole	76.2%	41.7%	25.0%	100.0%	44.0%	100.0%	100.0%

history was observed in 52% of cases. Topical corticosteroid misuse was seen in 74% patients. Most common clinical diagnosis in our patients was Tinea corporis et cruris (35%) followed by Tinea cruris (18%), Tinea corporis (16%), Tinea corporis et cruris et faciei 7%, and Tinea capitis 7%. Direct microscopic examination (KOH) showed positivity in 61% of samples. Culture positivity for dermatophytes was seen in 64% of samples.

Sensitivity of KOH considering culture to be gold standard was 59.4% and specificity of KOH was 36.1%. Sensitivity of culture considering KOH to be gold standard was 62.3% and specificity of culture was 33.3%.

Trichophyton mentagrophytes was most common species in 42 cases of dermatophytosis followed by T. rubrum 12, Trichophyton verrucosum 4, Trichophyton violaceum 3, Epidermophyton floccosum 2 and Microsporum audouinii 1. The present study showed that Voriconazole was the most sensitive drug showing sensitivity in the range of 75% - 100%

followed by Itraconazole 66.7%-97.6% for Trichophyton species.

Fluconazole was least sensitive drug 19%-66.7% followed by Terbinafine 25%-33.3 % and Clotrimazole 25%-100% for Trichophyton species.

All drugs were 100% sensitive for Epidermophyton floccosum and Microsporum audouinii except Fluconazole 50% and 0% respectively.

Table

Anti-Fungal Drug	Epidermophyton floccosum (n=2)	Microsporum audouinii (n=1)
Griseofulvin	100.0%	100.0%
Fluconazole	50.0%	0.0%
Itraconazole	100.0%	100.0%
Terbinafine	100.0%	100.0%
Voriconazole	100.0%	100.0%
Ketoconazole	100.0%	100.0%
Miconazole	100.0%	100.0%
Clotrimazole	100.0%	100.0%

KOH	Growth	Fungus Not Grown (n=36)	Total (n=100)	p-value
	Fungus Grown (n=64)			
Fungus hyphae seen	38(59.4%)	23(63.9%)	61(61.0%)	0.657 (Not Significant)
Fungus hyphae not seen	26(40.6%)	13(36.1%)	39(39.0%)	
Total	64(100.0%)	36(100.0%)	100(100.0%)	

DISCUSSION-

Our study demonstrated male preponderance in the 21–30 and 31-40 years age group similar to other studies. v m. KOH) showed positivity in 61% of samples parallel to abida malik et al. 64% of samples showed growth on culture parallel to pathania et al. we observed greater proportions of patients were from urban areas (around 80% of patients) .7 22

Trichophyton mentagrophytes was the predominant dermatophyte isolate in 42 cases of dermatophytosis followed by T. rubrum 12, finding similar to other studies khatri .

Voriconazole was the most sensitive drug followed by Itraconazole. Fluconazole was least sensitive drug followed by Terbinafine . Among topical drugs Ketoconazole was most sensitive and Clotrimazole was least sensitive drug. Similar findings were noted by other studies .

CONCLUSIONS

We observed that Trichophyton mentagrophytes has become predominant species nowadays . Antifungal resistance is increasing probably due to topical corticosteroid abuse , irrational use of antifungal drugs and environmental factors.

Declaration Of Patient Consent-

Written consent was taken from the patients or parents of patients about procedure and for images and other clinical information reported in the article.

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Nil

Conflicts Of Interest

None

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