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CLINICO-MYCOLOGICAL PROFILE OF DERMATOPHYTOSIS AND DERMATOMYCOSIS AMONG PATIENTS ATTENDING OUTPATIENT DEPARTMENT IN A TERTIARY CARE HOSPITAL.

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ABSTRACT This study was conducted to determine the common isolates causing dermatophytosis and other fungi causing dermatomycosis in patients presenting with skin lesions with or without involvement of hair and nails. A total of 75 patients were included in the study with 86 distinctive clinical lesions, of which Tinea corporis (46.7%,) was the most predominant followed by T. cruris (22.7%), T. unguium (14.7%) with a male to female ratio of 1.3:1. KOH mounts were made from samples and examined for fungal elements and samples were sent to Microbiology laboratory for culture, and identification among them 77.3% were skin scrapings alone and 14.7% were nail clippings along with skin scrapings and 8% were hair samples along with skin scrapings. Most common isolate was *Trichophyton rubrum* (40%) followed by *Trichophyton mentagrophytes* (17.3%), *Trichophyton tonsurans* (9.3%), *Microsporum gypseum* (4%) and no fungal growth was found in 17.3% of the samples. The study signifies the importance of mycological examination in the diagnosis of dermatophytosis.

KEYWORDS: Dermatophytes, Ringworm, Tinea, Skin scrapings, KOH mount and Sabouraud's Dextrose Agar

INTRODUCTION:

Dermatophytosis is the most common superficial fungal infection seen in man caused by a group of keratinophilic fungi, infecting the superficial keratinized layer of the skin, hair, and nails. World Health Organization declared that fungal disorders are emerging significant infections in the world.

Dermatophytosis remains a significant public health problem affecting children, adolescents as well as adults and is also of cosmetic importance. Dermatophytes possess the affinity for parasitizing the keratinous layer of the skin as well as other structures rich in keratin, such as hair and nails. They produce a dermal inflammatory response with intense itching and is also of cosmetic importance.²

The dermatophytosis infection is commonly referred as 'Ringworm' due to the appearance of the lesion. Dermatophytes comprise of three major genera, *Trichophyton, Microsporum* and *Epidermophyton*, of the class hyphomycetes and division deuteromycota. There are various species of dermatophytes that cause human infection, and the species varies according to geographical location as well as part of the body affected. The infection spreads easily by direct contact from infected humans and animals or through fomites.³

The dermatophytes are the most significant cutaneous fungi because of their widespread involvement of population at large and their worldwide prevalence. Dermatophytes are frequently encountered causative agent of superficial fungal infections leading to tinea infection which are generally classified according to body site affected. Dermatophytosis is the most common type of cutaneous fungal infection seen in man and animals.⁴

The fungi that are frequently known to cause dermatomycosis are *Candida* spp, *Aspergillus* spp, *Fusarium* spp, *Acremonium* spp, *Cladosporium* spp etc. These isolates vary according to geographical location and vary according to the seasonal variations of heat and humidity in the tropical regions. Tropical and subtropical countries like India makes dermatophytosis or ringworm a common superficial fungal skin infection may reach epidemic proportions in areas with high rate of humidity, over population and poor hygienic conditions.⁵

This present work was conducted to study the common clinical presentations of dermatophyte infections, epidemiology, and prevalence of causative strains of various dermatophyte genera among the patients attending the dermatology/ internal medicine outpatient departments in a tertiary care hospital in Visakhapatnam, Andhra Pradesh, India.

MATERIALS AND METHODS:

Study design, period, and population:

This was a cross sectional study conducted after obtaining Institutional Ethics committee approval. Patients showing lesions typical of dermatophyte infections as per physicians' preliminary diagnosis were included in the study during the period of August 2019 to April 2020. Patients of all age groups and sexes were included in the study and their demographic and other clinical data collected using structured questionnaire after obtaining informed consent from all the subjects. A total of 75 patients were included in the study and appropriate sampling was done as described below.

Specimen collection:

After cleaning the lesions with 70% ethyl alcohol and allowed to dry, skin scrapings were collected from the active growing/ developing margin of the lesion into a sterile pouch or container or a sterile petri dish and transported to Microbiology department under 30 minutes. A few hairs were epilated with extra care to involve basal region where fungus normally found with sterilized tweezers and transported to the laboratory. For nail infections (Onychomycosis), clippings of the infected part and scrapings beneath the nail after cleaning with 70% ethyl alcohol.

Microscopic examination:

Skin scrapings and hair samples were placed in a drop of 10% KOH, covered with coverslip on a glass slide and left at room temperature for 30 mins. Later the slides are observed under low power followed by high power objectives on a bright ground microscope to see the presence of fungal elements such as septate and branching filaments. whereas the nail samples were placed in few drops of freshly prepared 40% KOH in a test tube/ sterile container and kept at room temperature for overnight (16 to 18 hours) and observed under the microscope next day. 6

Culture and identification:

The specimens were then inoculated onto two plates of Sabouraud's dextrose agar (SDA) with cycloheximide to reduce or inhibit the growth of common skin flora/ contaminants. One plate incubated at room temperature and the other at 37°C in the incubator. Both plates were observed for growth daily and were discarded after 21 days with no growth. Plates showing growth were subjected to identification based on growth characteristics like colony morphology, colour of the colony surface and on the reverse, texture of the colony, rate of the growth, microscopic appearance, and other relevant tests.

For microscopic identification of the fungi, a tease mount using Lactophenol Cotton Blue (LPCB) stain using a teasing needle is made on a glass slide and observed. For actual undisturbed microscopic morphology, a slide culture is performed on a 1 cm² block of SDA with a cover slip placed on it. Later this cover slip is removed and placed on a glass slide with LPCB stain and observed for fungal elements.

Biochemical tests like production of urease were done to differentiate *Trichophyton mentagrophytes* and *Trichophyton rubrum* using Christensen's urease medium⁴ by determining hydrolysis of urea by the strains.

RESULTS:

A total of 75 patients were clinically diagnosed with Eighty-six various presentations of dermatophytosis like Tinea corporis (46.7%, n=35) followed by T. cruris (22.7%, n=17), T. unguium (14.7%, n=11), T. capitis (9.3%, n=7) etc. (see Table - 1). Males (56%, n=56) were marginally affected more than females (44%, n=44) in this study with a Male to female ratio of 1.3:1.

TABLE – 1: CLINICAL PRESENTATION OF DERMATOPHYTOSIS							
Clinical presentation	Number of samples (n=75)	%					
T. barbae	6	8.0					
T. capitis	7	9.3					
T. corporis	35	46.7					
T. cruris	17	22.7					
T. incognito	2	2.7					
T. mannum	3	4.0					
T. pedis	4	5.3					
T. unguium	11	14.7					
Ectothrix	1	1.3					



PIC - 1: VARIOUS CLINICAL PRESENTATIONS OF DERMATOPHYTOSIS

Dermatophytosis in relation to age showed predominance in 19 to 45 years age group (61.3%) followed by 46 to 70 years age group (26.7%) (see Chart -1). From the 75 clinically suspect patients, various samples were collected and sent to microbiology department for further analysis.

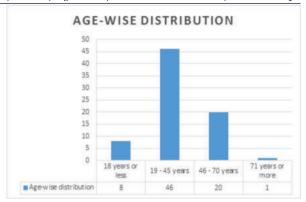


Chart-1: Age-Wise Presentation Of Clinically Suspect Cases

Among the 75 samples collected, 58 (77.3%) were skin scrapings alone and 11 (14.7%) were nail clippings along with skin scrapings and 6(8%) were hair samples along with skin scrapings.

Preliminary microscopic examination with KOH mount of the various samples sent to the microbiology department and 51 samples (68%) revealed fungal elements suggestive of dermatophytes and 24 (32%) were found to be negative. Culture on SDA were found revealed fungal growth in 62 (82.7%) and no growth in 13 (17.3%) of the samples. Twelve (50%) of the 24 samples that were KOH mount negative showed positive culture on SDA(see Table – 2).

TABLE – 2: RESULTS OF KOH MOUNT AND CULTURE ON

KOH Mount	Culture on SDA							
	Dermatophyte	Non-dermatophyte	No growth					
POSITIVE	50	0	1	51				
NEGATIVE	7	5	12	24				
Total	57	5	13	75				

The common dermatophyte isolates cultured were *Trichophyton rubrum* (40%, n=30) followed by *Trichophyton mentagrophytes* (17.3%, n=13), *Trichophyton tonsurans* (9.3%, n=7), *Microsporum gypseum* (4%, n=3) and others (see Chart – 2). No fungal growth was found in 17.3% of the samples.

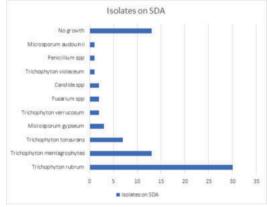
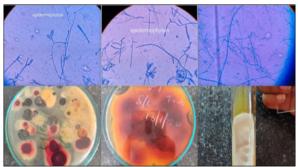


CHART-2: FUNGAL CULTURE RESULTS ON SDA



Pic – 2: Dermatophyte Growth On Sabouraud's Agar (sda) & Lactophenol Cotton Blue Mount Of The Isolates

Incidence of various species of dermatophyte and non-dermatophyte fungal isolates isolated from different clinical presentations and their frequencies are represented in the table -3.

TABLE – 3: FUNGAL ISOLATES FROM VARIOUS CLINICAL TYPES											
Clinical	Number	T. rubrum	T.	T. tonsurans	М.	T.	T.	М.	Fusarium	Candida	Penicilliu
Presentation			mentagro		gypseum	verrucosu	violaceum	audouinii	spp	spp	m spp
			phytes			m					
Tinea corporis	35	19 (54.3%)	8 (22.9%)	2 (5.7%)	0	0	0	0	0	0	0
	(46.7%)										
Tinea cruris	17	10 (58.8%)	0	4 (23.5%)	0	0	0	0	0	0	0
	(22.7%)										
Tinea unguium	11	1 (9.1%)	2 (18.2%)	0	0	0	0	0	2(18.2%)	2(18.2%)	1(9.1%)
	(14.7%)										
Tinea capitis	7 (9.3%)	0	0	0	3 (42.9%)	2 (28.6%)	1 (14.3%)	0	0	0	0
Tinea barbae	6 (8%)	2 (33.3%)	1 (16.7%)	0	0	2 (33.3%)	0	0	0	0	0
Tinea pedis	4 (5.3%)	2 (50%)	0	0	0	0	0	0	0	0	0
Tinea mannum	3 (4%)	1 (33.3%)	1 (33.3%)	0	0	0	0	0	0	0	0
Tinea incognito	2 (2.7%)	0	1 (50%)	0	0	0	0	1 (50%)	0	0	0
Ectothrix	1 (1.3%)	0	0	0	1 (100%)	0	0	0	0	0	0

Of the 75 patients included in the study, seven were found to have comorbidities (9.3%). Four (5.3%) with Type-2 Diabetes Mellitus, two (2.7%) were pregnant women and one (1.3%) patient was with hypertension and hyperthyroidism (see Chart -3).

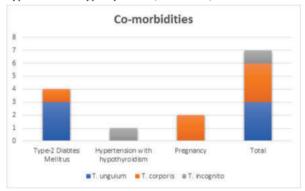


Chart - 3: Co-morbidities And Common Isolates Among The **Patients**

DISCUSSION:

Dermatophytosis has a wide geographical distribution; the species of dermatophyte causing infection may vary from region to region and are geographically restricted except some species like Trichophyton rubrum which have a cosmopolitan distribution.7 This present study was conducted to assess the clinicomycological profile of dermatophytosis.

In this study of 75 patients, T. corporis was the most common clinical presentation with 46.7% followed by T. cruris (22.7%), T. unguium (14.7%), T. capitis (9.3%), T. barbae (8%) etc which was comparable to studies conducted by Gopi et al.8 (2015), Rahman et al.9 (2015) and Majeed et al. 10 (2016).

Current study shows 61.3% of the patients belonged in the age group of 19 to 45 years with a male predominance of 1.3:1 which was consistent with other studies⁸⁻¹⁰ which showed similar incidence in the younger age group, which could probably be due to two reasons. First, young adult males are more exposed to the outer environmental elements which increases exposure chances. Second, women are less likely to go to hospital with skin conditions which could be a sensitive issue, and this may lead to marginally lesser incidence of dermatophytosis among women.

In this study, KOH mount was positive in 68% of the patients which yielded 50 (80.4%) of 62 total fungal isolates and one culture yielded no growth. Of the remaining 24 (32%) samples, 12 yielded fungal growth, 7 (58.3%) were dermatophytes (Two T. rubrum, Two T. tonsurans, One of M. gypseum, T. verrucosum and T. violaceum each) and 5 (41.7%) were non dermatophytes (Two Candida spp, Two Fusarium spp and One Penicillium spp).

The isolation rate of fungal pathogens was 82.7% which showed predominance of Trichophyton rubrum (40%), Trichophyton mentagrophytes (17.3%), Trichophyton tonsurans (9.3%), Microsporum gypseum (4%), followed by Trichophyton verrucosum,

Fusarium spp & Candida spp with 2.7% each and Trichophyton violaceum, Penicillium spp & Microsporum audouinii with 1.3% each. The higher isolation could be due to careful selection of clinical presentations and meticulous sample collection by microbiology resident postgraduates. This present study results were consistent with other studies like Gopi et al.8 (2015) and P Kannan et al.11 (2006) that showed similar outcomes.

The present 9.3% of the study subjects were associated with other medical conditions Type -2 Diabetes Mellitus, Systemic Hypertension, Hypothyroidism or Pregnancy which was similar to studies conducted by Ranganathan et al. 12 (1995) which showed 8.7% and Navya et al. showed 6.67%.

CONCLUSION:

Dermatophytosis is the most common cutaneous fungal infection. It is quite common in India due to several factors like hot humid climate owing it to be tropical country, poor hygiene, low socio-economic status, increased outdoor activities, occupational trauma, and immunosuppression. Dermatophytosis is a significant public health problem affecting children, adolescents, and adults. It is also of cosmetic importance. Indiscriminate and inappropriate use of corticosteroids or steroid based skin ointments, there is increased masking of clinical manifestations and misdiagnosis of dermatophytosis. KOH mount negativity in the samples and culture positivity observed in this study may be attributed to this. Rapid detection and diagnosis of species and knowledge of their host preference and ecology play an important role in treatment.

Determination of antifungal susceptibility patterns of dermatophytes may aid in understanding the emergence of resistance and proper clinical management of the infection.

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