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Original Research Article

Clinico-Mycological Profile of Clinically Diagnosed Cases of Dermatophytosis in North India - A Prospective Cross Sectional Study

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ABSTRACT

Background: Dermatophytosis is an important cause of superficial cutaneous fungal infections with dermatophytes being the most common etiological agents. Now a day, non dermatophytes are also being implicated as causative agents.
Aims and Objectives: This study is an attempt to find out the causative fungal agents in clinically suspected cases of dermatophytosis.
Materials and Methods: During one year of study from Jan 2013 to Dec 2013, various samples from clinically suspected cases of dermatophytosis were examined for presence of fungal elements using KOH preparation and culture on Sabouraud's dextrose agar. The organisms were identified using conventional methods.
Results: A total of 162 samples from 150 patients (12.8% patients had concomitant lesions) were obtained and processed. The most common age group involved was 21 to 30 years of age with male to female ratio of 1.6:1. The patients of rural (67%) area predominated over urban with most of the patients presenting in the monsoon season (43%). The commonest clinical presentation was Tinea corporis (40.7%) followed by Tinea unguium (21.6%). The KOH positivity was seen in 48% and culture positivity in 56% of samples. The dermatophytes (60.9%) predominate over non dermatophytes (39.1%) with *T. mentagrophytes* as the commonest fungal agent.
Conclusion: As fungal species may vary from place to place and time to time, thus mycological examination is necessary to diagnose, differentiate and treat dermatophytosis.

Key words: Dermatophytosis, Non dermatophytes, Dermatophytes, Trichophyton.

INTRODUCTION

Dermatophytoses is a group of superficial cutaneous fungal infections affecting skin, hair and nails. They are the most common type of cutaneous fungal infections seen in man and animals. The dermatophytic infections range from mild to severe depending on the host reaction to metabolic products of fungus, virulence of infecting strain and anatomic location of infection. [1] In tropical and sub-tropical countries like India, the prevalence is high and may reach epidemic proportions in areas with high rate of humidity, overcrowding and poor hygienic conditions. [2] The incorporation of aggressive therapeutical strategies like broad-spectrum antibiotics and cytotoxic drugs has further aggravated the prevalence of infection in community. [3] The causative agents responsible for dermatophytic infection vary widely in different geographical area, some

species exhibit worldwide distribution whereas others are restricted to particular continents or geographic regions. Common anthropophilic species with world-wide distribution are *Trichophyton rubrum*, *Trichophyton mentagrophytes*, *Trichophyton tonsurans*, *Microsporum audouinii* and *Epidermophyton floccosum*.[4]

Since the infections caused by fungi are often confused with other skin disorders, it is therefore necessary to make early laboratory diagnosis for better patient management. [5] Further, the epidemiology of dermatophytic infection is likely to alter with changing patterns of migration, change in socio economic conditions and growth in tourism. The nature of dermatophytoses may change with the passage of time due to evolution of preventive measures and hygienic conditions in society. [6] In this region of state of Himachal Pradesh, the geoclimatic conditions like rainfall, humidity, agricultural activities and exposure to animals are highly conducive for growth of various fungi. The current study was undertaken with the aim to isolate and identify the etiological agents in clinically diagnosed patients of dermatophytosis.

MATERIALS AND METHODS

The present prospective cross sectional study was conducted in the department of Microbiology at Dr Rajender Prasad Government Medical College and Hospital (DRPGMC & H), Kangra at Tanda, Himachal Pradesh, India. One hundred and fifty clinically diagnosed cases of dermatophytoses attending the outdoor patient department (OPD) of Dermatology, Venereology and Leprosy (Skin) of DRPGMC & H, Tanda from Jan, 2013 to Dec, 2013 were included in the present study after taking informed consent. The relevant clinical history and appropriate samples i.e. skin scrapings; hair or nail clippings were collected according to the site involved in Skin OPD and transported immediately to Department of Microbiology for processing as per standard protocol. [7] The study was approved by Institutional Ethics committee.

A total of 162 samples were collected as twelve patients (8%) had concomitant lesions at multiple sites. The specimens were subjected to KOH wet preparation using 10% and 20% for skin, hair and nails respectively for the presence of fungal elements. Following this, the specimen was inoculated into three sets of culture media i.e., Sabouraud's dextrose agar (SDA) without antibiotic, SDA with antibiotics (chloramphenicol and cycloheximide) and Dermatophyte Test Medium (DTM). The cultures were incubated at 25^0C and 37ºC and were examined daily for the first week and every alternate day thereafter up to 4 weeks for evidence of fungal growth. If no growth was obtained after 4 weeks, it was taken as negative for growth of fungus. The fungal isolates obtained were identified, based on colony morphology, pigmentation, growth rate, microscopically by lactophenol cotton blue (LCB) mount, slide culture, urease test, hair perforation test and corn meal agar test. All the data was entered and analyzed using SPSS 17.0 software (significance level of p value <0.05).

RESULTS

A total of 162 samples were collected from 150 patients of dermatophytoses as twelve patients (8%) had concomitant lesions at multiple sites. The demographic profiles of the patients are shown in (Table 1). The dermatophytosis was commonest (20.4%) among age group of 21-30 years with male to female ratio of 1.6:1. There was history of similar complaint in the past in 26.7% (40/150) of patients with no relation to the place of residence viz. rural versus urban area (p >0.05). The previous history of pets exposure were found in 48.7% (73/150) cases with more in case of rural (54.5%) than urban (36.7%) background patients (p value = 0.05). The similar complaints in the family member were seen in 12% (18/150) with more in rural (14.9%) than urban

(6.1%) cases (p value 0.18). The patient had taken treatment for the same problem in 36.7% (55/150) cases with no difference among rural and urban patient (p value = 1.0). Majority (33.3%) of the patients were students from rural background (67.3%).

The most common type of clinical presentation was Tinea corporis (40.7%) followed by Tinea unguium (21.6%), T. manuum (13%), T. pedis (10%), T. capitis (7%), T. cruris (4.3%) and T. faciei (2.5%) as shown in Table-2. The most common co-presentations were Tinea pedis with Tinea unguium (1.9%), Tinea corporis with Tinea unguium (1.9%) and Tinea manuum with Tinea unguium (1.9%), Tinea capitis with Tinea corporis (1.2%) and Tinea capitis with Tinea unguium (0.6%). The duration of disease ranged from 2 days to 13 years with mean of 21 months. Clinicomycological correlation was seen in only 61% cases. The chronic lesions were found more in nail (57.2%) than skin lesions (24.3%) (p value <0.001) (Table-3)

The KOH and culture correlation is shown in Table 4. Among 92 culture positive cases, dermatophytes and non-dermatophytes were obtained in 56 (34.6%) and 36 (22.2%) samples respectively. The KOH positivity was found in 50 (30.9%) and 25 (15.4%) dermatophytic and non-dermatophytic isolates respectively. The most common dermatophytes was *Trichophyton species* in 55 (34%) with *Trichophyton mentagrophytes* accounting for 48.2%. The *T. mentagrophytes* was predominant isolate in skin samples (18, 47.3%) followed by *T. rubrum* (12, 31.6%), *T. tonsurans* (4, 10.5%), *T. violaceum* (2, 5.3%) and *T. verrucosum* and *T. schoenleinii* in 1 (2.6%) each. In nail samples also, *T. mentagrophytes* was predominant isolate in 9 (60%) followed by *T. rubrum* and *T. tonsurans* in 4 (26.7%) and 2 (13.3%) samples each. *T. rubrum* was predominant in hair samples as the causative agent in 2 (66.6%) followed by *M. gypseum* in 1 (33.3%) sample. None of the sample showed growth of *Epidermophyton spp*. Non- dermatophytes were considered significant on repeated isolation (>2 times) and in pure culture. Among non-dermatophytes, the most common isolate was *Aspergillus species* 9 (25%) followed by *Penicillium species* 8 (22.2%) (Table-5)

Table 1: Demographic profile of patients

Particular	Variable	Values
Age distribution n (%)	Range in years	4 to 78
	Mean±2SD (in years)	33.38±18
	0-10 years	14 (9.3)
	11-20 years	28(18.5)
	21-30 years	30(20.4)
	31-40 years	24 (16)
	41-50 years	24 (16)
	51-60 years	16 (10.5)
	>60 years	14 (9.3)
Sex n (%)	Male	93(62)
	Female	57 (38)
Background n (%)	Rural	101 (67.3)
	Urban	49 (32.7)
Occupation n (%)	Students	50 (33.33%)
	Housewives	35 (23.33%)
	Employee	28(18.7%)
	Businessmen	12 (8%)
	army personnel	11 (7.33%)
	Farmers	6 (4%)
	Labourers	3 (2%)
	Drivers	2 (1.33%)
Seasonal distribution n (%)	Winters	23 (15.3)
	Summer	38 (25.3)
	Monsoon	65 (43.3)
	Post monsoon	24 (16)

Table 2: Distribution of lesions according to site of involvement

Site of involvement	Clinical Presentation	No. of sites of involvement*	Percentage
Skin	Tinea corporis	66	40.7%
	Tinea manuum	21	13%
	Tinea pedis	17	10.5%
	Tinea cruris	7	4.3%
	Tinea faciei	4	2.5%
Hair	Tinea capitis	12	7.4%
Nail	Tinea unguium	35	21.6%
	Total	**162**	**100%**

*Note: Twelve patients presented with lesions at 2 sites each leading to a total of 162 clinical presentations

Table 3: Correlation of duration of illness with site of lesion

Duration of illness	site of lesion*			
	Hair n (%)	Nail n (%)	Skin n (%)	Total n (%)
upto 6 months	10 (83.4)	13 (37.1)	86 (74.8)	109 (67.3)
7-12 months	1 (8.3)	2 (5.7)	1(0.9)	4 (2.5)
>1 year	1 (8.3)	20 (57.2)	28 (24.3)	49 (30.2)
Group Total	12	35	115	162

*% is calculated based on group total

Table 4: KOH and Culture correlation among various samples

Type of specimen			Culture		P value
			Negative n (%)	Positive N (%)	
Hair (n=12)	KOH	Negative n (%)	6 (50)	3 (25)	0.182
		Positive n (%)	0	3 (25)	
Nail (n=35)	KOH	Negative n (%)	7 (20)	3 (8.6)	<0.001
		Positive n (%)	0	25 (71.4)	
Skin (n=115)	KOH	Negative n (%)	49 (42.6)	10 (8.7)	<0.001
		Positive n (%)	8 (7)	48 (41.7)	
Cumulative (n=162)	KOH	Negative n (%)	62 (38.3)	16 (9.9)	<0.001
		Positive n (%)	8 (5)	76 (46.8)	

Table 5: Distribution of various dermatophytic and non-dermatophytic isolates obtained in the current study

Dermatophyte isolated	n (%)	Non dermatophyte isolated	n (%)
Trichophyton mentagrophytes	27 (48.2%)	*Aspergillus niger*	4 (11.1%)
Trichophyton rub rum	18 (32.1%)	*Aspergillus flavus*	3 (8.3%)
Trichophyton tonsurans	6 (10.7%)	*Aspergillus fumigates*	2 (5.6%)
Trichophyton violaceum	2 (3.6%)	*Penicillium spp.*	8 (22.2%)
Trichophyton verrucosum	1 (1.8%)	*Candida albicans*	6 (16.7%)
Trichophyton schoenleinii	1 (1.8%)	*Candida guilliermondii*	1 (2.8%)
Microsporum gypseum	1 (1.8%)	*Fonsecaea spp.*	5 (13.9%)
		Paecilomyces spp.	3 (8.3%)
		Alternaria spp.	1 (2.8%)
		Beauveria spp.	1 (2.8%)
		Fusarium spp.	1 (2.8%)
		Rhodotorula spp.	1 (2.8%)
Total	**56 (100)**	**Total**	**36 (100)**

DISCUSSION

Superficial fungal infections can be caused by dermatophytes, non dermatophytes like *Candida species*, *Scytalidium dimidiatum*, *Fusarium moniliforme* and *Scopulariopsis brevicaulis*, *Malassezia spp.*, *Hortaea werneckii*, *Piedraia hortae* and *Trichosporon spp*. [4] Dermatophytosis is widely prevalent infection in Northern India due to favourable environmental and climatic conditions. [8] Delay in diagnosis and improper treatment can lead to disseminated and refractory lesions. [4]

In the present study, people of rural background are involved in two third of cases. It could be because of their more involvement in agricultural activities, exposing them to animals and adverse weather conditions, less hygiene awareness, neglected early lesions, improper treatment in initial phases of disease. [9] The predominance of dermatophytic infections among young and adult could be attributed to their active nature and maximum involvement in outdoor activities. The male were affected more than the female, which may be due to more involvement in outdoor activities leading to increased sweating and more proliferation of fungi. [8,10,11] The hot and humid conditions favour fungus proliferation as observed in present study with maximum cases in monsoon season as has been seen earlier. [12,13]

The dermatophytic infections are transmitted from person to person by sharing common household items like clothes, fomites etc hence history of contact with cases, animals or similar episodes in the past is important to suggest possible source of infection. In accordance to previous reports, current study revealed history of similar problem in the family in 12%, pet exposure history in 48.7% and previous treatment history in 36.7% cases. [12,14,15] We observed more cases of pet exposure associated with these fungal infections in rural area compared to the urban area, it may be because of better hygienic care or early recognition of fungal infection in pet animals in the urban area. All the factors related to the demographic profile were not found to be statistically significant.

The commonest lesion of Tinea corporis observed in the current study is in agreement with various previous studies. [11,16,17] The concomitant Tinea infection

was present in 8% of cases and all these cases presented with nail involvement first followed by involvement of other body sites after a few days to weeks and this could be due to auto-infection from nail. [18,19]

The KOH examination did not show any fungal element in 48% of samples which could be due to bacterial contamination, severe inflammatory reaction which obscures fungal elements or due to minimal scaling in the lesions. The clinical importance of identifying the species of dermatophyte is to find the probable source and the prognosis of infection. [4] The culture positivity rate (56%) in our study correlated with earlier studies. [10,11] It was found to be statistically significant (<0.001). The culture negativity could be due to bacterial contamination, nonviable fungus due to prior use of topical anti-fungal agents or due to inappropriate collection of specimen. Bhagra *et al* reported culture positivity in 10% of samples. [20]

In present study, predominantly dermatophytes (60.9%) were isolated over the non dermatophytes (39.1%) as seen earlier. [21,22] In conformity to the previous reports *Trichophyton spp* was the most common genus responsible for dermatophytic infection. [14,20,23,24] The *T. mentagrophytes* was the predominant species which is in contrast to the earlier studies from adjoining region with T. *rubrum* to be the commonest one. [14,20,23] However, a study by Bhatia *et al* with samples from 3 different region of Himachal Pradesh has also found *T. mentagrophytes* to be the most common species. [24] The possible explanation for the lower incidence of *T. rubrum* in our study as compared to other studies may be because of increased association of *T. rubrum* with chronic lesions. [25] In the present study, maximum patients were of an acute duration of 0-6 months (>67%).

Thirty six (22.2%) non-dermatophytic fungi were isolated from superficial cutaneous lesions in the present study. Non- dermatophytes were considered significant on repeated isolation (>2 times), in pure culture and with a positive KOH finding. Among non- dermatophytes, the most common isolate was *Aspergillus species* 9 (25%) followed by *Penicillium species* 8 (22.2%). Several researchers have reported the association of non-dermatophytes and other fungi with dermatophytosis world over. [26,27] The findings of our study are similar to findings of Vyas *et al* [22] who reported *Aspergillus species* in 40% (8/20), *Candida species* in 15% (3/20) and *Alternaria spp.* in 10% (2/20) cases in their study. Sarma and Borthakur [11] isolated *Curvularia lunata* (3.27%), *Fusarium spp.* (3.27%) and *Aspergillus Niger*, *Aspergillus flavus* and *Penicillium spp*. in 1.63% cases each respectively.

As fungal species may vary from place to place and time to time, thus mycological examination is necessary to diagnose, differentiate and treat dermatophytosis. Hence all cases of dermatophytosis should be processed by direct microscopic examination followed by culture so as to identify the causative fungi. Early diagnosis and appropriate treatment will help in minimizing morbidity in these cases.

CONCLUSION

The study highlighted that T. corporis as most common clinical presentation of dermatophytosis followed by T. unguium with male predominance. In our region overall predominant etiological agent isolated was *T. mentagrophytes* followed by *T. rubrum*. As the number of non dermatophytic infections is increasing, hence fungal culture is mandatory to differentiate and treat dermatophytosis and non dermatophytosis.

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