



Mycological Study of Dermatophytosis in patients attending Skin OPD in JLNMC Bhagalpur

Ashif Ali Hassan

Tutor, Microbiology Department JLNMC Bhagalpur

Anjoo Anupama

Tutor, Microbiology Department JLNMC Bhagalpur

S N Tiwary

Prof. and HOD, Microbiology Department JLNMC Bhagalpur

ABSTRACT Dermatophytes affect millions of people worldwide. Dermatophytes are the most common cutaneous fungal infections seen in humans affecting skin, hairs and nails with a considerable morbidity

Material and Methods: This study involved mycological analysis of 110 cases of dermatophytosis attending the OPD of Skin and Venereology, JLNMC Bhagalpur, mycological examination with microscopy and culture using KOH and Sabouraud's Dextrose Agar (SDA) and Dermatophyte test medium (DTM) was done.

Result: KOH examination was positive in 82 (74.5%) cases while culture was positive in 77 cases (70%) *Trichophyton rubrum* was the most common isolate in 49 cases (63.6%) followed by *T. mentagrophytes* with 21 cases (27.3%) and *T. tonsurans* 2 (2.6%) and *E. floccosum* 4 (5.2%). Infection was more common in manual labourers and males.

Conclusion: They are very common infections and proper samples collection and processing are keys to diagnosis. Further epidemiological studies of dermatophytic fungus-induced dermatophytosis are needed.

KEYWORDS : dermatophytes, *Trichophyton*, SDA

INTRODUCTION

Dermatophytosis is a major public health problem worldwide today affecting millions of people. The estimated lifetime risk of acquiring a dermatophyte infection is between 10- 20 percent. {1} Superficial infections caused by a dermatophyte is known as dermatophytosis or ringworm. Dermatophytic infection of skin is often called as "ringworm". This term is a misnomer because worms are not involved [2]. Dermatophytes are the most common cutaneous fungal infections seen in humans affecting skin, hairs and nails with a considerable morbidity. "Tinea", the Latin name for worm, describes the serpentine appearance of the skin lesions. The classical presentation of tinea infection is a lesion with central clearing which is surrounded by an advancing red, scaly, elevated border (3). These infections generally remain limited to non-living superficial keratinized layers and rarely do they proceed into deeper layers or cause invasive infections (4). The etiologic agents of dermatophytosis are classified into three genera based primarily on differences in microscopic morphology and modes of sporulation as *Epidermophyton*, *Mirosporium* and *Trichophyton* {5}. Species of the genus *Trichophyton* are capable of invading the hair, skin and nails; *Mirosporium* species involve only the hair and skin; and *Epidermophyton* species involves the skin and nails.

The trend of living in communities, contact with animals, the use of antibiotics, corticosteroids and antineoplastic drugs are some of the factors that contribute to the increase in the risk of infection by fungi especially dermatophytes (6). These infections are especially common in tropical countries like India due to environmental factors like heat and humidity. In addition, the risk factors include socio-economic conditions like overcrowding, poverty and neglect of personal hygiene (7). Bihar is a socially, economically and educationally backward area of India. The climate in Bihar also remains hot and humid for major part of year which are favourable for growth of the dermatophytes. High environmental moisture content facilitates fungal growth resulting in a high incidence of fungal diseases in this area.

MATERIAL AND METHODS

A total of one hundred ten (110) clinically diagnosed cases of skin, hair and nail infection, of all age groups and of both sexes, attending Skin and Venereology outpatient department of JLNMC hospital bhagalpur (Bihar) were selected for the study over a period of 18 months; from October 2015 to March 2017

A detailed history of selected cases was taken regarding name, age, occupation, pets and address. After the detailed history, clinical examination of patients was made in well-lighted room, which included: The sites of lesion, Number of lesions and types, presence of inflammatory margins etc. Specimen was obtained when the patient had been off both topical and systemic antifungal drugs for two to four weeks under aseptic precautions.

The specimens for the study were skin scrapings, hair pluckings and nail clippings. All specimens were obtained from the active edge of the lesion after thorough cleaning with 70% alcohol to remove surface bacterial contamination (8).

Nail clippings: Samples were taken from the nail plate, nail bed and subungual region of the nails with sterile scalpel.

Skin: The skin scrapings were taken from the active margins of the lesions, and scraped from centre to edge, using blunt margin of a sterile scalpel blade. Suppurative lesions were sampled with a swab. The materials were then sent to the laboratory in sterile petridishes [9].

Hairs: The infected hairs were removed by plucking with the roots intact using epilating forceps, scales were scraped off from the advancing border of the lesions. Skin scrapings were also collected from sites where fungal infection of hairs is suspected.

Specimens were allowed to dry to avoid multiplication of bacteria and fungal spores. All the collected samples were then divided into two parts: one for (i) direct microscopy and the other for (ii) culture.

DIRECT MICROSCOPY: Potassium hydroxide (KOH) preparation:

A small portion of skin scrapings was taken on a clean glass slide. Then 2-3 drops of 10% KOH were added to it and clean cover slip was placed on it. Preparation was kept at room temperature for 30 minutes. Slides were observed first under 10X objective and then 40X power immediately for the presence of typical fungal elements such as branching or unbranching hyaline septate hyphae. In case of nail sample, the material was kept overnight in 40% KOH and then teasing was done. Hair was cut into 1 cm size and transferred on the glass slide. Hair was also examined for ectothrix and endothrix infections

CULTURE METHODS: Sabouraud's dextrose agar medium with antibiotics:

Each sample was inoculated in tube of Sabouraud's Dextrose Agar with chloramphenicol (0.05%) and cycloheximide (0.5%) and incubated at 30°C in a BOD incubator for 4 weeks. Another part of the sample was inoculated in the Dermatophyte test medium (DTM) and incubated at 25°C. The culture tubes were examined after every two days, for a period of 4 weeks for the presence of growth. The growth was relatively slow and usually observed after 6 days. Culture was reported as negative only after about 4 weeks of incubation. In DTM, growth of dermatophytes was associated with change of colour of medium to deep red within 3-6 days. Sample was declared as negative, if no change was seen upto 2 weeks.

Fungal isolate was identified based on: a) Colonial morphology on the culture medium and pigmentation b) Growth rate c) Microscopic

morphology in LCB stain d) Slide culture e) Urease test. Slide culture (Riddell's Method) was done for all the samples.

RESULTS :

Out of the 110 clinically suspected cases attending outpatient department of skin and venerology JLNMCB Bhagalpur 77 cases turned out to be dermatophytoses, which showed growth of different dermatophytes on culture. Remaining 33 were either contaminants, or due to fungi other than dermatophytes or did not show any positive finding either in KOH preparation or culture. Out of total 110 cases, 64 were males and 46 were females. Males outnumbered females with a ratio of 1.39:1.

The most common age group to be affected was 21-30 years 35 (31.8%) followed by 31-40 years 25 (22.7%). Least common age group affected was >60 years 2 (1.8%).

Amongst the various clinical patterns, Tinea corporis was the commonest 40 (36.4%) type followed by Tinea cruris 30 (27.3%), Tinea pedis 13 (11.8%), and Tinea unguium 2 (1.8%). Males 25 (62.5%) were more commonly affected with T. corporis than females 15 (37.5%) Males are also commonly affected with T. cruris, T. pedis and T. faciei 24 (80%) 8 (61.5%) 5 (62.5%) respectively.

In this study dermatophytosis was most commonly seen in the low income group with 70 cases (63.6%) which was followed by middle income group with 27 cases (24.6%) and high income group with 13 cases (11.8%)

Direct microscopy by KOH mount revealed fungal elements in 82 cases (74.5%). Of these 66 (60%) were culture positive. Out of the 77 culture positive, 11 cases (10%) were negative on microscopy. Thus out of 110 samples studied 33 (30%) did not show evidence of the fungi on culture and 17 cases (15.5%) were negative both by microscopy and culture. Sensitivity of the KOH mount technique was 85.71%, but the specificity was 51.51%.

Two media were used for the culture of the samples DTM and SDA DTM was found as efficient as SDA in primary isolation of dermatophytes. All the isolates were isolated on SDA while 94.8% were isolated on DTM.

Table 1: Distribution of clinical types

Clinical types of dermatophytosis	Cases	Percentage
Tinea corporis	40	36.4
Tinea cruris	30	27.3
Tinea pedis	13	11.8
Tinea faciei	8	7.3
Tinea corporis with Tinea cruris	6	5.5
Tinea manuum	5	4.5
Tinea capitis	4	3.6
Tinea unguum	2	1.8
Tinea barbae	2	1.8
Total	110	100

Table 2 : Distribution of different species of dermatophytes according to different clinical findings

Clinical Types	Dermatophyte Isolates, No. (%)					Total
	T. mentagrophytes	T. rubrum	T. tonsurans	M. gypseum	E. floccosum	
Tinea corporis	8(27.6%)	17(58.6%)	1(3.4%)	1(3.4%)	2(7%)	29(72.5%)
Tinea cruris	4(21.1%)	13(68.4%)	-	-	2(10.5%)	19(63.3%)
Tinea pedis	5(55.6%)	4(44.4%)	-	-	-	9(69.2%)
Tinea faciei	-	5(100%)	-	-	-	5(62.5%)
Tinea corporis with cruris	2(40%)	3(60%)	-	-	-	5(83.33%)
Tinea manuum	-	4(100%)	-	-	-	4(80%)
Tinea capitis	-	2(66.7%)	1(33.3%)	-	-	3(75%)

Tinea unguium	1(50%)	1(50%)	-	-	-	2(100%)
Tinea barbae	1(100%)	-	-	-	-	1(50%)
Total	21(27.3%)	49(63.6%)	2(2.6%)	1(1.3%)	4(5.2%)	77(70%)

Table :3 Study of clinical types in relation to occupation:

Clinical types	Manual Workers	Household workers	Students	Professionals	Others	Total
Tinea corporis	18(45%)	10(25%)	5(12.5%)	4(10%)	3(7.5%)	40(36.4%)
Tinea cruris	15(50%)	3(10%)	7(23.3%)	3(10%)	2(6.7%)	30(27.3%)
Tinea pedis	5(38.4%)	4(30.8%)	2(15.4%)	-	2(15.4%)	13(11.8%)
Tinea capitis	-	-	3(66.7%)	-	1(33.3%)	4(3.6%)
Tinea faciei	3(37.5%)	2(25%)	2(25%)	-	1(12.5%)	8(7.3%)
Tinea unguium	2(100%)	-	-	-	-	2(1.8%)
Tinea manuum	4(80%)	1(20%)	-	-	-	5(4.5%)
Tinea corporis with cruris	2(33.3%)	1(16.7%)	2(33.3%)	-	1(16.7%)	6(5.5%)
Tinea barbae	2(100%)	-	-	-	-	2(1.8%)
Total	51(46.4%)	21(19.1%)	21(19.1%)	7(6.4%)	10(9%)	110(100%)

DISCUSSION

In this study, T. corporis was the commonest type of dermatophytosis (36.4%). This finding is comparable with other studies done by Ellabib et al (45.9%) [10], Singh et al [11], and Jain et al. (37%) [12]. Tinea cruris was found to be the second most common clinical type (27.3%). This finding was comparable with studies done by Mishra M [13], Sen SS [14] and Peerapur BV [15]. T. pedis was seen in 11.8% cases. This finding was comparable with Chimelli PAV (9.9%) [16], Ellabib MS (8.1%) [10], Singh S (11.53%) [11] and Huda MM (7%) [17]. T. faciei was seen in 8% cases. T. manuum was seen in 4.5% cases, and is comparable with studies done by Huda MM, Chimelli PAS and who reported tinea manuum in 3%, 1.9% and 1.53% cases respectively [17,16].

In this study most common age group to be affected was 21-30 years 35 (31.8%) followed by 31-40 years 25 (22.7%). These observations are similar to findings of other authors like SS Sen et al (14), SS Singh et al (11) and BV Peerapur et al.[15]. The higher incidence in young males could be due to greater physical activity and increased sweating.

Direct microscopy revealed fungal elements in 82 cases (74.5%). Of these 66 (60%) were culture positive. These results are mostly comparable with the results of Sen SS et al [14], which showed- Forty nine (49%) cases were positive for fungal elements by direct microscopical examination

Males were more commonly affected with T. corporis (62.5%) and T. cruris (80%) than females. This was comparable with that of other studies done by Ellabib et al, Singh et al, Sen et al., S Mishra et al and and Peerapur et al, [10,11,13,14,15].

In this study, dermatophytosis was most common in the low income group with 70 cases (63.6%). This observation is almost similar to other observations by Ranganathan S et al (18) and Sivakumar N et al (19) who reported 69.2% and 74.7% of affected people are from low income group respectively. The reason behind this may be due to poor living conditions, large family size and close contact, either directly or by sharing facilities, including combs and towels between family members in low socioeconomic group.

In the present study, dermatophytosis was most commonly seen in

manual workers with 51 cases (46.4%), which included agricultural workers and manual labourers, followed by household workers the above findings are comparable with other observations (20).

In our study KOH mount revealed 82 (74.5%) cases and culture revealed fungal elements in 77 cases (70%). These findings are comparable with other studies done by Karmakar et al. [21], Bindu et al. [22].

T. rubrum was the commonest etiological agent in majority of clinical types with 49 cases (63.6%) followed by *T. mentagrophytes* with 21 cases (27.3%) and *T. tonsurans* 2 (2.6%) which is comparable to other studies done by Bindu et al. [22] *T. rubrum* was the commonest species isolated (66.2%) followed by *T. mentagrophytes* (25%). Singh et al. [11] who reported *Trichophyton rubrum* (73.27%) was the most common isolate, followed by *Trichophyton mentagrophytes* (17.24%) and Jain et al. [12] *T. rubrum* (45.71%) *T. mentagrophytes* (14.29%), followed by *T. violaceum* (10%), *T. tonsurans* (8.57%).

CONCLUSION :

This study gives an insight about the etiological agents of dermatophytosis in this area having rural population and lack of knowledge about the disease. As far as concerned commonest lesion species isolated and other variables it is similar to other parts of India. There is need for public education campaigns and socio-economic interventions at the community level, to overcome the risk factors like overcrowding and lack of personal hygiene. Although this infection responds to conventional antifungals, dermatophytosis has a tendency to recur at the same or different site. Hence a correct diagnosis is important to initiate an appropriate treatment and also for epidemiological purposes.

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