Clinico-mycological study of dermatophytosis in a tertiary-care hospital in Ghaziabad

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Abstract
Introduction: Dermatophytosis is a superficial infection, which varies according to the geographic region wise, socio-economic level of the population, time of study, climatic variations, presence of domestic animals and age.
Aim: To determine the clinical profile of dermatophytic infections and causative fungal species in the various clinical presentations.
Materials and Methods: A total of 105 samples of clinically suspected dermatophytosis were collected. Direct microscopy by Potassium Hydroxide (KOH) and culture on Sabouraud's Dextrose Agar (SDA) and Dermatophyte Test Medium (DTM) was done. Statistical analysis was done using SPSS 19.0 software.
Results: Out of 105 clinically suspected cases of dermatophytosis, Tinea corporis 47 (44.8%) was the commonest clinical type followed by tinea cruris 19 (18.09%), Commonest age group affected was 31-40 years (27.6%) with male to female ratio of 2.38:1. Of the dermatophytes isolated, T. rubrum 19 (51.35%) was the most common followed by T. mentagrophyte 16 (43.24%) and E. floccosum 2 (5.4%).
Conclusions: The major types of tinea were tinea corporis and tinea cruris. Among dermatophytosis, T. rubrum was the predominant etiological agent present and DTM being a good screening medium in the laboratory diagnosis of dermatophytosis when compared to SDA with antibiotics.

Keywords: Dermatophyte test medium, Dermatophytosis, Sabouraud dextrose agar, Tinea.

Introduction
Dermatophytosis is a superficial infection caused by a group of fungi, dermatophytes consisting of three major genera, Trichophyton, Microsporum and Epidermophyton of the class hyphomycetes and division deuteromycota. They have the ability to invade keratinized tissue (skin, hair and nails) but are usually restricted to the nonliving cornified layer of the epidermis because of their inability to penetrate viable tissue of an immunocompetent host.[1] The socio-demographic factors like poverty, overcrowding, poor personal hygiene are also prevalent in India. Dermatophytes are by far the most significant cutaneous fungi because of their widespread involvement of population at large and their worldwide prevalence.[2,3] The rising prevalence of dermatophytosis has been attributed to many factors including tropical climate, overcrowding, urbanization, shared accommodation such as living in hostels, the use of occlusive footwear, tight-fitting clothes, community showers and sports activity. There is a changing trend in the dermatophytic infections in that the cases are presenting as chronic, treatment unresponsive and recurrent cases.[2-4]

Although dermatophyte infection is non-invasive and easy to cure, its widespread nature, social embarrassment, impairment of quality of life and damage to the economic status due to the cost of the treatment are major public concerns. The present study was conducted to know the prevalence, etiology and common clinical profile and species causing dermatophytosis and the laboratory evaluation of SDA with DTM for diagnosing dermatophytosis.

Materials and Methods
A cross sectional study was carried out in Santosh Medical College and Hospital from March 2018 to February 2019 after obtaining the ethical clearance by Institutional Ethical Committee. A total of 105 samples were collected from clinically suspected cases having dermatophytosis. A detailed history regarding age, sex, occupation, social status, duration of complaint and clinical history were taken. Samples were collected after cleaning the affected surface with 70% alcohol. From skin lesions, scales were collected from erythematous growing margins of the lesion with a sterile blunt scalpel. For hair, plucking with epilating forceps along with the base of the hair shaft around the follicle was done. For nail, clippings of the infected part and scrapings beneath the nail were taken. Samples were collected in sterilized Whatman filter paper, envelope and transported to the microbiological laboratory as fungal spores resist drying and remain viable for several weeks when stored in the paper.

Specimens collected were subjected to potassium-hydroxide (KOH) wet preparation of various concentrations (10%, 20% and 40%) depending on the type of clinical specimen for the presence of fungal elements. Following direct microscopic examination, irrespective of demonstration of fungal elements, the specimen was inoculated onto Sabouraud's Dextrose Agar SDA with 0.1% gentamicin and the other to Dermatophyte Test Medium DTM (Hi-Media). Samples were inoculated in two sets of these culture media. One set was incubated at 37°C and another set at 25°C in BOD incubator. SDA with 0.1% gentamicin was incubated at 28°C for up to four weeks and was observed periodically for growth. If no growth was found after four weeks, it was taken as negative for the growth of fungi.[2,5]

Dermatophyte Test Medium (DTM) was incubated at 28°C for up to ten days and was observed for color change. Fungal isolate was identified based on colony morphology, pigmentation, growth rate, microscopy with Lactophenol Cotton Blue (LPCB) and slide culture. Urease test and in-

vitro hair perforation tests were also performed to differentiate Trichophyton rubrum and Trichophyton mentagrophytes when there was difficulty in identification by microscopic and macroscopic examination.

Data collection and statistical analysis was done using SPSS software version 20.0 (SPSS Inc., Chicago, IL, USA). $P = \leq 0.05$ was considered statistically significant.

Results

In the present study, 105 cases of clinically diagnosed dermatophytosis were studied. Mean age was 35.64 years with a range of 6-78 years. Most common age group affected was 31-40 years with 29 cases (27.61%) followed by 21-30 years with 26 cases (24.76%) and 41-50 years with 18 cases (17.14%). Least common age group affected was >70 years with 1 case (0.95%) followed by 0-10 years with 5 cases (4.76%). Males were more commonly affected with 74 cases (70.48%) than females with 31 cases (29.52%). Male to female ratio was 2.38:1. Distribution of clinical types is given in Table 1. Fig. 1 shows KOH preparation of Skin Scraping with Fungal Elements.

Majority of cases were from low-income group with 54 cases (51.43%) followed by middle-income group with 34 cases (32.38%) and high-income group with 17 cases (16.19%). Of the 105 clinically suspected cases of dermatophytosis, fungi were demonstrated in 74 cases (70.48%) either by direct microscopy and /or culture. [Table 2]

Culture positive samples were 37 (100%) on DTM, while on the other side SDA positive cultures were only 32 (86.49%). Dermatophytic species isolated were T. rubrum (58.06%) [Fig. 2], T. mentagrophytes (22.58%) [Fig. 3], and Epidermophyton floccosum (6.45%) [Fig. 4]. In the present study, species level identification was possible for 70.47% of isolates on primary isolation on SDA with gentamicin. All positive cultures in DTM also showed growth on SDA with gentamicin on primary isolation. All 37 (100%) isolates showed growth on DTM within 10 days of inoculation in comparison to SDA with gentamicin showing growth within 10 days in only 25 (78.12%) isolates.

Table 1: Sex wise distribution in relation to clinical types of dermatophytosis

S. No.	Clinical Types	No. of cases		Total
		Males	Females	
1	Tinea corporis	32	15	47
2	Tinea cruris	18	1	19
3	Tinea unguim	11	5	16
4	Tinea capitis	4	4	8
5	Tinea pedis	1	3	4
6	Tinea facei	2	1	3
7	Tinea manuum	1	1	2
8	Tinea corporis with Tinea cruris	5	1	6
Total		64	31	95

Table 2: KOH wet preparation (Microscopy) and culture (SDA with gentamicin and /or DTM) findings

S. No.	KOH and Culture Findings	No. of cases
1	Total KOH and/or culture positive	74
2	KOH-ve and Culture-ve	31
3	KOH-ve and Culture+ve	2
4	KOH+ and Culture-ve	37
5	KOH+ve and Culture+ve	35

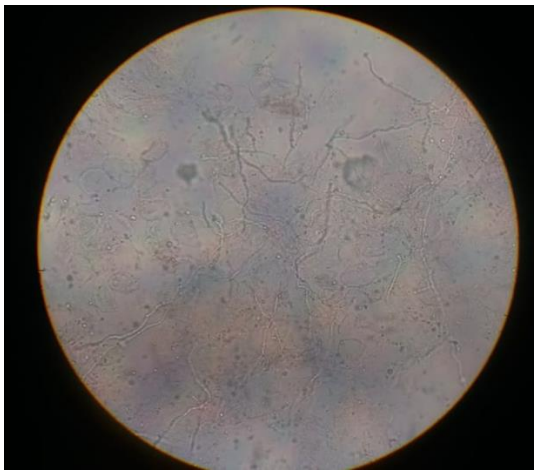

Fig. 1: KOH preparation of skin scraping showing fungal elements

Discussion

In the present study, dermatophytosis was observed maximum in the age group of 21–40 years and in males. This may be due to increased outdoor exposure, greater physical activity and increased sweating in this age group favoring the growth of dermatophytes. This was in correlation with other studies.[6,7] Increased percentage of males may be due to the fact of increased outdoor exposure and more physical work that results in increased sweating and less cosmetic consciousness compared to females.[8,9]

Tinea corporis followed by Tinea cruris were common clinical presentations of dermatophytosis in the present study which was in correlation with other studies from India.[3,6-8] Tinea capitis was more common in children below the age group of 10 years, which was also observed in some studies.[9,10] T. rubrum was the most common dermatophyte to cause all clinical types of dermatophytosis followed by T. mentagrophytes. This was in correlation with other studies.[7,8] Many other species of dermatophytes like T. ferrugineum, T. concentricum and M. audouinii, T. schoenleinii, T. tonsurans, T. verrucosum have been isolated by other workers, but we could isolate only T. rubrum, T. mentagrophytes, and E. floccosum. Chronic nature of infection and adaptation to human is mainly responsible for the predominance of T. rubrum to cause dermatophytosis.[11,12] Microsporum species were not isolated in the present study.

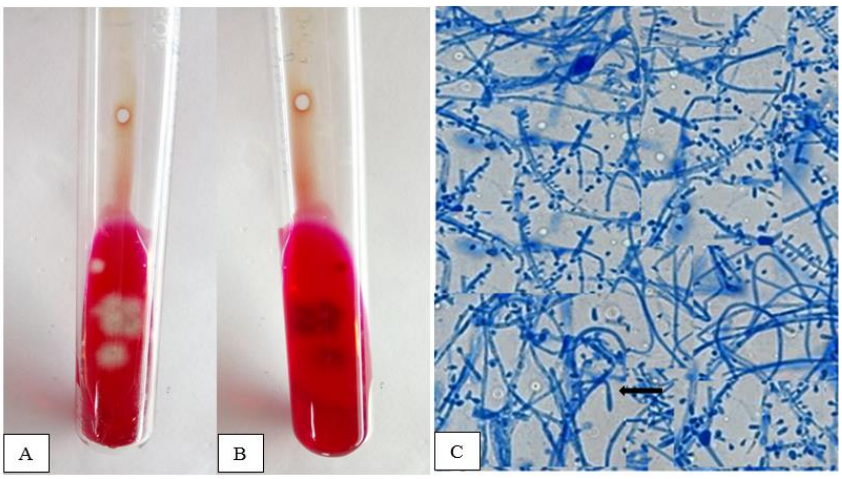

Fig. 2: Trichophyton rubrum. **(A):** Growth on DTM (obverse). **(B):** Growth on DTM (reverse no pigmentation). (C) Lacto phenol cotton blue mount showing tear drop shaped microconidia and pencil shaped macroconidia (×400)

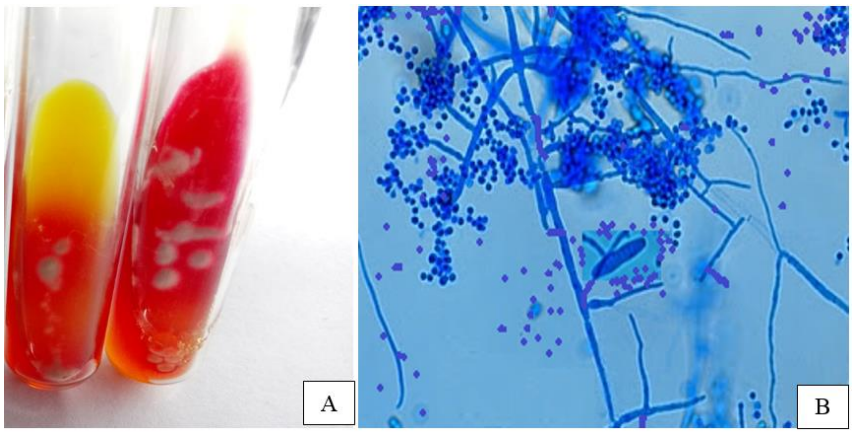

Fig. 3: Trichophyton mentagrophytes. **(A):** Growth on DTM **(B):** Lactophenol cotton blue mount showing clusters of microconidia with a single cigar shaped macroconidia in center (×400)

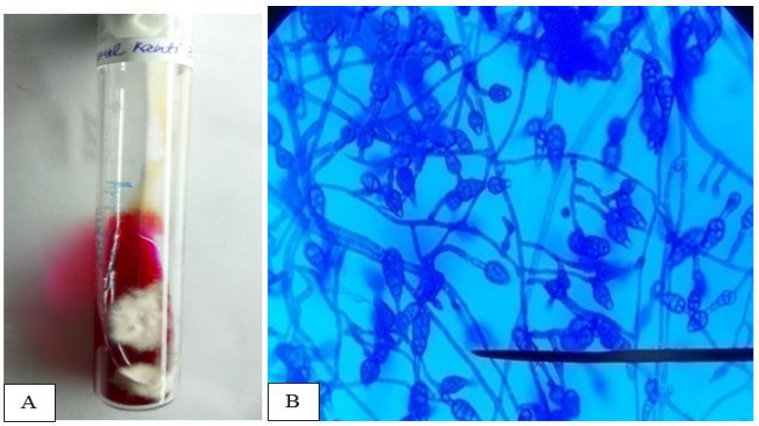

Fig. 4: Epidermophyton floccosum. **(A):** Growth on DTM (obverse); **(B):** Lactophenol cotton blue mount showing club shaped macroconidia (×400)

In the present study, KOH positivity was 68.57% and culture positivity was 35.23%. This difference is statistically significant ($P < 0.001$), this shows that direct microscopy by KOH mount is a good screening test in the laboratory diagnosis of dermatophytosis.

Present studied showed that dermatophytes grow earlier on DTM compared to SDA with gentamicin although efficacy of SDA with gentamicin and DTM in isolation of dermatophytes is equal as seen in a previous study.[4] In the present study even though all 37 culture positive samples yielded growth on DTM on primary isolation, the appearance

of growth was earlier on DTM that is, within 10 days (100%) compared to SDA (78.12%).

For dermatophytes classification up to species level, SDA with antibiotics is preferable compared to DTM. In the present study, species level identification was possible for 74.32% of isolates on primary isolation on SDA with gentamicin which is not possible with the growth on DTM on primary isolation, as conidial production was low on DTM, which is required for identification. Moreover, on DTM pigment production cannot be observed. For identification of growth on DTM, always it was required to subculture further on to SDA with gentamicin.

Conclusion

This research concluded that the major types of tinea were tinea corporis and tinea cruis. DTM is a good screening medium in laboratory diagnosis of dermatophytosis compared to SDA with gentamicin. T. rubrum and T. mentagrophyte were found to be the main causative agent of tinea corporis and tinea cruis.

Conflict of Interest: None.

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