A Study on Mycological Profile of Superficial Mycoses in a Rural Tertiary Care Hospital of Eastern India

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Abstract

Background: Superficial mycosis is a very common fungal infection worldwide of the skin, hair and nails by dermatophytes, non dermatophytes and yeasts. Though they do not cause mortality, but are important agents causing morbidity, cosmetic disfigurement and recurrence leading to a major public health problem. Aim: Determine the clinico-mycological correlation and prevalence of dermatophytosis. Material and Methods: A six months observational cross sectional study was conducted from July 2019-December 2019 on 156 clinically diagnosed cases of superficial mycosis with no systemic complaints. Specimens were examined macroscopically as well as microscopically by KOH and culture. Result: Dermatophytosis was more in the males <40 years. KOH positivity was 100% and dermatophytes were the commonest superficial fungal infection 101/156 (64.74%), followed by Candidiasis 36/156 (23.08%). Non dermatophyte moulds in 06/156 (3.85%) and Malassezia furfur in 13/156 (8.33%) cases. Trichophyton was the commonest dermatophyte isolated (52.47%). Conclusions: Non-dermatophytic fungi are emerging as an important cause of superficial mycoses.

Keywords: Dermatophytes, Non-dermatophyte Moulds, Superficial Mycosis, Trichophyton

1. Introduction

Fungal infections of the skin and its appendages caused by Dermatophytes, Pityriasis versicolor and Candidiasis are called superficial mycosis. Dermatophyte infections are very common worldwide¹. They produce the enzyme keratinase, which helps them to metabolize human keratin present in the skin, nails and hair producing dermal inflammation, profuse itching and disfigurement²,³. Dermatophytosis is common in the tropics because of high humidity and heat allows dermatophytes to grow profusely. The prevalence of dermatophytosis varies in different geographical locations and the chance of developing a dermatophyte infection is between 10 and 20%⁴,⁵. Dermatophytosis, commonly referred to as ringworm/tinea are hyaline septate fungal moulds having more than 100 species, 40% of which causes human infections⁶. They are divided depending on their natural habits and host preferences into three groups—anthropophilic, zoophilic and geophilic⁷. Emmon classified the dermatophytes according to their conidial morphology into three genera—Trichophyton, Microsporum and Epidermophyton⁸. Clinically, tinea is classified according to the site of involvement into tinea capitis, tinea corporis, tinea cruris, tinea pedis, and tinea barbae⁹. Nail plate infection by a dermatophyte is called tinea unguium while infection due to non-dermatophytes is called onychomycosis¹⁰. Infection by Candida spp. is called Candidiasis and causes infections of the superficial
skin, the mucosa and the deeper tissues which may lead to life-threatening conditions\textsuperscript{11}. Other fungi commonly causing superficial mycosis is \textit{Malassezia furfur}, a lipophilic fungus that affects the skin and hair causing, recurrent, superficial fungal infection of stratum corneum like dandruff, pityriasis versicolor (tinea versicolor), tinea cincinata and seborrhoic dermatitis\textsuperscript{12,13}. A large subcontinent like India with varied topography and climate leads to growth of various different fungi\textsuperscript{14}. All over Asia the epidemiological trend indicates that the commonest dermatophytes involved are the \textit{Trichophyton} species\textsuperscript{15}. Recently fungal infections are rising probably because of indiscriminate use of antibiotics, immune deficient diseases like Human Immunodeficiency Virus/ Acquired Immunodeficiency Syndrome (HIV/AIDS) and anticancer therapy\textsuperscript{16}. The present study was undertaken to find out the clinico-mycological profile of superficial fungal infections amongst patients attending a rural tertiary care hospital in Eastern India and to identify the common fungal pathogens responsible for it.

2. Materials & Methods

Six months cross sectional study was conducted in the Department of Microbiology, Burdwan Medical College & Hospital from July 2019–December 2019. Suspected cases of superficial mycosis attending the (OPD) Out Patients Department of Dermatology & Venereology were clinically examined and 156 patients willing to participate among them were selected for study purpose and were asked to report to the Department of Microbiology for fungal examination. Unwilling patients and those previously treated for the same infection were excluded from the study. Institutional ethics committee permission was taken beforehand.

Complete history taken and clinical examination was done once again in each patient; skin scrapings nail clippings and hair samples were collected after cleaning the area with 70% alcohol. The skin samples were collected from the edges of lesion; hairs were plucked from the infected areas & nail samples were collected from the infected nail area and nail plates. The materials collected were divided into two parts—one for KOH mount and another for inoculation into culture media. Skin scrapings were examined in 10% KOH solution while the hair samples were kept in 40% KOH solution for one hour before the nail scrapings were examined in 40% KOH after 3–4 hours. The other part of the sample was inoculated on culture media—Sabouraud Dextrose Agar with chloramphenicol (0.05%) and cycloheximide (0.5%) (SDCCA) with or without olive oil overlay. Cycloheximide selectively inhibits many fungi like \textit{Aspergillus} and \textit{Mucor} species while helping the dermatophytes to grow\textsuperscript{17}. All the inoculated culture media were incubated at 25°C and 37°C respectively for 4 weeks. The culture tubes were examined daily for one week; later twice a week for 4 weeks before declaring them as negative. Tubes showing growths were examined under Lacto Phenol Cotton Blue (LPCB) mount. The detailed colony characteristics—surface, texture, pigmentation and colour on the obverse and reverse were studied. Slide cultures were done to study the arrangement of macroconidia and microconidia to classify them into the 3 genera.

Chlamydospores production on corn meal agar and germ tube formation differentiated the \textit{Candida} species into the \textit{albicans} and \textit{non-albicans} group\textsuperscript{17}. Suspected Pityrosporum versicolor scrapings were examined only in KOH mount and not cultured. No further biochemical test was done because differentiation test media for yeasts and dermatophytes was not available in our laboratory. Cultures were repeated in culture negative cases for dermatophytes but positive for Non-Dermatophyte Moulds (NDM). For the diagnosis of NDM causing infection of the nails, the criteria of Walshe and English was followed in the study\textsuperscript{18}.

3. Results

Clinically, 156 suspected cases of superficial mycosis were included in this study, 93(59.62%) cases were male and 63(40.38%) were female; none of them had any systemic disease. The age group most commonly affected was 21–40 years (48 males and 32 females) followed by <20 yrs age group (24 males and 17 females) (Table 1).

Mycological analysis of the samples from 156 cases showed fungal elements in KOH mount in all the cases (100%), among which 101(64.74%) were dermatophytes, 6 (3.85%) were NDM, 36(23.08%) were Candida and 13 (8.33%) cases showed \textit{Malassezia furfur} (Table 2) (Figure 1).

Among the isolated organisms, 71/101(70.30%) dermatophytes were culture positive; 06/06 (100%) were culture positive NDM. \textit{Candida} were culture positive in all the cases 36/36 (100%) while \textit{Malassezia} was isolated in culture from 05/13(38.46%) cases only (Table 2) (Figure 1).
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Table 1. Age & sex wise distribution of superficial mycosis

| Age (Yrs) | Male (%) | Female (%) |
|-----------|----------|------------|
| <20 yrs   | 24 (15.4) | 17 (10.9)  |
| 21-40 yrs | 48 (30.8) | 32 (20.5)  |
| 41-60 yrs | 17 (10.9) | 11 (7.1)   |
| >60 yrs   | 4 (2.5)   | 3 (1.9)    |
| TOTAL     | 93 (59.6) | 63 (40.4)  |

According to anatomical site of involvement, Tinea corporis was the commonest site of involvement 29.70% (30/101) closely followed by Tinea cruris 24.75% (25/101). Tinea pedis was found in 14/101 (13.86%) cases only. Tinea capitis 10/101 (9.91%) was the least found clinical type of dermatophytosis. Total number of Onychomycosis cases were13; of these 07 (6.93%) were of Dermatophytic origin (Table 3) (Figure 2).

The commonest dermatophyte isolated was Trichophyton 53 (52.4%) cases while the least common was Microsporum 4(3.96%) (Table 3).

Table 2. Number of isolates showing KOH and culture positivity

| Isolates          | Number (%) | KOH (+) (%) | Culture (+) (%) |
|-------------------|------------|-------------|-----------------|
| Dermatophytes     | 101 (64.7) | 101 (100)   | 71 (70.3)       |
| Non-Dermatophytes | 06 (3.9)   | 06 (100)    | 06 (100)        |
| Candida           | 36 (23.1)  | 36 (100)    | 36 (100)        |
| Malassezia furfur | 13 (8.3)   | 13 (100)    | 05 (38.5)       |
| Total             | 156 (100)  | 156 (100)   | 118 (75.6)      |

Table 3. Distribution of different dermatophytes

| Fungal Infection | Tricho-phyton (%) | Epidermo-phyton (%) | Micro-sporum (%) | Number (%) |
|------------------|-------------------|---------------------|-----------------|------------|
| ♦ Tinea Corporis | 18 (60.0)         | 12 (40.0)           | 0               | 30 (100)   |
| ♦ Tinea Capitis  | 06 (60.0)         | 00                  | 04 (40.0)       | 10 (100)   |
| ♦ Tinea Cruris   | 11 (44.0)         | 14 (56.0)           | 0               | 25 (100)   |
| ♦ Tinea Pedis    | 08 (57.1)         | 06 (42.9)           | 0               | 14 (100)   |
| ♦ Tinea Mannum   | 06 (40.0)         | 09 (60.0)           | 0               | 15 (100)   |
| ♦ Onychomycosis  | 04 (57.1)         | 03 (42.9)           | 0               | 07 (100)   |
| Total            | 53 (52.5)         | 44 (43.6)           | 04 (3.9)        | 101 (100)  |

Onychomycosis cases were mainly due to Trichophyton (04) followed by Epidermophyton (03)–6.93%. All the non dermatophytic moulds were isolated from cases of Onychomycosis which included strains of Aspergillus in 02/06 cases (33.33%), Fusarium in 03/06 cases (50%) and Curvularia 01/06 cases (16.67%) (Table 4).
4. Discussion

Fungal infections are very common in the tropics causing serious and fatal infections ranging from superficial skin infections to internal organ invasion and may spread to other individuals also. They affect the quality of life of a patient and sometimes may even be life threatening. These infections are easily diagnosed and treated. In our study we found a high male prevalence (93/101) 59.62% of dermatomycoses similar to the study conducted by Sharath et al. Males were more affected worldwide probably because of their increased sweating and outdoor activity. Patients of all ages and both sexes were affected but maximum cases were observed in the age group of 21-40 years (48 in males and 32 in females) followed by <20 years age group (24 in males and 17 in females). Our findings correlated with that of Malik A et al. in 2014 who also reported 'higher frequency in age group of 11-40 years, predominantly in the physically active males'.

Fungus isolated rate was more by KOH mount (100%, 156/156) than by culture (74.64%, 118/156), in comparison to 7% to 49% by culture in other studies indicating that both direct microscopy and culture are required for the definitive diagnosis of fungal infection. Our study corroborated with that of Sharath et al. who found that all were positive for KOH (100%), but culture positivity was shown only in 72.9% of cases. In our study the most common clinical presentation was dermatophytosis (64.74%), followed by candidiasis (23.08%), Malassezia furfur or Pityriasis versicolor (8.33%) and the least common was onyehymycosis due to Nondermatophytes (3.85%). Our finding corroborated with that of Sharath et al. who found that the commonest clinical presentation was dermatophytosis (38.55%), followed by Pityriasis versicolor (31.3%), by candidiasis (18.75%) and the least common was onychomycosis (11.4%). It also correlated with findings of P Kannan et al.

In our study, among dermatophytosis, Tinea corporis (29.70%) was the commonest presentation, followed by Tinea cruris (24.75%) and the least common was Tinea capitis (9.91%). This is similar to the study conducted by Patel et al. who observed that Tinea corporis (64%) was the most common fungal manifestations. Our findings also corroborated with Hazarika D et al. where the commonest clinical pattern of infection was Tinea corporis (21.5%).

Onychomycosis refers to fungal infection of the nails due to various etiological agents, viz. dermatophytes, yeasts and non-dermatophyte moulds (NDM). It accounts for up to 50% of nail disorders and 30% of all superficial fungal infections of the skin. This is similar to our observation where onychomycosis constituted 13(7+6), 7 cases due to dermatophytes and rest due to NDM. NDM species are increasingly being implicated in causing primary invasion of the nails in onychomycosis. Malik A et al. in 2014 reported that 15 out of 32 NDM (46.8%) was isolated from infected nails. The most common NDM sp. isolated was Fusarium in 03/06 cases (50%) followed by Aspergillus in 02/06 cases (33.33%) and Curvularia 01/06 cases (16.67%). This result was found to be comparable to the study conducted by Hazarika D et al. who reported that the commonest isolate was Fusarium (03/09, 33.33%) followed by Aspergillus 02/09 cases (22.22%).

5. Conclusion

In conclusion, it can be stated that Non-Dermatophytes (NDM), which were previously considered to be laboratory contaminants, are increasingly being reported as important etiological agents of superficial mycoses along with dermatophytes. But the role of the NDM in causing such infections is still not well established. A study of longer duration with a larger sample size is needed to establish their role. Good hygiene, sanitation and proper hand wash are important preventive measures for such infections.

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| Table 4. Distribution of Non-dermatophyte isolates |
|-----------------|---------|-----------|
| **Non dermatophytes** | **Number** | **Percentage** |
| Aspergillus | 02 | 33.33 |
| Fusarium | 03 | 50.00 |
| Curvularia | 01 | 16.67 |
| Total | 06 | 100 |

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8. Conflicts of Interest

There are no conflicts of interest.

9. References

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