Clinico-Mycological Study of Superficial Mycoses in a Tertiary Health Care Centre of Southern Assam

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ABSTRACT

Background - Superficial Mycosis, which is the most common fungal infection affecting human beings, includes Dermatophytosis and Dermatomycoses, which are the infections of superficial keratinized layer of skin, nail & hair by Dermatophytes and non Dermatophytic moulds or yeasts respectively. This clinical entity is very common in hot, humid tropical climate of India with prevalence ranging from 30-60% but its precise case magnitude and epidemiology in North eastern India cannot be stated as there are only few studies conducted. So, this study is undertaken to 1) Find the prevalence of Superficial Mycosis in a tertiary health care centre of Southern Assam. 2) Study the clinical profile of the cases 3) Isolate and identify the causative agents of Superficial mycosis.

Materials & Method - The study has been conducted on 250 samples from clinically suspected and untreated cases of superficial mycosis from Aug 2017 to Dec 2018. 2 separate sets of samples from edge of skin lesion/nail /hair were collected, of which 1 sample was subjected to direct microscopy with (10-40) % KOH and the other part was subjected to 2 sets of fungal culture in SDA tubes at 25°Cand 37°C & followed for 3 weeks. In Culture positive cases, fungal identification was based on colony morphology, pigment production & LPCB mount. For confirmation of isolates, Slide Culture and biochemical tests were done.

Result - Out of total 250 samples, 115 samples (46%) showed presence of fungal elements in KOH examination, of which 73 were culture positive and of the KOH negative samples 10 samples were culture positive, thus making the prevalence 33.2% (83/250). Clinically, Tinea corporis was the most common form of both superficial mycosis & Dermatophytosis and Pityriasis Versicolor has been found the most common dermatomycosis. Males(21-50yrs) were affected by superficial mycosis more than Females(16-30yr). Trichophyton mentagrophyte was the mostly isolated agent causing superficial mycosis.

Key Words - Superficial mycoses, prevalence, Assam, Slide Culture, Urease

INTRODUCTION

Superficial mycoses are fungal infections of skin, and its appendages (hairs and nails) caused by dermatophytes, yeasts and non-dermatophyte moulds [1]. Superficial fungal infections are the most commonly encountered fungal infections prevalent in the world. According to World Health Organization (WHO), the prevalence rate of superficial mycotic infection worldwide has been found to be 20-25%. [2][3] Its prevalence varies in different countries. [3][4][5]. Among these, dermatophytes are by far responsible for significant and largest number of cases due to their widespread involvement of population at large and their worldwide prevalence [1][6]. Dermatophytosis is the most important group of superficial fungal infections caused by dermatophytes, which is a group of closely related keratinophilic fungi that are capable of growing by invading the keratin of skin, hair, and nail for obtaining nutrients. [5] Dermatophytes are
divided into three genera: Trichophyton, Epidermophyton and Microsporum. In addition, dermatophytes can also be divided into anthropophilic, zoophilic, and geophilic species on the basis of their primary habitat associations. Species of all three groups can cause human infection. Dermatophytic infections include several distinct clinical manifestations named according to the anatomic locations involved. The severity of the disease relies on the specific strain of the infecting dermatophyte, the sensitivity of the host and the site of infection. About 20-25% of the world’s populations are infected with Dermatophytic fungi and the incidence is increasing on a steady basis. At any topographical location no human race is free from Dermatophytes. Species distribution and prevalence of Dermatophytes varies with the geographical area and the course of time and is governed by environmental conditions, personal hygiene and individual’s susceptibility. The variance in the distribution pattern of dermatophytosis is attributed to the social practices, migration of labour, movements of troops, immigration and frequent worldwide travelling. It is more prevalent in tropical and subtropical countries like India where the heat and humidity are high for most part of the year which makes dermatophytosis or ringworm a very common superficial fungal skin infection. The climate of North East India where the monsoons are heavy and relative temperature and humidity is high for most part of the year, retards sweat evaporation due to high environmental moisture content, thus facilitating fungal growth resulting in a high incidence of fungal diseases in this area. Over the past decades, non-dermatophytes, as agents of superficial fungal infection in humans, produce lesions that are clinically similar to those caused by Dermatophyte infections.

Although common, the precise magnitude of the problem defies measurement. Studies on Dermatophytosis in India have received increased attention in recent years because one-fifth of world’s population suffers from mycosis. Although numerous studies on the clinico-mycological aspects of dermatophytosis have been undertaken from different parts of India, very few studies have been reported on the etiological profile from the Northeast and more specifically no study has been conducted in Southern part of Assam focussed on Superficial mycoses.

**PURPOSE OF THIS STUDY**
1. To find the prevalence of Superficial mycoses in a tertiary health care centre of Southern Assam.
2. To assess the clinical profile of the Superficial mycoses cases in this region of India.
3. To isolate and identify the causative agents of superficial mycoses in Southern Assam.

**MATERIALS AND METHOD**
This hospital based cross-sectional study was carried out from August 2017 to December 2018 in the Mycology section of Department of Microbiology of the only Tertiary health care centre of Southern Assam on 250 clinically diagnosed untreated cases of superficial mycosis attending the Dermatology OPD of this tertiary care hospital after obtaining Ethical clearance from the Institutional Ethical Committee. Before sample collection, medical, hygiene, socio-economic and habitual profiles were noted in each case. After thorough cleaning of the affected site with 70% Ethyl alcohol, skin scales and crusts from the edges of inflamed lesion, nail clippings with subungual scrapings and epilated lustreless affected hair with roots were collected in a dark background paper envelope for easy visualisation and kept overnight for absorption of moisture. Skin and hair samples were subjected to direct microscopy with 10% KOH and nail clippings were subjected to overnight treatment with 40% KOH and then subjected to direct microscopy the next day. While one set of clinical samples was used for direct microscopy, 2 more sets were...
inoculated into 2 sets of Sabouraud’s Dextrose agar tubes, one with 0.05% Chloramphenicol and the other with 0.05% Chloramphenicol and 0.5% Cycloheximide and incubated at 25°C and 37°C and followed up biweekly for growth for at least 3 weeks before declaring them negative. Growth of Fungal isolates were noted for colony characteristics in the form of texture, surface, colour on obverse and reverse and diffusible pigment if any. Identification of fungal isolate was done in most cases by direct microscopic Lactophenol Cotton Blue examination of tease mount from SDA tube growths. In cases of disputed identification or apprehended possible contamination, Slide Culture was performed on blocks of Corn Meal Agar to note the exact pattern of Microconidia, Macroconidia, Sporulation, Special structures and Chlamydospores. Urease test and invitro hair perforation test was done to differentiate Trichophyton mentagrophyte and Trichophyton rubrum. In case of Cutaneous Candidiasis, Germ tube test and subculture on CHROM Agar was done to identify the species.

STATISTICAL METHOD- Statistical analysis was done using Chi-square test and significance level set at p value ≤ 0.05

RESULT

Out of total 250 samples, 115 samples (46%) showed presence of fungal elements in direct KOH examination, of which 73 were culture positive and of the 135 KOH negative samples 10 samples were culture positive. Thus, taking culture positivity as gold standard, the prevalence of superficial mycosis comes as 33.2% (83/250) in this study. After microbiological confirmation, Tinea corporis was found the most common form of both superficial mycosis & Dermatophytosis and Pityriasis Versicolor by Malassezia furfur was been found the most common Dermatomycosis. Out of the total laboratory confirmed cases of superficial mycosis, 86% were due to Dermatophytosis and 14% were due to Dermatophytosis. 70% of the affected cases were male and 30% were females. Occupation wise, unskilled labourers were mostly affected by superficial mycosis. Males in the age group of (21-50) years and females in the age group of (11-30) years were affected more by superficial mycosis. Trichophyton mentagrophyte was the mostly isolated agent causing superficial mycosis.

Fig-1: result of direct microscopic examination by Koh

Fig-2: Presence of fungal hyphae noted in KOH examination
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Fig-3: OUTCOME OF FUNGAL CULTURE

Fig-4: Depicts Age distribution of affected cases of Superficial mycosis

Fig-5: Distribution of confirmed cases of Superficial mycoses-83/250, affected males-58/83, affected females-25/83

Fig-6: Occupational Distribution of positive cases of Superficial Mycoses

- AFFECTED FEMALES 30%
- AFFECTED MALES 70%
- NON WORKING 16%
- SKILLED LABOURER 25%
- UNSKILLED LABOURER 44%
- STUDENTS 15%
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Fig 7: Depicts the seasonal variation of cases of Superficial mycosis.

Fig 8: Prevalence distribution of Various types of Dermatophytosis.

Fig 9: Distribution pattern of isolated dermatophytes
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Fig-10

Fig-11: Photographs of Clinical lesions from cases studied.
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Fig-12: Various Clinical presentations of Superficial mycoses

Fig-13: Tease mount examination with Lactophenol Cotton Blue.
DISCUSSION

This study in a tertiary care centre of Southern Assam found the prevalence of superficial mycoses to be 33.2%. Similar prevalence of 33% was also reported by Lavanya V and S.S Solabannavar [6] from Kerala. The prevalence of Superficial mycoses and dermatophytosis reported by clinic-epidemiological study by Smita Sarma and A K Borthakur [11] from Northern Assam was 61%. And that found by Sen S S and Rasul ES in Guwahati Assam was 51%. [12] Whereas 79% prevalence was found in a hospital-based study by Lt Col P Roy and Wing Commander Sanjiv Grover [9] from Northeast India which might be due to the wearing of uniforms and closed shoes for prolong periods by soldiers in all weather- a milieu conducive to fungal growth. While in another study conducted 10 years later in Army Base Hospital Tezpur Assam by A. Naglot et al [7] the prevalence of Dermatophytosis only was found to be 59.6%. A prevalence of 45.3% was noted in a hospital-based study in Dermatology department of Calicut Medical College by V Bindu and K Pavithran. [13] 74% prevalence was found by KAK Surendran et al [14] from Mysore. Similar prevalence of 72% was reported by B. Janardhan and G. Vani from Telangana. [15]

In concordance with the present study, Tinea corporis was also found to be the commonest presentation of Superficial mycosis by A Naglot et al [7] from Tezpur Assam, report of Sen SS and Rasul ES in Guwahati Assam [12], Rekha Sharma et from Sikkim. [16] Sumit Kumar et al [10], U S
Agarwal et al [17] and Sonia Mahajan et al [18] from various parts of North India have also found Tinea Corporis to be the most common presentation of superficial mycosis in their respective studies. This has been seconded by studies of Lavanya V and S S Solabannavar [6], A Lakshmanan et al [3], V Bindu and K Pavithran [13], KAK Surendran et al [14] and B Janardhan and G Vani [15] from South India. But Lt Col P Roy and Wg Cdr Sanjiv Grover [9] found Tinea Pedis to be the commonest presentation of Superficial mycoses in their study. This discordance might be due to their study population consisting of Soldiers only who use closed boots/shoes in all types of weather which promotes sweating and prevents sweat evaporation- a milieu conducive for fungal growth.

Trichophyton mentagrophyte was the mostly isolated causative agent from cases of superficial mycosis in this study. This finding has been seconded by studies carried out by Soniya Mahajan et al [18] in Uttar Pradesh, US Agarwal et al [17] in Northwest India and Sanjeev Sahai and Devesh Mishra [18] in Central India and Noronha et al [19] in South India. While in discordance with our finding, Trichophyton rubrum was found to be the dominant causative organism in Superficial mycoses as well as Dermatophytosis by reports from Smita Sarma and A K Borthakur [10] from Northern Assam, A Naglot et al [6] from Tezpur Assam, Sen SS and Rasul ES in Guwahati Assam [11], Sumit Kumar et al [9] in North India and so has been found by A Lakshmanan et al in Chennai [3], V Bindu and K Pavithran [12] in Calicut, Lavanya V and S S Solabannavar [5], KAK Surendran et al in Karnataka [13], B Janardhan et al [14] in Telangana. On the contrast, Trichophyton tonsurans was the commonest isolate from Superficial mycoses in report of Lt Col P Roy and Wg Cdr S. Grover [8] from Northeast India.

Male gender was found to be significantly (Chi square – 3.9419, p value 0.047) associated with higher prevalence of Superficial mycoses which might be due to their higher physical activities associated with relatively higher sweaty outdoor activities and exposure to fungal infection than females (who mostly presented with cutaneous lesions in intertriginous areas in this study). Similarly, male preponderance of Superficial mycoses was also reported by studies carried out by Lavanya V and S.S Solabannavar [5], Sumit Kumar et al [10], Smita Sarma and AK Borthakur et al [11], Sen SS and Rasul ES [12], V Bindu and K Pavithran [13], B Janardhan and G Vani [15], US Agarwal et al [17], Sonia Mahajan et al [18]. The lower prevalence in females might be due to non-reporting to health care facilities from semi-urban and rural areas due to ignorance and social stigmas or due to over-the-counter use of topical steroids.

Unskilled manual labour was also found to be associated (Chi square- 10.3438, p value 0.0012) with higher prevalence of Superficial mycoses as far as occupational association is concerned. Higher incidence may be due to poor hygienic practices and overcrowding in the lower socio-economic strata.

In this study higher number of cases of superficial mycoses were reported midway between Spring and Summer. Similar observation has also been reported by B Janardhan and G Vani [15] while Smita Sarma and A K Borthakur [11] from Assam reported higher number of cases in August and September.

**CONCLUSION**

The prevalence of superficial mycoses in cases reporting to tertiary health care center of Southern part of Assam has been found to be 33.2%. Male subjects in the age group of 21-50 years and unskilled laborers from lower socio-economic strata have been found to be affected more by superficial mycoses. Tinea corporis has been found as the most common clinical presentation among the Dermatophytosis and Pityriasis versicolor to be the most common among non-dermatophyte superficial mycosis. Maximum cases were reported midway between spring and...
summer. Trichophyton mentagrophyte was the mostly isolated causative fungus for superficial mycosis in cases presenting to this tertiary care hospital. Occupations associated with activities of manual labour with prolong outdoor exposure and excessive sweating is more affected by superficial mycoses. Since different fungal infections run different disease courses and therapeutic options are different in each case and also some yeast-like isolates being intrinsically resistant to some antifungal drugs, identification of causative fungal genus and/or species by direct microscopy and culture is vital in clinically identified cases where molecular diagnostics is not available to curtail the emergence of drug resistance.

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