Int. J Med. Pharm. Res. (P-ISSN: 2958-3675 | E-ISSN: 2958-3683)
Available on: https://ijmpr.in/

International Journal
of Medical and Pharmaceutical Research

ORGINAL ARTICLE **OPEN ACCESS**

Speciation and Clinical Classification of Dermatophytosis in Tertiary Care Hospital

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Received: 19-07-2024
Accepted: 28-09-2024
Available online: 01-10-2024

A B S T R A C T

Introduction*:** There is a significant increase in the incidence of superficial dermatophytosis in India for last 5-6 years. Dermatophytosis poses a significant public health concern. According to Emmons Morphological criteria, dermatophytes are classified into three anamorphic genera (based on conidial morphology). Trichophyton (mainly infect skin, hair and nails) Microsporum (infect skin and hair not the nails) & Epidermophyton (infect skin as well as nails but not the hair). As a result of the variety of species and their habitat association, dermatophytes have long been classified as anthropophilic, zoophilic and geophilic, species that exclusively affect human are known as Anthropohilic while those inhabiting domestic and wild animals as well as birds are called zoophilic and third group isolated from the soil is known as geophilic. Zoophilic species tend to produce highly inflammatory reaction and anthropophilic species produce mild but chronic lesions. These include Tinea pedis (foot) or athlete's foot and Tinea barbae or barbers itch (bearded area of the face and neck). The disease caused by non Dermatophytic fungi infecting skin are called as dermatomycoses, where hair and nail are known as piedra and onychomycoses respectively. An example of a very common dermatophyte infection is the athlete's foot also called as Tinea Pedis. Another common dermatophyte infection affecting the groin area is jock itch, also known as tinea cruris. In these infections itching is severe and disabling lesions on the genitals and other area because social embarrassment and impair quality of life also. The epidemiology of dermatophytes varies among countries and even within different regions in the country. Surveillance studies are required to understand the changing clinicoepidemiology and prevalence of culprit agent for the effective treatment. ***Aims*:** This present study was conducted to identify the clinical profile and species of dermatophytic infection at our tertiary care centre IIMSR Lucknow Uttar Pradesh India. ***Study design*:** The present study was a hospital based observative prospective study. ***Statistical Analysis*:** Statistical analysis was done using SPSS version 29.0.1.1. Chi square test and appropriate tests of significance were applied. ***Method*:** It was a hospital based observational study of 156 clinically diagnosed case of dermatophytosis during five months duration. Sociodemographic details, clinical history and detailed examination were conducted from all patients. Skin scrapings were sent to microbiology department of our hospital for direct microscopy and culture. ***Results: Among 156 patients enrolled males were commonly affected than females, male to female ratio was 2.6:1. Incidence was maximum in rural area 56% as compared to urban. Multiple site infection (35.9%) was found to be commonest clinical type followed by Tinea cruris (16.67%). Trichophyton tonsuran was the commonest isolate obtained (35.2%) followed by Trichophyton mentagrophytes (22%). The outdoor activities, such as farming, labour work and poor personal hygiene in rural area were identified as risk factor for infection. ***Conclusion*:** The present study has provided recent data on etiological agent of dermatophytosis and risk factor in tertiary care

Centre.Microscopy with and without culture is an important diagnostic tool in dermatophytosis. It is important to develop measurement for disease prevention and control along with effective therapy**.**
Keywords: Dermatophytosis, microscopy, Tinea corporis, Tinea cruris.

INTRODUCTION

Dermatophytosis is a superficial fungal infection of keratinized tissues caused by dermatophytes that utilized keratin as the source of nutrient and colonized keratinized tissues namely stratum corneum of the epidermis of hair, nail and skin. The disease is the results of the host's reaction to the cell wall of fungus and the enzyme released by the fungus during its digestive process. Most of the infection are not life threatening but they can cause morbidity in immunocompromised, diabetic people, who use communal baths, sports person, wrestlers. Outbreaks of infection can occur in school households and institutional settings. The organism are transmitted by either direct contact with infected host or by direct or indirect contact with infected exfoliated skin or hair in comb, hair brushes, clothing, furniture, theatre seat, cap, bed linen, towel, hotel rugs, and locker room floor. In this infection itching is severe and disabling lesions on the genital and other area cause social embarrassment and impair quality of life also redness, scaling or fissuring of the skin or a ring with irregular borders and a cleared central area may occur. If the infection involves the scalp, there may be hair loss on the affected area. More aggressive infection may lead to an abscess or cellulitis. Area infected by dermatophytes may become secondarily infected by bacteria. Depending on the species the organism may be viable in the environment for up to 15 months. Symptoms typically appear between 4 to 14 days following exposure.

Reports indicate that the epidemiology of dermatophytes varies among countries and even within different region in the country. According to World Health Organization about 20% of the world's population is affected by dermatophytes and this infection is more prevalent in tropical and subtropical countries like India, where heat and humidity are high for most of the year. It is difficult to calculate the exact incidence and prevalence owing to a paucity of community based survey. The current reported prevalence in India falls in a very wide range (6.09%-61.5%). A prevalence of 6.09% to 27.6% has been reported in studies from south India, while a high prevalence of 61.5% has been recorded in north India. Interestingly, though dermatophytosis is expected to be more prevalent in south India hot and humid climates and less so in north India, no such association is apparent.

A significant increase in the incidence of the superficial dermatophytosis in India over the past 5-6 years has been observed. During different times frame studies from Kolkata, Ahmedabad and Chennai have revealed an increasing trend. The prevalence has increased many folds. Chronicity, relapse, recurrence, reinfection and treatment unresponsiveness became the norm. Certain infectious species are geographical restricted and endemic only in particular parts of the world. Other species may be sporadic but have worldwide distribution. The species that are endemic within a population are carried by that population to new places by troop movement, migration of labourers, emigration, social habits and rapid worldwide travel have all contributed to the changing distribution of infection.

Material and Methods

Total 156 clinically diagnosed cases of dermatophytosis attending the OPD of our tertiary care centre during five months from 1st January 2023 to 30th may 2023 were selected for study. Ethical clearance was obtained from the institutional ethical committee prior to the start of the study.

Inclusion Criteria

1. Clinically suspected patients with dermatophytic infection.
2. Patients who had given their consent of participation.

Exclusion Criteria

1. Those who had taken systemic antifungal drugs during the past week.
2. Those patients who took topical antifungal one day before.
3. Patients who refused to give their consent.

Sufficient clinical materials like skin scraping, plucked hairs and nail clipping were collected for both direct microscopy and culture.

Sample size Estimation:

The prevalence of dermatophytosis (p) was taken to be 61.3% with 8% allowable error on either side and a 95% confidence level. The sample size (n) was calculated according to Abraham Walds formula.

Statistical Analysis:

The collected data were coded and entered into Excel software (Microsoft Office Excel 2010) database. Data was analyzed using statistical package for social sciences (IBM SPSS Statistics Version 29.0.1.1) (SPSS, Inc. Chicago, IL, USA). Appropriate tests of significance were applied to find out the statistical significance of the difference in percentages or proportions. Statistical significance was assessed at $P < 0.05$.

Sample Collection:

Hair: After disinfecting the infected area with 70% alcohol, that infected hairs appeared dull broken and faded were epilated with the sterile forcep**.**

Skin: After disinfecting, w scraped the skin vigorously from the edge of the infected area using blunt side of a sterile scalpel blade no 23 and collected it on sterile white filterpaper.

Nail: After disinfection nail was collected either by nail clipper or scraping them using a scalpel blade no 15 or a sterile nail cutter. If there was any material under the leading edges of nail, we also scraped it and placed it in a sterile filter paper. To prepare the nail pieces for plating, we used a scalpel to pulverize or mince them into small fragments.

Direct microscopy: Emulsifying the collected specimen in 1 drop of 10% KOH for skin and 40% KOH for hair and nails. Leave it in a wet chamber for 10-15 min, in case of hair and nail, wait overnight at room temperature. Examine the slide under low power 10X for detection and high power 40X magnification for confirmation under bright field microscopy.

Culture: After conducting a direct microscope examination, regardless of whether fungal elements were observed or not, all the specimen were cultured in Sabourauds dextrose agar for colony morphology and characters.

Dermatophyte Test Medium (DTM):

This media selectively promotes the growth of dermatophytes, which cause a color change in the medium from yellow to red within 14 days. The objective of investigating the growth of the Dermatophytes test medium was to analyze the visible traits of both the front and back colonies, as well as the presence of diffuse pigmentation on the media. Dermatophytes colonies have various textures such as cottony, powdery or suede like. Colours, size and different morphologies. DTM contains a pH indicator (phenol red) that changes the colour of the media. The medium is yellow at pH below 6.8 which turns pink at pH >8.2 in the presence of alkaline metabolites produced by dermatophytes.

Lacto phenol cotton blue mount or tease mount (LPCB): LPCB mount is used for examining fungal specimen. It consists of phenol which acts as a cleansing agent, lactic acid which aids in the preservative of fungal structure, cotton blue, an aniline dye that stain the chitin in fungal wall and glycerol, a sticky ingredient that prevents that prepared slide specimen from drying out.

PROCEDURE

After wearing the appropriate PPE (personal protective equipment), petridish containing corn meal agar was taken, we have cut out 1cm square blocks of agar with a sterile blade from the corn meal agar for each slide culture to be inoculated, placing an agar block in the centre of the sterile slide by using a flat side of a sterile spatula. Inoculated three four fragment of the growth from the SDA tube around the periphery of the agar block using a Sterile straight wire- loop and then placed sterile cover slip on the agar block. We have thoroughly moisten a filter paper in a sterile petridish with sterile distilled water using a pipette, then placed a glass rod in the petridish, ensuring it is positioned to prevent the slide from sticking to the surface, placing the slide culture above the glass rod and labeled the petridish and then inoculated in BOD (Biological oxygen demand) incubator at 25 degree Celsius until visible growth is seen. When growth appeared beneath the coverslip, we took a clean slide and placed a drop of LPCB stain on it, examined the slide under low power (10X) and high power (40X).

DISCUSSION

Dermatophytes infection poses a significant public health issue. For efficient management of cases, it is important to determine their frequency, identify the main causative species, study risk factor and clinical persistence.

In the present study dermatophytosis was found to be common in the age group 20-40 yrs which is in accordance with Sumathi S *et al*., Grover S *et al*., [1, 2]. Higher incidence was noted among males (72.4%) than females (27.6%) because male were involved more in outdoor activities and female hormones such as estrogen and progesterone which suppress the growth of dermatophytes [3, 4].

The current research found that a majority of the patient (56%) were residing in rural area. It is possible that the higher occurrence of dermatophytosis in rural area is linked to poorer personal hygiene among rural individual compared

to their urban counterparts.In study conducted by Kaur *et al*., [5] observed that the patients from urban area (81.5%) was higher than that from rural area (18.5%), which contrast with our finding.

Multiple site infection was the commonest (35.9%) followed by Tinea cruris (16.6%). Our study showed (40%) incidence of dermatophytosis was among individual of daily wager category, farmers, laborers followed by students (25%) [6, 7].

Trichophyton tonsurans (35.2%) was found to be most prevalent species isolated in our study. In the study conducted by Lakshmanan A *et al*., [8], the predominant species identified was Trichophyton rubrum (79%) followed by T. menragrophytes. In contrast Siddiqui M *et al*., finding indicated that T.rubrum compromised (25.45%) of cases, T mentagrophytes accounted for (7.27%) and other species such as Microsporum audounii, Trichophytons choenleinii and Epidermophyton floccosum each constituted (0.91%) of the cases [7]. This research showed that most of the patients (42%) were already using antifungal medication while 40% patients were taking Over The Counter drugs.

In the present study patients were positive on both culture and KOH mount was 29.4% while 14.1% were negative on KOH but positive on culture and 29.4% were negative on both culture and KOH mount. The results of KOH and culture vary from study to study can be due to multiple factors such as the non-viability of fungal elements, the presence of contaminants and certain technical errors [7].

In previous research [9, 10] it was noted that more than half (53.3%) of the patients experienced symptoms for over three months, while 33.7% experienced symptoms for one to three months and 13% experienced symptoms for less than a month. Our study aligns with these findings, revealing that 42.4% of cases had an illness duration ranging from one to four months. This similarity of results indicates that the duration of the illness is consistent over time regardless of the population studied [11].

CONCLUSION

Present observational study evidence that multiple site infection is most common clinical presentation followed by Tinea cruris. Trichophyton tonsurans is the commonest specie in our tertiary care centre. Fungus were demonstrated by direct microscopy and culture (56%) out of 156 cases, hence direct microscopy with and without culture is an important tool for diagnosis of dermatophytosis. Use of antifungal drugs without culture positivity can leads to antifungal resistance. Antifungal susceptibility testing could not be done as it was a short duration study.

Foot notes:
Source of support: Nil

Conflict of Interest: Nil

Ethical Clearance: The study was approved by the Institutional ethics committee (IEC/IIMS&R/2023/37).

Consent to Participate Statement: Written informed consent was obtained from all patients to participate in the study.

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