

Isolation and Identification of Dermatophytes from Clinical Samples – One Year Study

A. Anupama*

Department of Microbiology, Akash Institute of Medical Sciences and Research Center
Bangalore, Karnataka, India

*Corresponding author

ABSTRACT

The main aim of this study to isolate and identify the fungi causing dermatophytosis. Also this study of dermatophytosis was carried out in the Department of Microbiology, KLE'S Dr. PrabhakarKore Hospital and Medical Research Centre Belgaum, over a Period of one year. All skin, hair and nail samples from clinically suspected cases of dermatophytosis were included in the study. The specimens collected were inoculated on to Sabourauds Dextrose agar containing Chloramphenicol and Cycloheximide irrespective of demonstration of fungal elements on KOH mount. The isolates were inoculated on potato dextrose agar for better conidiation. Out of 100 cases included in our study male preponderance of 73 (73%) cases was seen. 27 (27%) were females. Incidence of Dermatophytosis was high in males and male to female ratio of 2.7:1. Tinea cruris was the most common clinical type of dermatophytosis with incidence of 32. 37 samples were KOH negative and 63 samples were KOH positive. Out of 63 isolates of dermatophytes in our study *T. rubrum* was the most common dermatophyte. Dermatophytosis is the most common type of cutaneous fungal infection. The incidence of Dermatophytosis is increasing in India due to widespread and indiscriminate use of corticosteroids and antifungal agents without appropriate microbiological investigations. Dermatophytes isolated included predominately *Trichophyton* species, of which *T. rubrum* was the commonest dermatophyte isolated. *T. mentagrophytes*, *T. tonsurans*, *M. gypseum* were other isolates from clinical samples.

Keywords

Dermatophytes
from Clinical
Samples,
Keratinolytic fungi.

Article Info

Accepted:
12 September 2017
Available Online:
10 November 2017

Introduction

Dermatophytes are keratinophilic and keratinolytic fungi. They are responsible for dermatophytosis which are superficial mycosis affecting skin (Tinea corporis, Tinea cruris, Tinea pedis), hair (Tinea capitis), beard (Tinea barbae), nails (onychomycosis or Tinea unguium). Dermatophytosis is a nonfatal disease except in extremely rare cases of Hadida and Schousboe's dermatophytic disease.¹

Infection generally involves skin and restricted to the cornified layers because of the inability of the fungi to penetrate the deeper tissues or organs of immunocompetent hosts.² Dermatophytes are a group of closely related organisms that can use keratin as a nitrogen source. There are three genera of Dermatophytes; *Trichophyton*, *Microsporum* and *Epidermophyton*.

Because of difficulty in clinically differentiating dermatophytosis from other non-mycotic dermatosis, particularly in the dystrophic nails, it is important to establish an accurate diagnosis. Knowledge of the zoophilic or anthropophilic origin of the dermatophyte may allow setting up prophylactic measures¹.

Any clinical diagnosis needs to be supported by laboratory diagnosis. Culture is a necessary adjunct to direct microscopic examination for definitive identification of etiological agent and in many instances the choice of therapy depends upon the specific identification of the invasive mould. This is especially important in nail and skin infection, often caused by non-dermatophytic filamentous fungi, which are often resistant to usual dosage of the therapy used for dermatophytic infections. Before starting treatment for dermatophytosis, it is essential to establish the diagnosis of the disease, so that specific therapeutic modalities can be monitored during the course of the treatment.³

Rapid identification of dermatophyte species and knowledge of their host preference and ecology play an important role in epidemiology, public health and infection control.

The varied clinical presentation of Tinea, which results in delay in diagnosis, poor compliance in follow up of cases, and consequently spread of infection in the community has rekindled interest in rapid identification of species⁴.

The main aim of this study is to isolate and identify the fungi causing dermatophytosis.

Materials and Methods

This study of dermatophytosis was carried out in the Department of Microbiology, KLE'S

Dr. Prabhakar Kore Hospital and Medical Research Centre Belgaum, over a Period of one year.

Inclusion criteria

All skin, hair and nail samples from clinically suspected cases of dermatophytosis were included in the study.

Exclusion criteria

Patients who are already on treatment for dermatophytosis were excluded from the study

Statistical Analysis

Was done using percentage and chi-square test.

Specimen collection

From the skin

The affected area was first thoroughly swabbed with 70% alcohol to remove surface contaminants. After the alcohol dries, the skin scrapings were collected from the border of the active lesions with a beard parker blade in a sterile black paper envelop.

From the scalp

Hairs from the scalp were epilated with a flame sterilized forceps and the active border area was scraped with a scalpel to collect epidermal scales on a sterile black paper envelop.

From the nail

The affected nail was first cleaned with 70% alcohol. The upper portion of the infected nail was scraped away and material were collected from the deeper part of the distal end of the nail on to sterile black paper envelop.

The samples of skin and hair were subjected to 10% KOH and 20% KOH preparation for nail. After 15-20 minutes, the specimen was examined for the presence of hyphae or arthrospores.

Culture

The specimens collected were inoculated on to Sabourauds Dextrose agar containing Chloramphenicol and Cycloheximide irrespective of demonstration of fungal elements on KOH mount. Each sample was inoculated into two tubes. One tube with antibiotic and other without antibiotic and were incubated at 27⁰C. The cultures were examined daily for a period of 4 weeks. Slopes showing no growth for 4 weeks were discarded. If growth was obtained on Sabourauds Dextrose agar, identification was made based on colony morphology, microscopic appearance and other relevant tests. The isolates were inoculated on potato dextrose agar for better conidiation.

Macroscopic examination of culture

The growth on Sabourauds dextrose agar was observed to study the colony morphology, the color of the surface, the reverse of the colony, the texture of the surface, the topography and the rate of growth.

Microscopic Examination of Culture was done using LPCB preparation and slide culture was done in case the morphology was not clear in LPCB preparation.

Biochemical test

Urease test

This test is done on Christensen's medium. This is done to differentiate *T. mentagrophytes* from *T. rubrum*. *T. mentagrophytes*, hydrolyse urea and the

medium becomes deep red while *T. rubrum* does not hydrolyse urea. Urea broth may also be used which is more sensitive

Results and Discussion

Out of 100 cases included in our study male preponderance of 73 (73%) cases was seen. 27 (27%) were females. Incidence of Dermatophytosis was high in males and male to female ratio of 2.7:1.

Tinea cruris was the most common clinical type of dermatophytosis with incidence of 32, Tinea capitis was seen in 3 cases, Tinea unguium was seen in 20 cases, Tinea pedis was seen in 12 cases, Tinea manuum was seen in 2 cases and 31 cases presented with Tinea corporis (Table 1).

Tinea cruris was the most common and Tinea manuum was the least common clinical type of dermatophytosis encountered in our study.

Out of 100 clinical samples 58 samples were culture positive and KOH positive. 19 samples were Culture positive and KOH negative. 18 samples were Culture as well as KOH negative. 5 samples were KOH positive and culture negative.

Out of 63 isolates of dermatophytes in our study *T. rubrum* was the most common (33), followed by *T. mentagrophyte* (20), *T. tonsurans* (8) and *M. gypseum* (2).

In present study 100 clinically diagnosed cases of Dermatophytosis were studied. Of them 77 were skin scrapings, 20 were nail clippings and 3 were hair stubs. Out of these samples, dermatophytes were isolated in 63 cases.

Among 63 dermatophytes isolated, *T. rubrum* was the commonest species (33) followed by *T. mentagrophytes* (20). *T. tonsurans* was

isolated from (8), *M. gypseum* in (2), other than dermatophytes *Penicillium spp* was isolated in 3 cases, *A.niger* in 7, *Acremonium* in 3 and *Curvilaria* in 4. No fungal growth was seen in 22 clinical samples.

Present study shows that out of 100 clinically diagnosed cases of dermatophytosis 63 were affected by dermatophytes. Our results are comparable to studies conducted by Abu Elteen *et al.*, (1999) where in isolation rate was 56.8%. In a study conducted by Bindu *et al.*, (2002) and Seemabose *et al.*, (2013) isolation rate was 45% and 60.67% respectively, which is again comparable to results of present study. Extreme variation in isolation rate in few other studies may be due

to differences in techniques used for sample collection, culture and other methods of identification.

In present study most common type of dermatophytosis studied was Tinea cruris with incidence of 32 (32%). Study conducted by Sanchita Karmakar *et al.*, (1995) also showed similar results.

Studies conducted by Bindu *et al.*, (2002), Sumana *et al.*, (2002), Agarwalla *et al.*, (2001), Aghamirian *et al.*, (2004) and Aruna Aggarwal *et al.*, (2013) showed T. corporis being most common dermatophytosis with incidence of 54.6%, 48.7%, 43% 20.7% and 36.2% respectively.

Table.1 Various clinical types of dermatophytosis

Dermatophytosis	Number of Cases
Tinea cruris	32
Tinea capitis	3
Tinea unguium	20
Tinea pedis	12
Tinea manuum	2
Tinea corporis	31

Out of 100 clinical samples 37 samples were KOH negative and 63 samples were KOH positive. Out of 100 clinical samples 88 were culture positive and 22 were culture negative.

Dermatophytosis	<i>T.rubrum</i>	<i>T.mentagrophytes</i>	<i>T.tonsurans</i>	<i>M.gypseum</i>	Total
Tinea cruris	13	6	2	0	21
Tinea capitis	0	0	1	0	01
Tinea unguium	2	2	1	0	05
Tinea pedis	5	4	0	1	10
Tinea manuum	1	0	0	0	01
Tinea corporis	12	8	4	1	25
Total	33	20	08	02	63

Table.2 Dermatophytes isolated from various clinical types

Studies	Sample size	Isolation rate
Abu Elteen <i>et al.</i> , (1999) ⁵	350	56.8%
Agarwalla <i>et al.</i> , (2001) ⁶	100	94%
Seemabose <i>et al.</i> , (2013) ⁷	150	60.67%
SanchitaKarmakar <i>et al.</i> , (1995) ⁸	250	8.6%
Asticcioli <i>et al.</i> , (2008) ⁹	100	97%
Bindu V <i>et al.</i> , (2002) ¹⁰	150	45.3%
Sumana MN <i>et al.</i> , (2002) ¹¹	150	24%
Present Study	100	63%

Study	Commonest Dermatophytosis	Percentage
Bindu V <i>et al.</i> , (2002) ¹⁰	Tinea corporis	54.6%
Sumana MN <i>et al.</i> , (2002) ¹¹	Tinea corporis	48.7%
Agarwalla <i>et al.</i> , (2001) ⁶	Tinea corporis	43%
SanchitaKarmakar <i>et al.</i> , (1995) ⁸	Tinea cruris	34.8%
Aghamirian MR <i>et al.</i> , (2004) ¹²	Tinea corporis	20.7%
ArunaAggarwal <i>et al.</i> , (2002) ¹³	Tinea corporis	36.2%
	Tinea cruris	32%
Present Study	Tinea cruris	32%
	Tinea corporis	31%

Studies	Dermatophyte	Percentage
Abu Elteen <i>et al.</i> , (1999) ⁵	<i>T.mentagrophytes</i>	32.7%
Agarwalla <i>et al.</i> , (2001) ⁶	<i>T.rubrum</i>	45.74%
Asticcioli <i>et al.</i> , (2008) ⁹	<i>T.rubrum</i>	42.3%
Bindu V <i>et al.</i> , (2002) ¹⁰	<i>T.rubrum</i>	66.2%
Sumana MN <i>et al.</i> , (2002) ¹¹	<i>T.rubrum</i>	52.7%
Seema Bose <i>et al.</i> , (2013) ⁷	<i>T.rubrum</i>	33.3%
Present Study	<i>T.rubrum</i>	33%
	<i>T.mentagrophytes</i>	20%

In present study *T.rubrum* was the commonest dermatophyte isolated from 33 clinical samples. Second most common dermatophyte isolated was *T. mentagrophyte* with the incidence of 20. In various studies conducted by Agarwalla *et al.*, (2001), Asticcioli *et al.*, (2008), Bindu *et al.*, (2002) and Sumana *et al.*, (2002) *T. rubrum* was the commonest isolate with incidence ranging from 42.3% to 66.2%. The present study was comparable with Seema Bose *et al.*, (2013). Some species

of dermatophytes are endemic in certain parts of the world. Various studies done in India have shown *T. rubrum* as the commonest isolate (Table 2).

Dermatophytosis is the most common type of cutaneous fungal infection. It is very common in our country with several contributing factors like hot humid climate, poor hygiene, increased outdoor activities, occupational trauma and immunosuppression.

The incidence of Dermatophytosis is increasing in India due to widespread and indiscriminate use of corticosteroids and antifungal agents without appropriate microbiological investigations.

Dermatophytes isolated included predominately *Trichophyton* species, of which *T. rubrum* was the commonest dermatophyte isolated. *T. mentagrophytes*, *T. tonsurans*, *M. gypseum* were other isolates from clinical samples.

The overall isolation rate of dermatophytes was 63%. Tinea cruris accounted for maximum number of cases

References

- Robert R., Pihet M. Conventional methods for the diagnosis of dermatophytosis. *Mycopathologia* 2008; 166: 295-306.
- Cas E. Parasitic adaptation of pathogenic fungi to mammalian hosts. *Critical reviews in microbiology*. 1986; 13 (2): 173—218.
- Chander J. Textbook of Medical Mycology. 2nd ed. Pune: Mehta Publishers; 2009. chapter 10, Dermatophytoses; 91-100 & 376-88.
- W T. Topley & Wilson's microbiology and microbial infections. 9th ed. London: Hodder Arnold; 2005; 215-235
- Abu-Elteen KH, Abdul M. Prevalence of dermatophytoses in the Zarqa district of Jordan. *Mycopathologia* 1999; 145(3): 137-42.
- Agarwalla A, Jacob M, Sethi M, Parija SC, Singh NP. A clinico-mycological study of dermatophytosis in Nepal. *Journal of Dermatology* 2001; 28(1): 16-21
- Bose S, Barapatre R. Clinico--mycological profiles of dermatophytoses in a tertiary care rural hospital. *International Journal of Biomedical and Advance Research*. 2013; 4 (1): 31-34.
- Karmakar SG, Kalla KR, Joshi, Karmakar S. Dermatophytoses in a desert district of Western Rajasthan. *Indian J Dermatol Venereol Leprol* 1995; 61(5): 280-83.
- Asticcioli S, Di Silverio A, Sacco L, Fusi I, Vincenti L, Romero E Dermatophyte infections in patients attending a tertiary care hospital in northern Italy. *New Microbiologica* 2008; 31: 543-48.
- Bindu V, Pavithran K. Clinico - mycological study of dermatophytosis in Calicut. *Indian J Dermatol Venereol Leprol* 2002; 68(5): 259-61.
- Sumana MN, Rajagopal V.A study of dermatophytes and their in-vitro antifungal sensitivity *Indian J Pathol Microbiol*. 2002 Apr; 45(2): 169-72.
- Aghamirian MR, Ghiasian SA Dermatophytoses in outpatients attending the Dermatology Center of Avicenna Hospital in Qazvin, Iran. *Mycoses* 2008 Mar; 51(2): 155-60.
- Aggarwal A, Arora U, Khanna S, Others. Clinical and mycological study of superficial mycoses in Amritsar. *Indian Journal of Dermatology*. 2002; 47 (4): 218.

How to cite this article:

Anupama, A. 2017. Isolation and Identification of Dermatophytes from Clinical Samples – One Year Study. *Int.J.Curr.Microbiol.App.Sci*. 6(11): 1276-1281.
doi: <https://doi.org/10.20546/ijcmas.2017.611.152>