

Original Article

A STUDY OF PATTERN OF ONYCHOMYCOSIS IN A TERTIARY CARE HOSPITAL OF CENTRAL INDIA

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ABSTRACT

Objective: This study was undertaken to study the clinico-mycological profile of clinically suspected cases of onychomycosis in the region.

Methods: In this cross-sectional study, 65 non-repetitive, clinically diagnosed cases of onychomycosis visiting Dermatology, Venereology and Leprosy OPD of People's Hospital, Bhopal were included. Nail clippings and scraping beneath the nails were taken from the affected sites. Collected specimens were subjected to standard mycological procedures.

Results: Males were more affected than females. The male to female ratio was 1.4:1. Distal and lateral subungual onychomycosis (DLSO) was the commonest clinical pattern. Out of the total 35 fungal culture isolates, dermatophytes (71.43%) were the most common. Among dermatophytes, *Trichophyton rubrum* and among non-dermatophytes, *Candida spp.* was the most common isolate.

Conclusion: Dermatophytes were the most common aetiological agent of onychomycosis in our study with *Trichophyton rubrum* as the most common isolate. This study also reveals the fact that nowadays, non, dermatophytes are increasingly isolated from cases of onychomycosis.

Keywords: Onychomycosis, Dermatophytes, KOH wet mount, Fungal culture

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INTRODUCTION

Onychomycosis is a fungal infection of the nails and can be caused by dermatophytes, yeasts, or non-dermatophytes. Dermatophytes are considered the most common etiologic agents of onychomycosis and infection caused by them is referred to as *Tinea unguium* [1]. In India, it mainly affects the low socio-economic sections of the population [2].

The prevalence of onychomycosis across the world was found to be around 3-18 % in different studies [3-7]. It is one of the most common nail diseases and accounts for 20-50% of all nail diseases [1, 3].

Depending upon the morphology of nails during the infection, different clinical patterns of onychomycosis have been described, namely distal and lateral subungual, white superficial, proximal subungual, and total dystrophic onychomycosis (fig. 1)

Onychomycosis affects the male population more frequently and the most common clinical pattern is distal subungual onychomycosis. Toenails are affected more commonly than fingernails [3].

The clinical presentation of onychomycosis mimics many nail diseases such as traumatic onycholysis, nail psoriasis, onychogryphosis, and Lichen Planus making clinical diagnosis difficult and, therefore, laboratory confirmation is necessary. Also, laboratory confirmation is necessary as treatment depends on the fungus species involved whether dermatophyte, yeast or a non-dermatophytic mould [8].

Onychomycosis requires targeted and several months of treatment, as the growth of nails, is very slow, especially in the elderly. Failure to start treatment at the appropriate time may lead to permanent damage to the nail plate. Therefore early institution of antifungal agents is required. Confirmation of an etiological agent by laboratory diagnosis is often time-consuming and therefore starting empirical treatment based on prevalence studies is required till diagnosis is confirmed.

As there is a paucity of such studies in this part of Central India, we have undertaken the study to determine the clinico-mycological profile of suspected cases of onychomycosis in the region.



Fig. 1: Different clinical presentations of Onychomycosis

MATERIALS AND METHODS

This cross-sectional study was undertaken after obtaining clearance from Institutional Ethics Committee, People's College of Medical Sciences and Research center, Bhopal (PCMS/OD/2013/3808) and after taking written informed consent from the study subjects.

Study population: Clinically diagnosed cases of onychomycosis visiting Dermatology, Venereology and Leprosy OPD of People's Hospital, Bhopal.

Study period: November 2013-October 2015.

Place of study: Department of Microbiology, PCMS and RC, Bhopal.

Inclusion criteria: -Patients of all age groups and both sex are included in the study.

Exclusion criteria: -Bacterial isolates were excluded. Patients already on antifungals were excluded from the study.

Study type: -Hospital-based, cross-sectional study.

Sample size: -Non-repetitive 65 suspected cases of onychomycosis.

Specimen collection and processing

The affected nail was wiped with 70% ethanol. Then nail clippings and scraping beneath the nails were taken. Samples were collected in clean black paper packets. Collected specimens were subjected to standard mycological procedures.

• Direct microscopic examination

KOH wet mount-This was prepared by placing a portion of each sample collected on a glass slide. Then 1-2 drops of 20% or 40% KOH were added and kept for 1-2 h. The slide was then screened for the presence of fungal hyphae.

• Fungal culture

The other portion of the collected sample was inoculated onto three test tube slants in duplicate on Sabouraud's dextrose agar (SDA) with chloramphenicol (0.005%), Sabouraud's dextrose agar (SDA) with chloramphenicol (0.005%), and cycloheximide (0.05%), and Dermatophyte test medium (DTM). SDA with chloramphenicol and SDA with chloramphenicol and cycloheximide were incubated at 25 °C and 37 °C for up to 4 w and observed periodically for growth. If there was no growth even after 4 w of incubation, it was reported as negative. Dermatophyte test medium (DTM) was incubated at 25 °C and 37 °C for ten days and was observed for color change.

• Identification of isolates

Fungal isolates were identified on the basis of distinctive colony characteristics, microscopy features through Lactophenol cotton's blue (LCB) tease mount, slide culture, urease test, and hair perforation test [1, 9] (fig. 1-10).

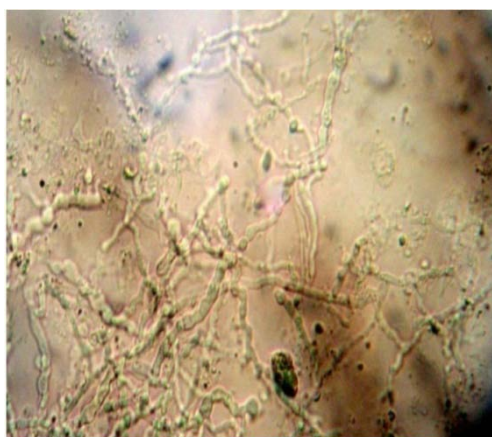


Fig. 2: KOH mount of nail specimen



Fig. 3: DTM agar showing growth



Fig. 4a: Growth of *Trichophyton rubrum* on SDA slant



Fig. 4b: LCB mount



Fig. 5a: Growth of *Trichophyton mentagrophytes* on SDA plate

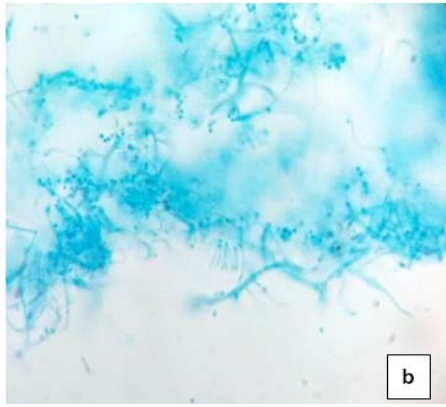


Fig. 5b: LCB mount

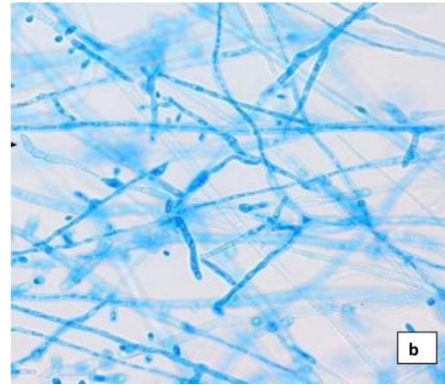


Fig. 7b: LCB mount



Fig. 6a: Growth of *Epidermophyton floccosum* on SDA slant

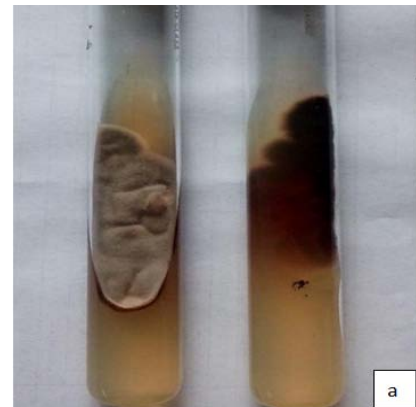


Fig. 8a: Growth of *Alternaria spp.* on SDA slants



Fig. 6b: LCB mount

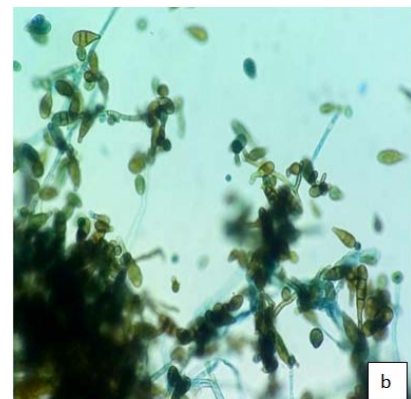


Fig. 8b: LCB mounts



Fig. 7a: Growth of *Trichophyton tonsurans* on SDA plate



Fig. 9a: Growth of *Curvularia spp.*



Fig. 9b: LCB mounts

Fig. 10a: Growth of *Aspergillus niger*

Fig. 10b: LCB mount

RESULTS

In this study of 65 suspected cases of onychomycosis, patients were distributed between the age ranges of 5-69 y. The mean age of the study group was 36.32 y. Males were more affected than females contributing to 58.46% of total cases. The male to female ratio was 1.4:1. The age group of 31-40 y was most commonly affected with 26 cases (40%) followed by the 41-50 y age group with 14 cases (21.54%) (table 1).

In our study, onychomycosis was most common in agricultural workers (46.15% cases) followed by housewives (23.08%) (table 2)

In our study, distal and lateral subungual onychomycosis (DLSO) was the commonest clinical pattern (63.08%) followed by proximal subungual onychomycosis (21.54%), total dystrophic onychomycosis (13.85%), and Superficial white onychomycosis (1.54%). The toenails were more frequently involved (55.38%) followed by fingernails (29.23%). Both toenails and fingernails were found to be involved in 15.38% of cases (table 3).

Out of 65 clinically suspected cases of onychomycosis, in 45 cases (67.69%) we were able to detect fungi either by direct microscopy and/or culture. In 33cases (50.77%) both microscopy and culture were positive. 9cases (13.85%) were positive only by microscopy but culture turned out to be negative. In 2 cases (3.08%) culture was positive but microscopy was negative. In 21 cases (32.31%) both microscopy and culture were negative (table 4).

Considering fungal culture as the gold standard, direct microscopy (KOH) findings were evaluated. The sensitivity of direct Microscopy (KOH) was found to be 94.29% and the specificity turned out to be 70% (table 5).

Out of the total of 35 fungal isolates, the dermatophytes were the most common (71.43%). Amongst dermatophytes, there were 12 (48%) isolates of *Trichophyton rubrum* followed by 09 (36%) isolates of *Trichophyton mentagrophyte*, 03(12%) of *Trichophyton tonsurans*, and 01(4%) of *Epidermophyton floccosum*. Non-dermatophyte isolates were 10(28.57%) of which 4(40%) were *Candida spp.* and 2 isolates each of *Alternaria spp.*, *Curvularia spp.*, and *Aspergillus niger* (table 6).

Table 1: Age-wise distribution of dermatophytes

Age in years	Male	Female	Number of cases
<10	01(100%)	00(00%)	01 (1.54%)
11-20	02(66.67%)	01(33.33%)	03(4.62%)
21-30	06(60%)	04(40%)	10(15.38%)
31-40	15(57.69%)	11(42.31%)	26(40%)
41-50	09(64.29%)	05(35.71%)	14(21.54%)
51-60	04(44.44%)	05(55.56%)	09(13.85%)
61-70	01(50%)	01(50%)	02(3.08%)
Total	38(58.46%)	27(41.54%)	65(100%)

M: F=1.4:1

Table 2: Occupation details of the study population

Occupation	Number	%
Agricultural worker	30	46.15
Housewife	15	23.08
Students	07	10.77
Office workers	06	9.23
Others	07	10.77
Total	65	100

Table 3: The clinical pattern of onychomycosis

Clinical pattern	Fingernails	Toenails	Both	Total (%)
DLSO	13	21	07	41(63.08)
PSO	04	08	02	14(21.54)
SWO	-	01	-	01(1.54)
TDO	02	06	01	09(13.85)
Total	19(29.23%)	36 (55.38%)	10 (15.38%)	65 (100)

DLSO-Distal and lateral subungual onychomycosis, PSO-Proximal subungual onychomycosis, SWO-Superficial white onychomycosis, TDO-Total Dystrophic onychomycosis

Table 4: KOH and culture findings

	KOH and/or culture+ve	KOH and culture+ve	KOH+ve culture-ve	KOH-ve culture+ve	KOH and culture-ve
Number of cases	45	33	09	02	21
Percentage	67.69%	50.77%	13.85%	3.08%	32.31%

Table 5: Correlation of direct microscopy (KOH) findings with culture

	Culture positive	Culture negative	Total
KOH Positive	33	09	42
KOH Negative	02	21	23
Total	35	30	65

Sensitivity = 94.29%, Specificity = 70%, p value < 0.00001

Table 6: Culture isolates in the study population

Fungal isolate (n=35)			
	Dermatophytes (n=25, 71.43%)		Non-Dermatophytes (n=10, 28.57%)
<i>Trichophyton rubrum</i>	12(48%)		<i>Alternaria spp.</i> 02(20%)
<i>Trichophyton mentagrophyte</i>	09(36%)		<i>Curvularia spp.</i> 02(20%)
<i>Trichophyton tonsurans</i>	03(12%)		<i>Aspergillus niger</i> 02(20%)
<i>Epidermophyton floccosum</i>	01(04%)		<i>Candida spp.</i> 04(40%)
Total	25(100%)		10(100%)

DISCUSSION

In our study, 65 clinically suspected cases of onychomycosis visiting the Dermatology, Venereology and Leprosy outpatient department of People's Hospital, Bhopal, were studied. In this study, males (58.46%) were more affected than females, the ratio being 1.4:1. These findings are comparable with the other similar studies by Adhikari L *et al.*, Vinod S, Grover S whereas Jesudanam TM *et al.* have reported a slightly higher prevalence in females. Males are probably more affected due to their frequent indulgence in outdoor work, thereby increasing chances of exposure [10-13].

The age group of 31-40 y was most commonly affected with 26 cases (40%) followed by the age group of 41-50 y with 14 cases (21.54%) which was found similar to studies done by Vinod S, Grover S and Veer P *et al.* [11, 12, 14].

In our study, onychomycosis was most common in agricultural workers (46.15%) followed by housewives (23.08%), which was comparable to the study conducted by Niranjana HP *et al.* [15]. This may be due to increased chances of occupation-related trauma in agricultural workers and the involvement of housewives in household wet work.

In our study, distal and lateral subungual onychomycosis (DLSO) was the commonest clinical pattern (63.08%) and the toenails were more frequently involved (55.38%) than fingernails. Similar findings were reported by Beena *et al.* and Lone B *et al.* [16, 17].

In our study diagnostic utility of KOH mount for laboratory diagnosis of onychomycosis was found to be highly significant with a p-value of < 0.00001 when compared to the gold standard i.e. culture.

In our study, most of the fungal isolates (71.43%) turned out to be dermatophytes, with *Trichophyton rubrum* (48%) as the most common isolate, followed by *Trichophyton mentagrophyte* (36%). These findings were similar to studies done by Veer P *et al.* and Niranjana HP *et al.* [14, 15].

Among non-dermatophytes, *Candida spp.* (40%) was the most common isolate, followed by 2 isolates (20%) each of *Alternaria spp.*, *Curvularia spp.*, and *Aspergillus niger*. Vijaya D *et al.* and Gupta M *et al.* [18, 19] also reported *Candida spp.* as the most common non-dermatophyte isolate, while Veer P *et al.* [14] reported *Aspergillus spp.* as the most common non-dermatophyte isolate.

CONCLUSION

In our study, distal and lateral subungual onychomycosis (DLSO) was the commonest clinical presentation. Dermatophytes were the most common aetiological agent of onychomycosis in our study and

Trichophyton rubrum was the most common isolate. This study also reveals the fact that nowadays, non-dermatophytes are increasingly isolated from cases of onychomycosis.

LIMITATIONS OF THE STUDY

Our study is a hospital-based study therefore clinico-mycological profile may vary from the general population. Many of the patients in the general population may not seek medical advice. There is a great need for a carefully conducted population-based prevalence study.

FUNDING

Nil

AUTHORS CONTRIBUTIONS

Concept, design, and defining intellectual content were attributed to Dr. G. Saxena. Literature search and data acquisition was done by Dr. G. Saxena and Dr. K. Sadawarte. Data analysis and interpretation was done by Dr. M. Kulmi and Dr. A. Mehta. The manuscript was prepared by Dr. G. Saxena and Dr. K. Sadawarte. Manuscript Editing and review were done by Dr. M. Kulmi and Dr. A. Mehta. All authors read and approved the final version of the manuscript.

CONFLICT OF INTERESTS

The Authors declare no conflict of interest.

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