

Original Research Article

Onychomycosis: dermatophytes to yeasts: an experience in and around Mumbai, Maharashtra, India

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ABSTRACT

Background: Onychomycosis is a fungal nail infection having wide range of prevalence in different geographical regions. It becomes imperative to know prevalent causative agent in local area to improve quality of life of patients. Present study was aimed to study epidemiological, clinical and laboratory aspects of onychomycosis

Methods: Study was carried out prospectively at a tertiary care teaching hospital. Nail scrapings of 630 clinically suspected cases of onychomycosis over a period of 5 years (January 2012 to December 2016) were subjected to KOH examination and culture.

Results: Young adults in age group of 21-40 years (67.61%) were predominantly affected with male to female ratio of 1.8:1. Overall prevalence of onychomycosis of the present study is 58.41%. Yeasts were isolated in 47.86%, dermatophytes in 30.71% and non dermatophytic filamentous fungi in 21.43%

Conclusions: Present study demonstrates a shift in causative agents from dermatophytes to yeasts.

Keywords: Dermatophytes, Non dermatophytic filamentous fungi, Onychomycosis, Yeasts

INTRODUCTION

Onychomycosis is a fungal infection of nails which can be caused by dermatophytes, yeasts or non-dermatophytic moulds.¹ It is responsible for up to 50% of all nail diseases and 30% of all fungal infections.²

Onychomycosis has varied worldwide distribution ranging from 2% to 50%.³ Various risk factors associated with this condition are reduced peripheral circulation, diabetes, nail trauma, difficulty to maintain proper nail hygiene, chronic smoking, communal bathing etc. Clinically, onychomycosis is classified as distal lateral subungual onychomycosis, proximal subungual onychomycosis, white superficial onychomycosis and total dystrophic onychomycosis.⁴ Onychomycosis is not a life threatening condition. It causes cosmetic problem to the patient. The infected nail may serve as a chronic reservoir, giving rise to repeated mycotic infection thus

posing an important public health problem.⁵ It can have significant negative effects on patient's emotional, social, occupational functioning.

There are many skin conditions like psoriasis, lichen planus, onychogryphosis which may be clinically confused with onychomycosis. Therefore, it is necessary to have a clinical suspicion and correct laboratory diagnosis of this condition before starting treatment for better results.

Species identification is of paramount importance as the clinical outcome of antifungal agents varies as to whether the etiological agent is yeast, dermatophyte, or other mould. There is paucity of such data available in tropical country like India. Prevalence of regional onychomycosis is not available. Hence this study was conducted to know epidemiological, clinical and laboratory aspects of onychomycosis.

METHODS

After obtaining institutional ethics committee permission, study was carried out prospectively over a period of 5 years i.e. January 2012 to December 2016. Suspected cases of onychomycosis referred to mycology division of a tertiary care hospital were included in the study. Detailed clinical history and examination findings were noted. Patients on antifungal agents were excluded from the present study. Specimen collection was done from most severely affected nail. Nail scrapings were collected on filter paper after cleaning the affected nail with 70% alcohol to remove contaminants.

All specimens were subjected to microscopy and culture. Specimen was kept in 10% KOH for a period of 24 hours to dissolve the keratin and examined microscopically for presence of fungal elements.⁴ Specimens were inoculated onto Sabouraud's dextrose agar with antibiotic, Sabouraud's dextrose agar with antibiotic and 5% cycloheximide and Dermatophyte test medium (DTM) and were incubated in a B.O.D. incubator at 30°C for 4 weeks. The pathogenic filamentous organisms were identified by gross morphology, microscopic examination with lactophenol cotton blue preparation and slide culture. If a dermatophyte was isolated, it was considered as a pathogen. Nondermatophytic mould or yeast was considered significant if they were isolated repeatedly in pure culture in atleast three culture tubes. Yeast like growth was further speciated by use of germ tube, urease, corn meal agar and chrome agar inoculation.

RESULTS

630 clinically suspected cases of onychomycosis were included in the study. 405 (64.28%) were males and 225 (35.72%) were females with male to female ratio of 1.8:1. Young adults were most commonly affected in the age group 21 to 30 years (36.98%) followed by 31 to 40 years (30.63%) (Table 1).

Table 1: Distribution of patients of onychomycosis according to age and gender.

Age group (years)	Male	Female	Total (Percentage)
0-10	0	0	0
11-20	27	10	37 (5.87%)
21-30	155	78	233 (36.98%)
31-40	121	72	193 (30.63%)
41-50	70	40	110 (17.46%)
51-60	20	15	35 (5.56%)
61-70	10	8	18 (2.86%)
> 70	2	2	4 (0.63%)
Total	405	225	630

When occupational history was analyzed, it was observed that there were 55.87% farmers, 19.04% housewives, 12.69% laborers, 3.65% students and remaining 8.75%

were doctors, office workers, and retired persons. Fingernails were affected in 440 (69.84%) patients while in 125 (19.84%) patients toenails were affected. In 65 (10.32%) patients, both the finger and toenails were affected. Distal lateral subungual onychomycosis was the most common manifestation in 320 (50.79%) cases followed by proximal subungual onychomycosis (26.34%), white superficial onychomycosis (17.14%) and total dystrophic onychomycosis (5.72%). Of 630 clinically suspected cases, 368 (58.41%) were confirmed either by direct microscopy and/or culture (Table 2). Direct microscopy could identify 274 cases (43.49%) whereas 280 (44.46%) cases were confirmed by culture. 186 (29.52%) cases were positive by both microscopy and culture.

Table 2: Comparison of microscopy and culture in clinically suspected cases of onychomycosis.

KOH	Culture		Total
	Positive	Negative	
Positive	186 (29.52%)	88 (13.96%)	274 (43.49%)
Negative	94 (14.92%)	262 (41.58%)	356 (56.51%)
Total	280 (44.46%)	350 (55.54%)	630

Among the 280 isolates, yeasts (47.86%) were predominant followed by dermatophytes (30.71%) and non-dermatophyte filamentous fungi (21.43%) (Table 3). Of the yeast isolates, isolation of non-albicans candida (25.71%) was more than *C.albicans* (20%). Non albicans candida isolated were *C.parapsillosis* (13.57%), *C.tropicalis* (11.43%) and *C.kefyr* (0.71%)

T.rubrum (15.36%) accounted for majority cases of dermatophytic onychomycosis followed by other trichophyton species namely *T. mentagrophyte*, *T.tonsurans*, *T. verrucosum*, *T. violaceum*. There were few cases of *M. ferrugineum* (0.71%) and *E. floccosum* (0.71%). Among the non-dermatophyte filamentous fungi *Fusarium spp* (10%) was the most common isolate followed by *Aspergillus spp* (5%). Phaeoid fungi namely *Cladosporium* (2.86%), *Curvularia* (2.14%), *Alternaria* (0.71%) were also isolated.

DISCUSSION

Onychomycosis is a chronic infection of nails affecting quality of life. Though this condition has got poor attention in earlier days due to less awareness in community, its prevalence is now increasing with wide spectrum of causative agents.

The disease occurs worldwide with varying prevalence in different geographical areas. In this study, prevalence of onychomycosis is 58.41%. In India, different studies reported prevalence ranging from 37.78% to 54.5%.⁶⁻⁹

It has been reported to involve both the sexes of all age groups. In our study, maximum numbers of patients having onychomycosis were in the age group of 21 to 40 years (67.61%) (Table 1). Patients of this age group are more prone to trauma because of their outdoor activities making their nails more exposed to fungi. Alternately, young population may be more sensitive towards cosmetic damage of nails and approach clinicians on time. In the present study, there were no patients below age group of 10 years. This could be attributed to size of

nail bed and rate of growth of nail plate. The small size of nail bed provides lesser area for fungal invasion, whereas increased growth rate of nail plate in children helps to the elimination of fungi. This finding correlates well with other studies.^{8,10,11} Few authors have reported higher prevalence of onychomycosis in elderly age group.^{12, 13} But in our study only 9.05% of patients above age of 60 years were affected. This may be due to ignorance about the disease in this age group and partly due to asymptomatic nature of this condition.

Table 3: Isolation of fungi over period of 5 years (2012-2016).

Species	No of isolates					Total	Percentage
	2012	2013	2014	2015	2016		
All yeast and yeast like fungi							
<i>C. albicans</i>	12	6	18	14	6	56	20.00%
<i>C. tropicalis</i>	4	6	10	6	6	32	11.43%
<i>C. parapsillosis</i>	0	4	12	4	18	38	13.57%
<i>C. kefyr</i>	0	0	2	0	0	2	0.71%
<i>Tricosporoon spp</i>	0	0	2	2	2	6	2.14%
Total	16	16	44	26	32	134	47.86%
Dermatophytes							
<i>T. rubrum</i>	4	6	5	12	16	43	15.36%
<i>T. mentagrophyte</i>	6	6	4	4	2	22	7.86%
<i>T. tonsurans</i>	3	1	1	1	2	8	2.86%
<i>T. verrucosum</i>	1	0	2	0	0	3	1.07%
<i>T. violaceum</i>	1	0	2	2	1	6	2.14%
<i>M. ferrugineum</i>	0	0	2	0	0	2	0.71%
<i>E. floccosum</i>	0	0	2	0	0	2	0.71%
Total	15	13	18	19	21	86	30.71%
Non dermatophytic filamentous fungi							
<i>Fusarium spp</i>	12	6	4	0	6	28	10.00%
<i>Aspergillus spp</i>	4	2	4	2	2	14	5.00%
<i>Paecilomyces spp</i>	0	0	0	0	2	2	0.71%
<i>Cladosporium</i>	0	0	4	0	4	8	2.86%
<i>Curvularia</i>	0	0	4	2	0	6	2.14%
<i>Alternaria</i>	0	0	2	0	0	2	0.71%
Total	16	8	18	4	14	60	21.43%

It is said that onychomycosis is a disease of women because of their more involvement in wet work.^{5, 6} However, in our study, males were found to be more affected than females (Table 1). This male preponderance could be due to their increased outdoor activities, type of occupation and increased use of ill fitting foot wares. This makes them more vulnerable to trauma and subsequent entry of fungi. This finding is similar with other studies.¹⁴ Some authors have postulated that this gender difference may be due to differences in hormone levels that result in a different capacity to inhibit the growth of dermatophytes.¹⁴⁻¹⁷

We observed that fingernails (69.84%) were most commonly affected site followed by toenails (19.84%). In 65 (10.32%) patients both the finger and toenails were

affected. This finding is similar to that reported by other authors.^{9,10}

Distal lateral subungual onychomycosis (50.79%) was the most common pattern followed by other clinical patterns namely proximal subungual onychomycosis (26.34%), white superficial onychomycosis (17.14%) and total dystrophic onychomycosis (5.72%). K. Narotham Reddy et al, Kaur R, et al have also reported similar findings.^{10,18}

Microscopic examination with KOH and culture are the two laboratory tools used to confirm onychomycosis. In this study, direct microscopy using KOH could identify 43.49% cases whereas 44.46% cases were confirmed by culture. 29.52% cases were positive by both microscopy

and culture (Table 2). In 14.92% culture positive cases, KOH was negative. This might be due to difference in sensitivity of these methods. Sensitivity of both these methods depends upon various factors right from method of sampling, preparation of sample up to final interpretation of result.¹⁹ Hence both the tools are complimentary to each other. Use of both these methods is advocated in routine diagnostic mycology laboratories.

Dermatophytes are said to be the most common fungi associated with onychomycosis.^{19,20} They have been responsible for 90% of toe nail and 50% of finger nail infections.¹⁸ Few workers have reported equal incidence between dermatophytes and yeasts.²¹ Scenario is changing from dermatophytes to yeasts which is the case even in our study. In our study yeasts (47.86%) are the predominant pathogen followed by dermatophytes (30.71%) and non-dermatophyte filamentous fungi (21.43%). Similar findings were shown by other researchers.^{6,13}

Overall prevalence of *C.albicans* in our study was 20% whereas that of non-albicans was 25.71%. The prevalence of *C.albicans* showed steady decline over a period of 5 years (2011 to 2015). Among non albicans candida, the prevalence of *C.parapsilosis* was 13.57% and *C.tropicalis* was 11.43%. There was uniform increase in number of isolates of *C.parapsilosis* over 5 years showing shift from *C. albicans* to non-albicans candida mainly *C.parapsilosis*. Our finding is in contrast with the findings of Gelotar et al, M Gerami shoar et al who have not reported any shift from *C. albicans* to non-albicans candida.^{6, 13}

Onychomycosis due to candida is a sign of immune suppression.²² Systemic treatments with itraconazole or fluconazole is usually effective against all candida species and not much resistance has been reported from candida isolates of onychomycosis.²² Hence with changing trends and a shift to non-albicans candida, antifungal susceptibility testing should be recommended. More studies will be required to analyze resistance pattern.

Among dermatophytes isolated, *T.rubrum* (15.36%) was the most common isolate followed by *T.mentagrophyte* (7.86%). Some workers have shown *T.rubrum* as most common agent while some found *T.mentagrophyte* as the most common pathogen.^{7, 9, 10} Thus there may be a change in the species of dermatophytes with respect to geographical area. There were two cases of *E. floccosum* in our study. Interestingly both these isolates were seen from health care workers. Therefore more studies are needed to enlighten association of *E. floccosum* to health care workers.

Isolation rate of non-dermatophytic filamentous fungi was 21.43% of which *Fusarium* (10%) was the predominant isolate. M. Leelavathi et al, *Shrijana Gurung et al* have also reported similar findings in their

study.^{12, 23} It is interesting to note that dematiaceous fungi namely *Cladosporium* (2.86%), *Curvularia* (2.14%), *Alternaria* (0.71%) have been isolated in our study. Thus, role of these nondermatophytic fungi as a cause of onychomycosis should not be ignored.

Due to presence of periungual inflammation, non-dermatophytic onychomycosis can be clinically suspected. Treatment with only systemic antifungals is effective in cases of *Aspergillus* infection whereas *Fusarium* infections are difficult to eradicate. Their treatment should always be associated with topical treatment and avulsion of affected nail along with systemic antifungals.^{24, 25} Hence laboratory confirmation of these fungi is a must for betterment of patients.

CONCLUSION

To conclude, it is observed that young adults were most commonly affected owing to their outdoor activities. Commonly affected site was fingernail with distal lateral subungual onychomycosis being usual clinical pattern. There is change in causative agent over a period of time from dermatophytes to yeasts. *C.parapsilosis* and *C.albicans* have emerged as leading cause of onychomycosis.

Therefore, though onychomycosis does not impose serious public health problem, the etiological diagnosis this condition is imperative to improve quality of life.

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