Current mycological profile of onychomycosis in Kashmir valley: A hospital-based study

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Abstract:
INTRODUCTION: Onychomycosis is a common nail ailment associated with significant physical and psychological morbidity. It is caused by dermatophytes, nondermatophytic molds, and yeasts. Increased prevalence in the recent years is attributed to enhanced longevity, comorbid conditions such as diabetes, avid sports participation, and emergence of HIV.

AIMS AND OBJECTIVES: This study was undertaken to identify the mycological profile and etiological agents associated with this infection.

MATERIALS AND METHODS: One hundred and twenty-nine cases of clinically suspected onychomycosis were included in this study. Clinical patterns were noted, and samples collected from most severely affected nail. They were subjected to direct microscopy and culture.

RESULTS: The microscopic and/or culture examination was positive in 64.34% of cases. The most common clinical type was distal lateral subungual onychomycosis in 42 (50.60%) Candida sp. was the most common fungal agent among yeasts, followed by Trichophyton sp among dermatophytes and Aspergillus among nondermatophytic molds. Females were affected more than males and fingernails were affected more frequently than toenails.

CONCLUSION: Onychomycosis is a significant and important disease which can cause many physical, physiological, and occupational problems, considerably impairing patient quality of life. Hence, fungal cultures are of paramount importance in all suspected cases of onychomycosis because the antifungal agent with an appropriate spectrum of activity can only be used if the underlying fungal pathogen is identified correctly.

Key words: Candida sp., dermatophytes, nails, nondermatophytic molds, onychomycosis

Introduction

Onychomycosis is the most common infective disorder of nails and is responsible for about 50% of all consultations for nail disorders.[1] Onychomycosis results from invasion of the nail plate by dermatophytes, yeasts, or mold species of Fungi and represents about 30% of mycotic cutaneous infections.[2-4]

Onychomycosis affects approximately 5% of the population worldwide.[9] However, in India, various workers have reported the incidence to vary from 0.5% to 12% in the general population.[6,7] The prevalence of onychomycosis seems to vary across the world because of various socioeconomic and cultural factors. Several factors such as reduced peripheral circulation, diabetes, nail trauma, difficulty to maintain proper nail hygiene, chronic smoking, and communal bathing have been implicated in the increase in the prevalence rates of onychomycosis.

Onychomycosis is the most difficult to treat superficial mycosis. It is a chronic infection that is prone to relapse as infected nails serve as a chronic reservoir of infection, which can cause mycotic infection of skin.[8] Onychomycosis is classified into five clinical types, according to the fungal invasion of the nail: Distal and/or lateral subungual onychomycosis (DSLO), white superficial onychomycosis (WSO), proximal subungual onychomycosis (PSO), total dystrophic onychomycosis, and candida onychomycosis.[9]

Increasingly, onychomycosis is being viewed as more than a mere cosmetic problem. In spite of improved personal hygiene and living environment, onychomycosis continues to spread and persist.[2] Therefore, it is important...
to identify the causative agent to ensure that the appropriate treatment is employed for each case.\(^1\)

This study seeks to improve the current knowledge of the mycological etiologies of onychomycosis in Kashmir valley.

**Materials and Methods**

**Study population**

This prospective study was conducted over a period of 1 year, from November 2014 to October 2015, on 129 patients with clinically suspected nail infections, who attended the Dermatology Out-Patients Department of SMHS Hospital, Srinagar, Kashmir, India.

**Inclusion criteria**

All patients with a clinical diagnosis of onychomycosis were included in the study. The samples of patients who had taken antifungal drugs were excluded from the study. A detailed history of patients was taken. The clinical pattern and location of disease were also documented.

**Sample collection**

The specimens were collected for microbiological analysis, on the basis of the results of clinical evaluations. The selected nail was cleaned with 70% alcohol to remove contaminants. As sites of invasion and localization differ in different varieties of onychomycosis, separate approaches were taken to collect the nail specimens.

**Processing of samples**

- **Step 1:** The sampled material was divided into two portions: One for direct microscopy and the remainder for culture. Direct microscopy of all specimens was done with the help of 20% potassium hydroxide (KOH) solution to determine the presence of any fungal elements such as fungal hyphae, arthrospore, or yeast forms.
- **Step 2:** All samples were inoculated on each of two isolation media: (1) Sabouraud dextrose agar (SDA, HiMedia Laboratories) with 5% chloramphenicol and (2) SDA with 5% chloramphenicol and cycloheximide. The culture tubes were incubated at 25°C and 37°C and examined biweekly for 6 weeks.
- **Step 3:** The pathogenic organisms were identified by [Figure 1] gross morphology and microscopic examination with KOH and lactophenol cotton blue preparation [Figure 2]. If a dermatophyte was isolated in culture, it was taken as a pathogen. If a nondermatophytic mold or yeast was isolated, it was considered to be significant. If they were isolated repeatedly in pure culture (three times) on two media and with a positive KOH finding. To identify Candida albicans, we did germ tube test and Corn Meal Agar inoculation. We also did urease test to differentiate Trichophyton species.

**Results**

A total of 129 (58 male and 71 female) patients were examined. Among the 129 cases included in our study, it was observed that a total of 83 (64.34%) clinically suspected onychomycosis patients were confirmed either by direct microscopy and/or culture, 85 (65.89%) samples were positive by KOH, whereas 76 (58.91%) samples were positive by culture. Nearly 47 (36.43%) samples were positive by both the methods and 27 (20.93%) samples were KOH negative but culture positive [Table 1].

While analyzing the prevalence of onychomycosis in different age groups, it was observed that the young adults ranging from 31 to 40 years (28.91%) and from 21 to 30 years (26.50%) were most commonly affected followed by 11–20 years (24.09%). The lowest affected age was 4 years, whereas the highest affected age was 65 years [Graph 1].

Furthermore, in our study, females were affected (55.42%) more than males (44.57%).

Fingernails were most commonly affected site in 86.74% cases whereas toenails were affected in 27.71% patients. Both the finger and toenails were affected in 33.73% patients [Graph 2].

DLSO was the most common pattern in 42 (50.60%) found in our study. Other clinical patterns found were PSO in 23 (27.71%), followed by WSO in 9 (10.84%) and chronic paronychia in 9 (10.84%).

Among the culture-positive cases, the most common Fungi isolated [Table 2] in our study were Candida species (55.42%), followed by dermatophytes (40.96%) and nondermatophytic molds (3.61%). Of the Candida species isolated in the study, maximum were C. albicans (54.34%), followed by Candida krusei (23.91%) and Candida tropicalis (21.73%). While speciating the dermatophytes, it was found that Trichophyton mentagrophytes was the most common isolate (47.05%), followed by Trichophyton rubrum (38.23%), Trichophyton tonsurans (3.61%), and Epidermophyton sp (2.94%). The nondermatophytic molds isolated in our study were Aspergillus spp. (3.61%) and Scopulariopsis (1.20%).

**Table 1: Correlation of direct microscopy with culture (n=129)**

| Test procedure                        | n (%) |
|---------------------------------------|-------|
| KOH positive and/or culture positive  | 83 (64.34) |
| KOH positive culture negative         | 85 (65.89) |
| KOH negative culture positive         | 27 (20.93) |
| Culture positive                      | 76 (58.91) |
| Both positive                         | 47 (36.43) |

KOH = Potassium hydroxide

Graph 1: Figure showing the percentage distribution of age groups having onychomycosis infections
The distribution of the different clinical types of onychomycosis with occupation type was also observed [Table 3]. Homemakers showed the highest rate of onychomycosis (31.32%), and candida was isolated from their nail samples. This was followed by office workers and students (19.27%) each and farmers and/or laborers (13.25%).

**Discussion**

Onychomycosis is a chronic infection of the nails and is considered to be a serious public health problem. The worldwide incidence of onychomycosis is increasing due to a number of factors such as diabetes, poor peripheral circulation, increase in the number of persons who are immunocompromised, increase in avid sports use of health clubs, communal swimming pools, and occlusive footwear for exercise.[10] However, the data regarding the etiology of onychomycosis and its prevalence in particular geographical areas are lacking. In the present study, an attempt was made to understand the etiology and distribution of onychomycosis in a population of Kashmir Valley.

In our study, 129 clinical cases of onychomycosis were included in the study. Out of these, a total of 83 (64.34%) cases were confirmed either by direct microscopy and/or culture. These results are in corroboration with previous studies where both the methods have been shown to be useful in diagnosis.[2,11] We observed direct microscopic examination (65.89%) to be more sensitive than culture positivity (58.91%). However, culture positivity helps in the identification of genus and species of different Fungi and remains the gold standard. Weinberg et al. reported that the sensitivity of KOH and culture in detecting positive infections were 80% and 59%, respectively.[12] The study conducted by Shenoy et al. showed that KOH and culture had 64% and 42% sensitivity in detecting positive infections.[13]

Furthermore, in our study, females were affected (55.42%) more than males (44.57%), the ratio being 1.2:1. Homemakers (31.32%) were affected more, possibly because of the constant submerging of their hands in water during household chores.[14-16]

It was observed in our study that the young adults ranging from 21 to 40 years (55.41%) were most commonly affected followed by 11–20 years (24.09%). The lowest affected age was

| Table 2: Etiologic agents obtained from nail samples |
|-----------------------------------------------|
| **Category of fungus** | **Species** | **Male (n)** | **Female (n)** | **Total (%)** |
|------------------------|-------------|--------------|----------------|---------------|
| Candida sp.            | Candida albicans | 7            | 18             | 25 (54.34)    |
|                        | Candida krusei | 4            | 7              | 11 (23.91)    |
|                        | Candida tropicalis | 3         | 7              | 10 (21.73)    |
| Dermatophytes          | Trichophyton mentagrophytes | 10      | 6              | 16 (47.05)    |
|                        | Trichophyton rubrum | 8         | 5              | 13 (38.23)    |
|                        | Trichophyton tonsurans | 1        | 2              | 3 (3.61)      |
|                        | Epidermophyton sp.   | 1           | 1              | 2 (2.94)      |
| Nondermatophyte molds  | Alternaria sp.       | 1           | 1              | 2 (3.61)      |
|                        | Scopulariosis        | 1           | 0              | 1 (1.20)      |

| Table 3: Distribution of onychomycosis with occupation |
|-----------------------------------------------|
| **Occupation** | **Males** | **Females** | **Total (%)** |
|----------------|-----------|-------------|---------------|
| Homemakers    | 0         | 26          | 26 (31.32)    |
| Office workers| 12        | 4           | 16 (19.27)    |
| Students      | 10        | 6           | 16 (19.27)    |
| Farmers and laborers | 9     | 2           | 11            |
| Miscellaneous | 6         | 8           | 15            |

Graph 2: Sites of involvement of onychomycosis

Figure 1: KOH mount with plenty of hyphae (×40)

Figure 2: LCB with hyphae lined with microconidia (×40)
4 years, whereas the highest affected age was 65 years. High prevalence of onychomycosis during the third decade has also been found in other studies.[17,18] The increase in cases with age may be justified by repeated nail microtrauma, due to more prolonged exposure to pathogenic Fungi, as well as greater work activity and venous insufficiency.[19] Furthermore, they may also be more conscious cosmically with age.[20]

In our study, the predominant pathogens were yeasts (64.71%), which included 54.34% cases of C. albicans, followed by C. krusei (23.91%) and C. tropicalis (21.73%). This may be due to the repeated contact with water, which is the common mode of transmission of Candida spp. Dermatophytes were present in 17.65% cases, among which T. mentagrophytes was the most common isolate (47.05%), followed by T. rubrum (38.23%), T. tonsurans (3.61%), and Epidermophyton sp (2.94%). The nondermatophytic molds isolated in our study were Aspergillus spp. (3.61%) and Scopulariopsis (1.20%). These findings are in accordance with the results of the work of other authors also.[14,21,22] However, these findings are in contrast with many studies which have demonstrated a greater prevalence of dermatophytes as the etiological agents of onychomycosis.[16,23] Although some studies have reported T. rubrum as the most common dermatophyte,[23] our finding is in concordance with many other studies which found T. mentagrophytes as the most common dermatophyte responsible for onychomycosis.[20] Such variations may be related to local environmental conditions.

DLSO was the most common clinical presentation of onychomycosis in our patients (50.60%) which is in agreement with many previous studies.[25-27]

Fingernails were affected more often than toenails in females, which can be explained by the work habits of such patients who generally work performing domestic chores with their hands remaining wet for most of the day.[28] The increased prevalence of toenail infection in males could be the result of more traumas in the nails and the more common use of occlusive footwear. Finger nails were also more frequently affected than toenails in many other studies.[20,23]

Conclusion

This study shows that onychomycosis is potentially a significant problem and can no longer be considered a simple cosmetic nuisance confined to the nails. It is important to undertake a complete mycological study (KOH and culture) in patients with suspicion of onychomycosis. Furthermore, since the pattern of onychomycosis observed in any geographical area changes with time, it is necessary to conduct periodic studies to obtain information regarding the prevalence of the causative agents of onychomycosis.

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Conflicts of interest

There are no conflicts of interest.

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