



## An Epidemiological Study of Onychomycosis in Kashmir Valley

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### Authors' contributions

This work was carried out in collaboration between all authors. Author FK carried out the study performed the statistical analysis and managed literature searches. Authors TW and GB designed the study, wrote the protocol and the first draft of the manuscript. Authors SM, IHS and GM managed the analyses of the study and literature searches. Author NB wrote the final draft of the paper and managed the literature searches. All authors read and approved the final manuscript.

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### ABSTRACT

**Background:** Onychomycosis is a common fungal infection of the nails that apart from causing disfigurement of the nails acts as a source of other fungal infections. Prompt diagnosis along with appropriate antifungal therapy can help prevent such complications.

**Aims:** To find out the prevalence and fungal aetiology of onychomycosis in patients presenting to the Department of Dermatology Sher-i-Kashmir Institute of Medical Sciences Medical College/Hospital Srinagar.

**Settings and Design:** This prospective study was carried out in the Department of Microbiology, Sher-i-Kashmir Institute of Medical Sciences, Srinagar in collaboration with the Department of Dermatology Sher-i- of Kashmir Institute of Medical Sciences College, Bemina for a period of two

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years (August 2010 to September 2012).

**Materials and Methods:** A total of 300 samples from patients attending the Dermatology OPD with features suggestive of onychomycosis were included in this study. Microscopy of the samples was done in 20% potassium hydroxide (KOH) and culture on Sabouraud dextrose agar (SDA) with chloramphenicol and SDA with chloramphenicol and cyclohexamide. Tubes were incubated at 37°C and 25°C for 4-6 weeks and examined biweekly for growth. Microscopic analysis of the growth was done using lactophenol cotton blue (LCB).

**Results:** Age of cases ranged from 3 months to 85 years with female preponderance. Distal and lateral subungual onychomycosis (DLSO) was the most common clinical type. Overall, positivity of microscopy was 73.5% and of culture was 29%. Sensitivity and specificity of KOH as compared to culture was 83.9% and 30.3% respectively. Among the fungi isolated, 98% were dermatophytes and 2% were non-dermatophytic fungi (*Fusarium spp.*). Commonest dermatophytes isolated were *Trichophyton rubrum* followed by *Trichophyton violaceum*.

**Conclusion:** KOH and culture are complementary to each other and should be included in the diagnostic evaluation of onychomycosis which is commoner in our population.

*Keywords: Dermatophytes; KOH; onychomycosis.*

## 1. INTRODUCTION

Onychomycosis refers to the fungal infection of the fingernails or toenails, caused by dermatophytes, yeast and non-dermatophyte molds that lead to distortion, discoloration, thickening and detachment from the nail bed [1]. It accounts for up to 30% of all superficial infections of skin and constitutes about a half of all nail abnormalities; affecting approximately 5% of population worldwide [2,3]. The prevalence of onychomycosis is determined by age, social class, occupation, climate, living environment and frequency of travel. The infection is more common in adult males (particularly elderly > 60 yrs of age), diabetics, immunocompromised individuals (e.g. HIV positive), individuals with peripheral vascular (arterial) disease, previous tinea pedis infection, history of trauma to the nail, or those with a family history of onychomycosis [1,3-5]. The disease affects toenails more frequently than fingernails due to slower rate of growth of the former, reduced blood supply and usual confinement in dark moist environments [4,5].

Onychomycosis can be divided into four major clinical types on the basis of their presenting clinical features; distal and lateral subungual onychomycosis (DLSO), proximal subungual onychomycosis (PSO), white superficial onychomycosis (WSO) and total dystrophic onychomycosis (TDO) [5-7]. Among these, DLSO is the most common form in which patients present to the clinician [7]. Accurate diagnosis is critical for appropriate treatment and requires identification of physical changes and positive laboratory analysis. As only 50% of nail problems

can be attributed to onychomycosis; clinical diagnosis by physical examination alone can be inaccurate. In addition conditions that mimic onychomycosis like lichen planus, psoriasis etc. need to be ruled out [6]. Various techniques have been used to accurately diagnose onychomycosis, with microscopy and culture being the most frequently used. When the index of suspicion is high, all specimens should be cultured, even if the KOH preparation is negative [1]. Needless to mention that clean healthy nails are important and dystrophic nails can be a social impediment causing significant embarrassment that affects patient's self-esteem. In addition, thickened nails can be painful, interfere with the function of the nail unit and may cause discomfort in walking, standing and exercising [6].

Although fungal infections of the nail are common dermatological presentation, the prevalence of onychomycosis remains largely unknown as no microbiological studies have been done in this part of the country till date. The present study was carried out to ascertain the prevalence, fungal aetiology and clinical characteristics of onychomycosis in patients presenting to the Department of Dermatology SKIMS Medical College/ Hospital Srinagar.

## 2. MATERIALS AND METHODS

This prospective study was conducted in the Department of Microbiology, Sher-i-Kashmir Institute of Medical Sciences (SKIMS), Soura Srinagar in collaboration with the Department of Dermatology SKIMS Medical College, Bemina from August 2010 till September 2012.

A total of 300 patients attending the dermatology OPD at SKIMS MC with features suggestive of fungal infection of nails were included in the study. A detailed history was taken and proforma separately filed for each patient that included history of any other cutaneous fungal infections and systemic diseases like hypertension, diabetes, peripheral vascular disease, thyroid disease and any immunodeficiency disorder. Apart from this, a detailed examination of nails was conducted that included site of involvement (fingernails and/or toenails) and type of nail changes [clinical pattern]. Written consent was taken from all the patients. Ethical approval for the study was sought from the institute's ethical clearance committee.

## 2.1 Collection, Transport and Processing of Specimens

Sterile scalpel/serrated curette were used to collect nail scrapings, clippings and curetting's after the area was cleaned with 70% alcohol. The specimen was placed in small dark brown pill packets and processed in the Mycology section in the Department of Microbiology, SKIMS. In patients of DLSO, the abnormal nail was clipped proximally and the nail bed and underside of the nail plate were scraped with a 1-2 mm serrated curette. For patients affected with PSO, abnormal nail was clipped proximally and underside of nail plate and nail bed were scraped. In patients of WSO, white spots on the nail were scraped. In patients suffering from TDO, any abnormal area of the nail plate or bed was clipped.

## 2.2 Microscopy and Culture

The sampled material was divided into two portions one for direct microscopy and the other for culture. Direct microscopy was done using a solution of 20% potassium hydroxide (KOH) to establish the presence or absence of fungal hyphae. For culture, specimen collected in dark colored pill packets was reduced in size to approximately 1 mm. Media used were Sabouraud dextrose agar (SDA) with chloramphenicol and SDA with chloramphenicol and cycloheximide. The tube of SDA with chloramphenicol was incubated at 37°C and SDA with chloramphenicol and cycloheximide was incubated at 25°C, for a period of 4-6 wks. These were examined biweekly for growth characteristics like; color of the colony, its consistency and topography as well as any pigment production.

Any growth on the culture media was then subjected to morphological assessment using lacto-phenol cotton blue (LCB) mounting medium. Characteristics like presence or absence of microconidia, macroconidia their shape, arrangement and hyphal structures were looked for. Additional tests like urease and hair perforation tests were also performed for differentiation wherever needed. For inducing sporulation/conidiation, media like water agar was used for identification in some cases. All the media and reagents were procured from HiMedia Laboratories Pvt. Ltd Mumbai.

## 2.3 Statistical Analysis

Statistical tests like Chi-square and Fischer's exact test were used to determine the association between the different types of variables. SPSS 16 software was used for statistical analysis.

## 3. RESULTS

Out of a total of 300 patients recruited for the study, 190 (63%) belonged to urban areas whereas 110 (37%) were from rural areas. Age of cases ranged from 3 months to 85 years (mean SD 38.5±16.2 yrs), with female preponderance. Majority of the patients were in the age group of 21-40 yrs (48%) followed by age group of 41-50 years (18%). Most of the patients had involvement of the toenail (61%) whereas fingernail involvement was seen in 32% of them. Involvement of both sites was seen in 7% of the cases. Involvement of toenails was equal among both sexes; however, fingernail involvement was more in females as compared to males.

The most common clinical presentation was DLSO (79%), followed by PSO (11%), WSO (6%) and TDO (4%). Both DLSO (52%) and PSO (65%) were common in female than male patients. Maximum cases in our study were house wives (32%) followed by students (25%) and office workers (23%). Farmers represented only 2% of the cases. Miscellaneous group included teachers, businessmen, retirees etc. and constituted 18% of the cases.

### 3.1 Magnitude of Onychomycosis

Out of the 300 cases, 235 (78.3%) were found to have onychomycosis. A total of 321 samples were taken from 300 cases, of which 252 yielded

**Table 1. Comparison of microscopy and culture in the diagnosis of onychomycosis**

Results	Culture positive	Culture negative	Total	
KOH positive	77	159	236	Chi square = 5.786
KOH negative	16	69	85	Odds ratio = 2.088
Total	93	228	321	P-value = 0.018

a fungal isolate. Overall positivity on microscopy was 73.5% and on culture it was 29%. 159 (49.5%) samples were KOH positive and culture negative, 77 (24%) samples were KOH and culture positive and 16 (5%) were KOH negative and culture positive (Table 1 above). A significant association between KOH and culture was seen, the two variables being dependent on each other. Sensitivity and specificity of KOH as compared to culture was 83.9% and 30.3% respectively. Positive predictive value (PPV) and negative predictive value (NPV) was 32.9% and 82.1% respectively.

Most common systemic diseases associated with onychomycosis were hypertension, hypothyroidism and diabetes. Family and past history of fungal infections were the common risk factors associated with the disease. Fungal infections of other parts of the body were seen in 19 (8%) of patients with onychomycosis.

A total of 93 fungal isolates were recovered of which 91 (98%) were dermatophytes and 2 (2%) were non-dermatophytic fungi (Table 2). Overall, among the dermatophytes, *Trichophyton (T) rubrum* was the most common species isolated (81.3%). As far as site specific species is concerned, *T. rubrum* and *Epidermophyton floccosum* were most commonly isolated from toe nails whereas *T. violaceum* was commonly isolated from finger nails. In two patients, a non-dermatophytic fungus '*Fusarium solani*' was isolated. The isolation of different species from these sites was not statistically significant though. *T. rubrum* was isolated from all clinical types of nail involvement while *T. violaceum* was isolated from all except WSO. Other fungal species were isolated only from DLSO pattern of nail involvement.

**Table 2. Dermatophytes isolated in cases of onychomycosis**

Dermatophyte	Number	Percentage
<i>Trichophyton rubrum</i>	74	81.31
<i>Trichophyton violaceum</i>	7	7.69
<i>Epidermophyton floccosum</i>	5	5.49
<i>Trichophyton tonsurans</i>	4	4.39
<i>Trichophyton verrucosum</i>	1	1.09
Total	91	100.00

The commonest fungal infection seen to coexist with onychomycosis was tinea pedis (n=11; 4.68%) followed by tinea corporis (n=4; 1.7%) and tinea cruris and tinea unguium (n=2; 0.85%).

#### 4. DISCUSSION

The incidence of onychomycosis is increasing worldwide with social, cultural and economic factors contributing to it. Although the disease is asymptomatic yet it poses a serious concern to the patient as it becomes a chronic source of recurrent superficial skin infections. The peculiarities of the nail unit with respect to its structure and its immune mechanism make onychomycosis an adversity, which once established is difficult to eradicate.

This study was carried out with the intent to assess the magnitude of onychomycosis, identify the organisms responsible for it and ascertain various risk factors associated with the disease in Kashmir Valley. We found a preponderance of cases from urban as compared to rural areas which is in accordance with other studies where similar results were seen. [2,8] Urban people being more conscious about their health than rural ones and seek early medical advice which could be one of the reasons for the increased number of cases from these areas.

Onychomycosis was seen more in female (53.3%) than male patients (46.7%). Various other studies have reported a higher incidence of this disease in females [8-10]. The fact that females are more conscious about their appearance and disfigurement in nails can affect their self-esteem can be the reason behind this scenario. Some studies have however reported onychomycosis to be more common in males as compared to females where frequent trauma to nails and the use of occlusive footwear have been cited as reasons for such preponderance. [2,9,10].

Adults in the age group of 21 to 40 years were seen to be affected more than other age groups, a finding consistent with other studies carried out around the globe [2,6] Nonetheless, the actual incidence of onychomycosis may be higher in elderly in our state but the disease being

mostly asymptomatic and such patients being dependent upon others may not seek medical help for onychomycosis. Children on the other hand seem to be less susceptible to this infection due to less exposure to environments containing the causative pathogens, faster nail growth, smaller nail surface for invasion and lower prevalence of tinea pedis [6].

In our study toenail involvement was seen in the majority of patients. Similar pattern of nail involvement has been reported by many authors [9,11]. Slower growth of toenails as compared to fingernails is one of the contributing factors [3]. Also use of occlusive footwear during winter season could be one of the reasons as to why toenails are affected more than fingernails in our patients. This is in contrast to what has been reported from other parts of the country where authors have seen a predominant involvement of fingernails [2,5].

In our study the commonest clinical type of involvement was DLSO, seen in 78.8% of the patients most of them being females. This finding is consistent with the observation of many other workers who reported this type of nail involvement as the commonest [2,5,6,9]. However, in a study conducted in Nepal even though DLSO was the most common clinical type of presentation yet it was seen more commonly in male than female patients, a finding contrary to what we saw [5].

The predisposition of cutaneous fungal infections in patients having onychomycosis has been found in various studies [5,8]. The commonest cutaneous infection coexisting with onychomycosis in our study was found to be tinea pedis followed by tinea corporis. Most common systemic diseases associated with onychomycosis were hypertension and diabetes mellitus in our study. Of the 42 patients who were on anti-hypertensive treatment, 30 had onychomycosis. Predisposing factors for onychomycosis are multifactorial and hypertension being one of the contributing factors for its development has been reported by various authors [2,5,12,13]. Diabetes mellitus in our study was present in 19 patients, 15 of whom had onychomycosis. This is in concordance with other studies where patients with diabetes mellitus were found to have onychomycosis as well [3,6,7].

A higher percentage positivity of direct microscopy in our study (73.5%) was in contrast

to other studies where the positivity was 40% or even lower. Sampling error or low prevalence could be the reason behind this low positivity [8,9,13]. Overall positivity of culture was 29%. Combined positivity of culture and microscopy was 24% which is in accordance with studies conducted in India, Turkey and Indonesia [10,11,14] whereas in other studies the positivity of both culture and microscopy has been reported to be 40.2% [9,11].

A predominance of dermatophytes as a cause of onychomycosis was seen in this study, with *T. rubrum* being the most common dermatophyte isolated. Results similar to ours have been reported in India and elsewhere [2,6,9,10,13]. These fungi can invade keratinized tissue and thus are an important cause of onychomycosis [12]. Two patients in our study were infected with a non-dermatophyte fungus (*Fusarium solani*). Both patients were immunocompetent and both had a DLSO type of nail involvement.

## 5. CONCLUSION

Onychomycosis contributes to a substantial proportion of conditions affecting the nail. Though initially presenting as a cosmetic problem, it can eventually lead to permanent disfigurement of the nails and serve as a source of other fungal infections. Thus a proper microbiologic diagnosis is imperative for institution of early and appropriate treatment for best outcome.

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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