A Study on Clinico-Mycological Profile, Aetiological Agents and Diagnosis of Onychomycosis at a Government Medical College Hospital in Kashmir

ABSTRACT
Background: Onychomycosis is a major public health problem with a high incidence, associated morbidity and a long lasting treatment with anti-fungal agents. This study was carried out to know the clinico-mycological pattern of onychomycosis, which could help in the control of this infection.

Aims: The aim of this study was to determine the prevalence of various causative agents of onychomycosis and to study the clinical and mycological patterns of onychomycosis.

Material and Methods: This was a prospective study which was carried over a period of one year, from 1st February 2011 to 31st January 2012 on samples from 150 patients with clinically suspected nail infections, who attended the Dermatology Department of SKIMS Medical college, Kashmir, India. The nails were evaluated clinically and the nail samples were subjected to direct microscopy and culture.

Results: 60% samples were found to be positive by direct microscopy and culture. Males were infected more than females. The commonest age group which was infected was the 21-30 years age group. Finger nails were affected more frequently than toe nails and distolateral subungal onychomycosis was the most common clinical type of infection which was seen in 64.44% patients. The aetiological agents were dermatophytes (61.66%), Non–Dermatophyte Moulds (NDM) (31.66%) and yeasts (6.66%). Among dermatophytes, T. rubrum was the commonest aetiological agent.

Conclusion: Although dermatophytes were the main causative agents, NDM and yeasts were also not uncommon aetiological agents of onychomycosis. This study also emphasized the need of performing both a direct examination and culture to improve sensitivity. Since onychomycosis can cause physical, psychological and occupational problems, the clinico-epidemiological data can be helpful in development of preventive and diagnostic strategies.

Key Words: Onychomycosis, dermatophytes, non-dermatophyte moulds, yeasts

INTRODUCTION
Onychomycosis is a term which is used to describe a fungal infection of one or more of nail units and it can be caused by dermatophytes, yeasts or non-dermatophyte moulds. It represents upto 30% of mycotic cutaneous infections [1]. Clinically, onychomycosis is classified into various types; Disto–Lateral Subungal Onychomycosis (DLSO), Superficial White Onychomycosis (SWO), Proximal Subungal Onychomycosis (PSO), Endonyx Onychomycosis EO), Candidal Onychomycosis (CO), and Total Dystrophic Onychomycosis (TDO) [2-4]. The prevalence of onychomycosis is determined by age, predisposing factors, social class, occupation, climate, living environment and frequency of travel [5]. The worldwide incidence of onychomycosis is increasing and a number of factors contribute to its rise, like an immunocompromised status which is caused by HIV, immunosuppressive therapy and cancer chemotherapy or increased antibiotic usage [6]. Although onychomycosis is often regarded as merely a cosmetic problem which is rarely life threatening, its high prevalence and the associated morbidity makes it an important public health problem [1]. Onychomycosis resembles several diseases in the field of dermatology and medicine, so it is necessary to diagnose the infection with some laboratory evidence before treatment with anti-fungal agents, whose duration of treatment is long and may have some serious side effects [7]. The incidence of onychomycosis is high in Indian sub-continent because warm and humid climate, poverty, overcrowding and lack of medical facilities contribute to high prevalence of disease. Since the patients with dystrophic nails who seek medical advice is increasing, the present study was carried out to determine the prevalence of various causative agents of onychomycosis, to identify the clinical pattern of this disease in our part of world and to analyze the potential risk factors.

MATERIAL AND METHODS
Study Population and Period
This study was conducted over a period of one year, from 1st Feb 2011 to 31st Jan 2012, on samples from 150 patients with clinically suspected nail infections, who attended the Dermatology Out-Patients Department of SKIMS Medical College, Srinagar, India.

Inclusion Criteria
All patients with a clinical diagnosis of onychomycosis were included in the study.

Exclusion Criteria
The samples of patients who had taken anti fungal drugs were excluded from the study. Detailed history of patients was taken. The clinical pattern and location of disease was also documented.

Sample Collection and Processing
The specimens were collected for microbiological analysis, on the basis of the results of clinical evaluations. Samples from clinical abnormal nails were collected by vigorously scraping the distal portion of the nail, the nail undersurface as well as nail bed, after cleaning the area with 80 % alcohol, to remove contaminants with
a no. 15 scalpel blade. The specimens were analyzed by direct microscopy and culture.

**Direct Microscopy**
Specimens were placed on slides and one drop of 20% Potassium Hydroxide (KOH) was added to each slide. A microscopic examination was carried for the presence of fungal elements after incubating the slides for two hours or until digestion of specimens occurred [8].

**Culture**
**Culture was done by using:** 1. Sabouraud’s dextrose agar without antibiotics and 2. Sabouraud’s dextrose agar with 5% Chloramphenicol and cycloheximide. Both media were used in duplicate and they were kept at 25°C and 37°C. They were examined daily for six weeks before they were declared as negative. The growths were noted for colony characteristics in the form of rate of growth, texture of growth, surface colour, and colour on reverse and diffusible pigments. For microscopic morphology, tease mounts, cellophane tape mounts and slide cultures were done [9,10].

Yeasts were identified on the basis of germ tube tests, microscopic morphologies on corn meal agar and colour production on CHROMAGAR candida culture medium (Becton Dickinson).

The criteria for reporting NDMs as pathogens were positivity on direct microscopy and isolation of same fungi in second samples which were obtained some days later.

**RESULTS**
Out of 150 patients, 90 (60%) showed positive results on direct examination and/or culture. Direct examination was positive in 84 (56%) patients and fungal culture was positive in 60 (40%) patients [Table/Fig-1].

Among 90 patients with onychomycosis, 55 (61.11%) were males and 35 (38.88%) were females, with a male to female ratio of 1.57:1.

<table>
<thead>
<tr>
<th>Age Group</th>
<th>Male</th>
<th>Female</th>
<th>Total And % Age</th>
</tr>
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<tbody>
<tr>
<td>0 - 10</td>
<td>2</td>
<td>-</td>
<td>2 (2.22)</td>
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<tr>
<td>11 – 20</td>
<td>5</td>
<td>4</td>
<td>9 (10)</td>
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<tr>
<td>21 – 30</td>
<td>25</td>
<td>16</td>
<td>41 (45.55)</td>
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<td>31 – 40</td>
<td>14</td>
<td>9</td>
<td>23 (25.55)</td>
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<tr>
<td>41 – 50</td>
<td>6</td>
<td>4</td>
<td>10 (11.11)</td>
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<tr>
<td>51 – 60</td>
<td>2</td>
<td>2</td>
<td>4 (4.44)</td>
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<tr>
<td>&gt; 60</td>
<td>1</td>
<td>-</td>
<td>1 (1.11)</td>
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<tr>
<td>Total</td>
<td>55</td>
<td>35</td>
<td>90 (100)</td>
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**DISCUSSION**
In the present study, 60% samples were positive by direct examination and/or culture. In studies which were conducted by Kaur et al., Das et al., Jesudanam et al., and Aghamirian et al., 54.5%, 51.76%, 45.53% and 40.2% samples respectively were found to be positive by direct examination and/or culture [11-14]. In our study, direct microscopy with KOH mount and mycological culture showed positive results in 84 (56%) and 60 (40%) patients respectively. In our study, direct microscopy was positive in more cases than culture. The results were in accordance with the findings of study which was carried by Manjunath Shenoy et al., which showed positive results in 53% and 35% cases by direct microscopy and culture respectively. However, in the study which was conducted by Das et al, direct microscopy was positive in only 32.94% cases, while culture was positive in 49.4% cases [7,12].

In our study, 61.11% patients with onychomycosis were males and 38.88% were females, with a male to female ratio of 1.57:1. Although many reports had shown a greater susceptibility of females to this infection [15,16], in our study, males were dominant. In the study which was conducted by Garg et al., and Veer et al., males were infected more than females [17,18]. The increased prevalence of onychomycosis in men could be due to nail trauma and more common use of occlusive footwear. In the present study, highest numbers of patients (46%) were in the age group 21-30 years, followed by those in age group of 25-45 years (25.55%). Adhikari et
al., also found a higher prevalence of onychomycosis in similar age groups [19]. In contrast, Velez et al., and Mercantini et al., reported higher prevalences among adults who were over 50 years of age [16,20]. The increased prevalence of onychomycosis at young ages could be because of occupation related trauma, cosmetic awareness and shoe wearing habits. In elderly, prevalence may be higher than what was observed, but as the disease is asymptomatic they are not mostly bothered about it.

In our study, distolateral subungal onychomycosis was the commonest clinical pattern in 64.44% cases, followed by proximal subungal onychomycosis (15.55%), as was found in many other studies [14,21]. The present study showed that finger nails were involved more often than toe nails. Although toe nails have been reported to be more commonly involved [1], our finding was in accordance with those of many other studies in which finger nails were found to be more frequently affected [14,21]. The lower incidence of toe nail onychomycosis could be because of lesser cosmetic awareness on their disfigurement.

In our study, the most common organisms which were isolated in culture were dermatophytes (61.66%), NDM (31.66%) and yeasts (6.66%). This finding was in accordance with those of many studies, which had demonstrated a greater prevalence of dermatophytes as the aetiologic agents of onychomycosis [11, 14, 22] and it was in contrast to those of other studies which had found yeasts as the most common agents [23,24]. Among the dermatophytes, T. rubrum was the most common aetiologic agent which was found in our study, followed by T. mentagrophytes. Although some studies had reported T. mentagrophytes as the most common dermatophyte [11], our finding was in concordance with those of many other studies which had found T. rubrum as the most common dermatophyte which was responsible for onychomycosis [15,18]. The increased prevalence of T. rubrum could have been due to increased virulence and better adaptation to hard keratin of nails. Among the NDM, A. niger was the commonest isolate which was obtained. Kaur et al., and Grover et al., also found A. niger to be the most common NDM which was responsible for onychomycosis [11,21]. Candida albicans accounts for a majority of cases of onychomycosis which are caused by yeast. Candida parapsilosis, Candida tropicalis and Candida krusei account for the remainder of the cases [25]. In our study, Candida albicans was isolated from 3 (5%) cases and Candida parapsilosis was isolated from 1(1.66%) samples.

CONCLUSION
In our study, dermatophytes were the most common aetiologic agents of onychomycosis; the roles of NDM and yeasts in causing infections were also demonstrated. In our study, the combined sensitivity of direct microscopy and culture was greater than those of direct microscopy and culture alone. This emphasizes the need of performing both tests. In the present study, men were more commonly infected and the age group of 20-40 years was more commonly involved. Distolateral subungal onychomycosis was the common clinical pattern. The clinico-epidemiological data can be helpful for creating public awareness and for the development of diagnostic, preventive and treatment strategies.

REFERENCES