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## RESEARCH ARTICLE

### MYCOLOGICAL STUDY OF ONYCHOMYCOSIS IN DIABETIC PATIENTS

Dr. Prathap, R., Dr. Jyoti S. Kabbin, \*Dr. Bhavana C. and Dr. Ambica R.

Bangalore Medical College and Research Institute, Bangalore, India

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#### ABSTRACT

**Background:** Onychomycosis refers to fungal infection of nails with various etiological agents, involving dermatophytes, yeasts and moulds. Onychomycosis continues to be worldwide problem constituting a large bulk of cases attending the dermatology outpatient department. Diabetics are more prone to onychomycosis, most of the time the infection is subclinical and may lead to secondary infections and bad prognosis. Hence this study was undertaken to diagnose onychomycosis earlier in diabetics which will help to start suitable antifungal therapy and prevent further complications.

##### Objectives:

1. To identify the most predominant fungus isolated from diabetics suffering from onychomycosis.
2. To know the percentage of diabetics suffering from onychomycosis.
3. To identify the age and sex group of people at a higher risk of acquiring onychomycosis in diabetes mellitus patients.

**Methods:** The study was conducted from July 2015 to August 2015, in the department of Microbiology of a tertiary care hospital in Bangalore, including 50 patients clinically suspected of onychomycosis. Nail scrapings and clippings were collected depending upon site, type of nail involvement and subjected for microscopic examination using 40% KOH, followed by fungal culture. Species identification was done based on colony characteristics, pigment production, LPCB mount and slide culture.

**Results:** Out of 50 patients, 22 (44%) were diagnosed having onychomycosis. The common isolates were non-*albicans Candida* (n=8/22, 36.36%) followed by *Aspergillus niger* (n= 5/22, 22.72%), *Penicillium* (4/22, 18.18%) and others being *Paecilomyces*, *Gliocladium* and *Rhizopus* species each being 4.54%. Mixed infection with *Aspergillus niger* and non-*albicans Candida* species was seen in 2/22 (9.09%) of cases.

**Conclusion:** The present study concludes that screening test for fungal infections on regular basis is important for long term diabetic patients and they should be started on the suitable antifungal therapy as soon as possible. Early diagnosis of onychomycosis in diabetics helps in the prevention of further complications.

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## INTRODUCTION

Onychomycosis or Tinea unguium is a fungal infection of the nail plate or nail bed. It accounts for up to 50% of all nail disorders and 30% of all superficial fungal infections (Kaur *et al.*, 2008). Dermatophytes, non-dermatophytemolds and yeasts are its causative agents. At first it presents with thickening of nail with discoloration of nail to white, yellow patches on the nail bed, black, and foul smell (James *et al.*, 2006). Generally it is not associated with pain, but may be associated with psychological symptoms in upper socioeconomic classes (Szepietowski and Reich, 2008). Toe nails are 25 times more susceptible to infection than finger nails as causative molds are ubiquitous fungi seen in soil, water and decaying vegetation.

Ageing, immunodeficiency, trauma, hyperhidrosis, socioeconomic condition, frequency of travel, climatic conditions predispose to onychomycosis. Diabetics are more prone to get onychomycosis. The disease incidence increases in reduced peripheral circulation, diabetes, nail trauma, poor nail hygiene. If left untreated it may lead to significant clinical consequences such as secondary bacterial infections, cellulitis, and therapeutic difficulties (Ahuja *et al.*, 2011). Diabetes mellitus affects all socioeconomic and age groups, its incidence has increased to 381 million in 2013 according to international diabetic federation. Diabetic foot complication is one of the severe complications in diabetes. A large prospective study showed that onychomycosis is among most significant predictors of foot ulcers (Mayser *et al.*, 2009). Distal lateral subungual and dystrophic onychomycosis are the most frequent clinical types. Onychomycosis in patients with diabetes increases the risk of other foot disorders and limb

\*Corresponding author: Dr. Bhavana C. ,  
Bangalore Medical College and Research Institute, Bangalore, India.

amputation. Thickened nail can cause erosion of nail bed because of pressure, just as tight shoes can cause friction blisters in these patients. When combined with peripheral neuropathy blisters and erosion may progress to cellulitis or osteomyelitis of the underlying bone. Extension of the fungal infection to surrounding skin may lead to fissures in the plantar and interdigital skin, providing a route for the entry of the bacteria leading to secondary complications (Jason *et al.*, 2006). Certain skin conditions such as psoriasis, lichen planus, onychogryphosis and nail trauma can mimic onychomycosis. Hence laboratory investigations are needed to differentiate accurately between fungal infections and the above mentioned skin diseases (Golia *et al.*, 2012). Hence this study was undertaken to diagnose onychomycosis earlier in diabetics which will help to start suitable antifungal therapy and prevent further complications.

## MATERIALS AND METHODS

This prospective study was carried out in the Department of Microbiology, Bangalore Medical College and Research Institute for a period of 2 months from July 2015 to August 2015. Diabetics of all age groups with slightest change in their nail appearance were included in the study. Patients on oral antifungal medications during the previous three weeks or who used the topical antifungal nail lacquer were excluded. Following the approval of the ethical committee of the institution, the samples were collected from 50 diabetic patients of all age groups with slightest change in their nail appearance attending the surgical minor OT, diabetic patients from diabetic wards at Victoria and Bowring and lady Curzon hospitals, and also clinically diagnosed onychomycosis patients with diabetic history attending dermatology department at Victoria hospital, attached to BMCRI, Bangalore. The procedure and importance of collecting the samples was explained to every subject in their own language.

**Collection and processing of sample:** Samples were taken from the toe nail and the affected nails. Nails were cleaned with 70% alcohol prior to sample collection. The nail scrapings and clippings were collected from a clean sterile scalpel blade over a clean white paper and were sent to microbiology lab within two hours of collection.

The collected samples were examined directly under 40% potassium hydroxide solution after 24 hours incubation. The samples were inoculated on Sabouraud's dextrose agar (SDA) with and without antibiotics. The tubes were incubated at room temperature and at 37°C for 4 weeks before they are reported as no growth. The tubes were examined for growth everyday for the first week and thrice weekly thereafter for 3 weeks. The findings were recorded as no growth/growth/yeast/mould (dermatophyte or non dermatophyte). Isolates were further identified by standard mycological methods<sup>8</sup>. The growth was confirmed by repeated isolation of the fungus. The data obtained was calculated in the form of percentages and projected in form of tables, pie charts and bar graphs.

## RESULTS

Out of 50 patients studied, 37 (74%) were male and 13 (26%) were female. Male to female ratio was 2.8:1. The most common age group studied were 41-45 years, 51-55 years and 61-65 years. Out of 50 samples, 22 samples were positive for growth of various mycological agents with the isolation rate being 44%. The common isolates were Non-*albicans Candida* (n=8/22, 36.36%) followed by *Aspergillus niger* (n= 5/22, 22.72%), *Penicillium* (4/22, 18.18%), *Paecilomyces* species (n=1/22, 4.54%), *Gliocladium* species (n=1/22, 4.54%), *Rhizopus* species (n=1/22, 4.54%). Mixed infection with *Aspergillus niger* and non-*albicans Candida* species was seen in 2/22 (9.09%) of cases (Figure-1). Among the positive cases, there was male preponderance. 35.13% of males (13/37) and 69.23% of females (9/13) were affected. Majority of patients belonged to the age group of 51-65 years. The occurrence of onychomycosis was 1.45 times higher in males than in females. Male: Female distribution among different age groups is shown in Figure 2. Out of the total population, 32 individuals had diabetic foot ulcer, 6 of them had limb amputations. 12 patients with foot ulcer were found to have onychomycosis and in those 12 patients, 4 patients had leg amputations. Out of the 22 affected individuals 7 individuals had a diabetic history of 7 to 9 years and the remaining 15 individuals had a history of 3 to 5 years. All the affected individuals were from low socio economic class, with an occupation of coolies, construction workers, farmers, some of them were retired teachers and retired policemen.

Table 1. Comparison of organisms isolated with other studies

|                   | Present study | Dogra <i>et al.</i> , (2002) | Saunte <i>et al.</i> , (2006) | Gulcan <i>et al.</i> , (2011) | Leelavathi <i>et al.</i> , (2013) |
|-------------------|---------------|------------------------------|-------------------------------|-------------------------------|-----------------------------------|
| Yeasts            | 36.36%        | 48.2%                        | 7%                            | 56.1%                         | 25.2%                             |
| Dermatophytes     | -             | 37%                          | 93%                           | 43.9%                         | 0.81%                             |
| Non-Dermatophytes | 54.54%        | 14.8%                        | -                             | -                             | 48.79%                            |
| Mixture           | 9.1%          | -                            | -                             | -                             | 25.2%                             |

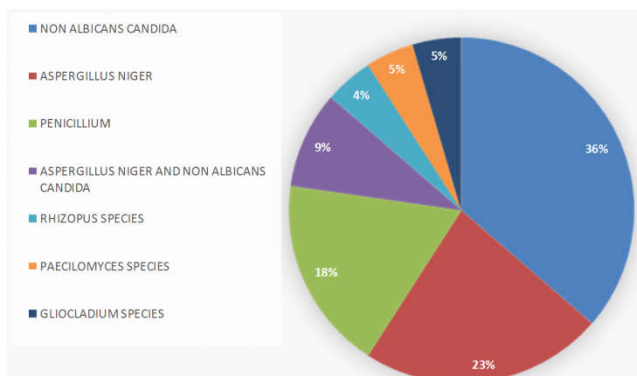


Figure 1. Pie chart showing species causing onychomycosis in diabetics

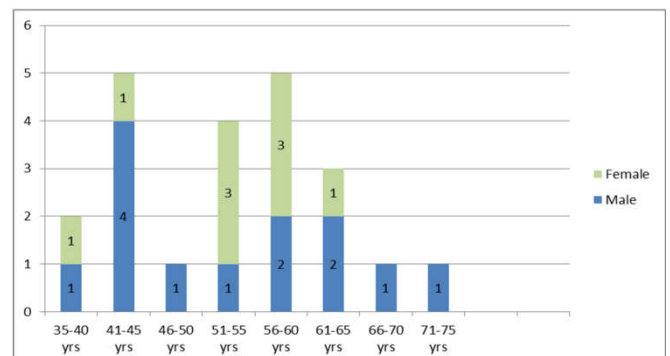


Figure 2. Relative distribution of positive cultures between different age groups

## DISCUSSION

Onychomycosis among diabetics is being increased every day. The organisms causing onychomycosis are dermatophytes, yeasts and non-dermatophytes where the proportion of these causing the infection of nails varies in each studies. Comparison of organisms isolated with other studies is depicted in Table 1. The percentage of yeasts (*Candida* and non *albicans Candida*) causing the onychomycosis in our study is 36.36, as compared to 25.2% in Leelavathi *et al.*, Malaysia, 2013; 56.1% in Gulcan *et al.* 2011 and 48.2% in Dogra *et al.* India, 2002. Non dermatophytic molds like *Aspergillus niger*, *Penicillium*, *Paecilomyces species*, *Gliocladium species*, *Rhizopus species* also causes the infection in low proportions but in the recent studies the proportion has been increased to 48.79%. (Leelavathi *et al.* Malaysia, 2013) and to 54.54% in our study. The onychomycosis is also caused by the mixed infections, 25.2% (Leelavathi *et al.* Malaysia, 2013). In our study it was 9.1%. 2 cases out of 22, the causative agents being *Aspergillus niger* with *non albicans Candida* together. Dermatophytes are the main cause of onychomycosis in many studies (Dogra *et al.*, 2002; Saunte *et al.*, 2006; Gulcan *et al.*, 2011; Gupta *et al.*, 1998) however dermatophytes were not found to cause the onychomycosis in our study may be due to the climatic variations and other environmental and occupational variations. Our study shows significant correlation between the occurrence of onychomycosis and the increasing age as the mean age for the culture positive patients was 53.31% and the age class 51 to 65 showed 46.15% (n=12/26) of the patients with the infection, which is in accordance with the studies (Gupta *et al.*, 1998). Some studies did not find any correlation between onychomycosis and age (Dogra *et al.*, 2002; Leelavathi *et al.*, 2013; Gulcan *et al.*, 2011). We also found that onychomycosis cases were more observed in the patients with increased duration of diabetics which is similar to the study done in India (Dogra *et al.* India, 2002). Our study also showed that the occurrence of onychomycosis is 1.45 times higher in males than in females, similar results were found by (Al-Mutairi *et al.*, 2010; Gupta *et al.* London 1998), and some studies did not found any significant correlation (Gulcan *et al.*, 2011; Leelavathi *et al.*, 2013). This study also found that 54.54 % (n=22) of the infected patients had diabetic foot ulcers with 18.18 % (n=22) of them had leg amputations. Many studies state that onychomycosis in diabetics may lead to secondary complications like diabetic foot ulcers, cellulitis and gangrene (Jason *et al.*, 2006; Leelavathi *et al.*, 2013). Diabetic patients with onychomycosis were more frequently diagnosed with plantar 62.2% and interdigital infections 46.7%, these abrasions combined with impaired circulation can lead to secondary complications. Onychomycosis has high prevalence in subjects suffering with diabetic foot complications. Since the patient's occupations are like house building, coolies, vegetable vendors, they are exposed to humidity most of the times which can give favorable condition for the fungus to colonize. This combined with peripheral neuropathy may make the individual to not to notice any abrasion leading to secondary complications.

## Conclusion

Diabetics are more prone to onychomycosis, most of the time the infection is subclinical. Since pain is which brings the patient to the hospital, patient with onychomycosis may not visit the hospital until the infection is severe. So the infection

may lead to bad prognosis. Yeasts and non-dermatophyte molds are equally responsible for causing the onychomycosis in diabetic patients in our area. Onychomycosis has increased incidence in diabetic patients with increasing age, male gender and the duration of diabetes. Onychomycosis in diabetics can be a risk factor for diabetic foot ulcers. Good hygiene and good care of legs can prevent onychomycosis and secondary complications. Early diagnosis of onychomycosis in diabetics helps in the prevention of further complications.

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