Etiological agents of onychomycosis diagnosed in the medical mycology laboratory of the University of Costa Rica.

Ingrid Salas-Campos, Norma T. Gross-Martínez

Abstract

Background and aim: Among the onychodistrophies, onychomycosis are the most frequently encountered. This infection not only constitutes an esthetic problem for the patients, but can also affect their daily activities. For dermatologists, it is crucial to make a differential diagnosis; thus, the medical mycology laboratory plays an important role to achieve this purpose. The fungal agents most frequently encountered are the dermatophytes, however, other filamentous non dermatophyte fungi have been isolated and are known to be less susceptible to antifungals. In the present work, 115.

The frequency of onychomycosis among patients attending the medical mycology laboratory, UCR, was studied during four years, according to the age and sex of the patients, as well as the isolated etiological agents identified.

Methods: The study included all patients that requested the community service provided by the Department of Medical Mycology, School of Microbiology, UCR, between January 2007 and December 2010 and that showed nail alterations suspicious for onychomycosis. The age and sex of each patient were registered and samples were processed for direct microscopy and culture.

Results: A total of 431 nail samples were collected, of which 85.4% were toenails and 14.6% fingernails. The mean age of the patients was 49 years, of which 64% were females and 36% males. Onychomycosis was diagnosed, either by direct microscopy and culture, or only with positive direct microscopy, in 73.4% of the sample population, of which 89.4% were toenails and 10.6% fingernails. Trichophyton rubrum was the etiological agent most frequently isolated from toenails, followed by Fusarium spp. C albicans was the most frequent fungal agent observed in fingernails.

Conclusion: The diagnosis of onychomycosis relies upon both the clinical and laboratory diagnosis. Dermatophytes, yeasts and non-dermatophyte filamentous fungi were identified in the population studied. These findings should be considered due to their implications to the choice of the most appropriate treatment.

Key words: onychodystrophies, onychomycosis, dermatophytes, filamentous non dermatophyte fungi.

Date received: June 20, 2011 Date accepted: February 17, 2012

Among onychodystrophies, onychomycoses account for 36-50% of nail-related pathologies and have a prevalence of 2.5-16% in the general population. These figures depend on geographic and population-specific factors. However, the real dimension of this pathology among the population is unknown because of reasons such as a lack of consultation by affected patients.
onchomycoses, but also to be able to identify the etiologic agent involved because for non-dermatophyte filamentous fungi recently described in Costa Rica, a priori prescription is not possible since some of these fungi, such as *Fusarium*, are resistant to imidazoles, while for others, such as *Scytalidium dimidiatum*, there is no efficient treatment. This situation makes necessary to look for other therapeutic options.

Onychomycoses are not only an aesthetic problem for the patient, but they can also affect daily activities such as walking, standing, exercising, recreation, nail trimming, and even shoe preference because of the appearance of their nails if wearing sandals or the microenvironment and the thickening of the nails when wearing closed shoes. This infection’s effect on the appearance of nails can also alter the patients’ psychological status by inducing shame, low self-esteem, anxiety and social effects among others. It can also cause complications in elderly diabetic or vascular disease-affected patients, such as cellulitis, and even systemic dissemination of the fungi from nails in immunocompromised patients.

In this study, we analyzed the frequency of onychomycoses that were diagnosed at the Medical Mycology laboratory of the University of Costa Rica (UCR) during 4 years and classified by patient age and gender, as well as the etiologic agent that was identified.

**Methods**

This study included all the samples from patients with ungual lesions that were received for examination on the suspicion of onychomycoses between January 2007 and December 2010 as part of the service provided by the Medical Mycology Section of the Faculty of Microbiology, project ED-539 of the Social Action Vice rectory, UCR. For each patient, name, age and gender were recorded, and then a sample of subungual detritus or, if there was periungual inflammation, a sample of the affected tissue was obtained. All samples were examined by direct observation in 40% KOH and fungal elements were searched for under the microscope. Part of the material was cultured on Saburaud’s agar and media with actidione and chloramphenicol. Cultures were incubated at room temperature for at least 15 days. The identification of filamentous fungi was based on macroscopic and microscopic characteristics of the colony while the identification of the yeasts was carried out with metabolic tests including the semi-automated API system or an automated Vitek®.

**Results**

During the study’s period a total 431 nail samples were processed. Of these, 85.4% were toenails, and the remaining 14.6% were fingernails. Patient’s mean age was 49 years (range 5-87 years; Figure 1) and 64% of them were female and 36% male.

Among the samples, 119 (27.6%) had a negative result for fungi in both the direct examination and the cultures. In the remaining 312 samples (73.4%) a diagnosis of onychomycosis was established by either direct examination, culture or both, of which 279 (89.4%) were toenails and 33 (10.6%) were fingernails.

Among the 279 toe onychomycoses, 122 (43.7%) were from males and 157 (56.3%) from females. The age distribution is shown in Figure 1. For 273 samples (97.8%) the direct examination was positive, 135 (48.4%) of which had negative culture while from 142 (51.6%) samples 147 isolations were obtained. Of these, 103 (70.1%) were dermatophytes, 9 (6.1%) were yeasts, and 35 (23.8%) were non-dermatophyte filamentous fungi (Table 1).

Among the 33 finger onychomycoses, 7 (21.2%) were from males and 26 (78.8%) from females. The age distribution is shown in Figure 1. Thirty samples (90.9%) had a positive direct examination. In 12 samples (36.4%) it was impossible to determine an etiologic agent and in 21 (63.6%) there was a positive isolation. Of these, 1 (4.7%) was a dermatophyte, 10 (47.6%) were yeasts, and 10 (47.6%) were non-dermatophyte filamentous fungi (Table 1).

**Discussion**

The majority of the cases that are received by our laboratory are due to onychomycoses as has been reported by other studies. Pathologies similar to this infection include pachyonychia, ungual dyschromia, acquired dystrophies, or changes due to diseases such as pityriasis rubra and psoriasis, which could explain the onychodystrophies that some patients present.

Nowadays, onychomycoses represent a public health problem because of their interpersonal transmission, their high prevalence among the population, the difficulty of their treatment and because they are favored by modern activities such as exercising in gymnasiuks, the use of public pools and...
baths, the easiness of travel and the use of occlusive shoes.\textsuperscript{9,16,18} Moreover, it has been reported that the global incidence of this condition is rising as well as the factors that facilitate its development, such as diabetes, vascular problems and nail trauma.\textsuperscript{16}

As it has been reported in the literature,\textsuperscript{14,16,19} in our laboratory some samples for which a positive direct examination was obtained, it was impossible to isolate an etiologic agent. This could be due to the fact that the sample was not representative because there is a higher possibility that the fungus is alive in the proximal area of the nail where access for sample collection is more difficult. Moreover, some patients apply to themselves topical treatments or take antimycotics that impair the isolation of the fungal agent. For this, it is indispensable to recommend the patient not to apply or take antimycotic medication before the sample collection.

It is important to know the epidemiology of onychomycoses in a country because it can vary between different geographic areas in terms of their frequency by gender, age and etiologic agent.\textsuperscript{15} In this study, a higher amount of cases were seen among women as has been described in other countries.\textsuperscript{1,15,16,20} This could be due to the fact that in many countries women are more frequently employed in domestic labor,\textsuperscript{16} which favors the maceration of the skin, a predisposing factor for finger onychomycoses. In turn, it could also be due to the fact that women are in general more aware of their health, their physical appearance and have easier access to medical consultation, factors that could make the statistics not reflect the real situation regarding onychomycoses.\textsuperscript{20,21}

With regards to the age of the patients, a higher number of consultations and diagnoses of onychomycosis was registered for patients between 40-60 years of age, which agrees with what has been reported in other latitudes.\textsuperscript{1,15,16,20} Even though children and adolescents suffer onychomycoses, it is expected that most cases will be observed among adults because of factors such as a slower growth of the nail, the presence of microtraumata due to occlusive shoes or sport (onychomycoses among barefooted people are rare),\textsuperscript{15} as well as higher work activity, venous insufficiency and even a higher exposure to the fungus.\textsuperscript{16} Also, the low frequency among children could be due to the structure of the ungual plaque, a lack of accumulated traumata, fast ungual growth and the subsequent elimination of the fungus.\textsuperscript{16,20} The reduction in cases after 60 years of age could be due to low motivation to consult for a problem that many consider to be aesthetic or even to follow a treatment regime.\textsuperscript{9} However, if we consider the possible complications that could arise in elderly patients, these conditions should not be left without a proper diagnosis and treatment.\textsuperscript{10,12}

\begin{table}
\centering
\caption{Onychomycosis etiologic agents and number of isolations from toe and finger nails in the samples processed at the Medical Mycology section, Faculty of Microbiology, UCR, 2007-2010.}
\begin{tabular}{|l|l|l|l|l|}
\hline
\textbf{Etiologic agent} & \textbf{Toenail isolations} & \textbf{Fingernail isolations} \\
 & \textbf{Number} & \textbf{\%} & \textbf{Number} & \textbf{\%} \\
\hline
\textit{T. rubrum} & 95 & 66,9 & 1 & 4,8 \\
\textit{T. tonsurans} & 1 & 0,7 & 0 & \\
\textit{T. mentagrophytes} & 2 & 1,4 & 0 & \\
\textit{M. canis} & 1 & 0,7 & 0 & \\
\textit{Fusarium} & 25 & 17,6 & 4 & 19,0 \\
\textit{Scopulariopsis spp.} & 2 & 1,4 & 0 & \\
\textit{S. dimidiatum} & 4 & 2,8 & 1 & 4,8 \\
\textit{A. versicolor} & 1 & 0,7 & 0 & \\
\textit{C. albicans} & 2 & 1,4 & 7 & 33,3 \\
\textit{Candida spp.} & 6 & 4,3 & 3 & 14,3 \\
\textit{Trichosporon spp.} & 0 & & 5 & 23,8 \\
\textit{Mixed}\textsuperscript{1} & 3 & 2,1 & 0 & \\
\textit{Total} & 142 & 100 & 21 & 100 \\
\hline
\end{tabular}
\footnotesize{1 two of the mixed isolations were of \textit{+ T. rubrum} and one of \textit{Fusarium + C. haemulonii}.}
\end{table}
**Ethiology of onicomycosis**

*T. rubrum* was the most common etiologic agent isolated from the toenails. This fungus is the most commonly reported in many countries, possibly due to its anthropophilic character, which is favored by modern activities such as those described previously.

Among the non-dermatophyte filamentous fungi, *Fusarium* spp. was the most common agent in toenails, while *Trichosporon* spp. was the most common in fingernails. In European countries, non-dermatophyte filamentous fungi represent 1.5-6% of all onychomycosis cases, while in countries such as India they are very important, accounting for 39% of the isolations.

Onychomycoses caused by *Fusarium* spp. do not respond to treatment with fluconazole, and because of this these infections must be treated with terbinafine and itraconazole with satisfying results, as well as with ciclopirox in spray after a total or partial elimination of the nail with 40% urea.

In this study, we were able to isolate *Scopulariopsis brevicaulis* and *Aspergillus versicolor* from toenails. For the treatment of these cases oral terbinafine or itraconazole, as well as the partial or total extraction of the nail with ciclopirox as an ointment or a spray, bifonazole or terbinafine as nail cream.

Regarding *Scytalidium dimidiatum*, even though it was isolated only in a few cases, its identification is relevant because it is generally considered incurable, requiring chemical ablation of the nail along with ciclopirox or 5% amorolfine. For this fungus, voriconazole has been tested in vitro, showing a low minimal inhibitory concentration; hence its use in these infections should be considered.

In fingernails, *C. albicans* was the most commonly isolated species. *Trichosporon* spp. was only isolated from fingernails. This fungus has been reported in other countries, although its role as a causative agent is disputable and it could even be considered as an invader secondary to damage to the nail.

In conclusion, because not all onychodystrophies are of fungal origin, a precise laboratory diagnosis of onychomycosis is important, as has been shown in the present work. Also, the identification of the etiologic agent is indispensable considering that, even though most isolations in this population involved *T. rubrum*, a fungus that is sensitive to various antifungotics, other fungi more resistant to conventional treatments that are commonly used in our country (e.g. fluconazole). Furthermore, it is necessary to stress that, since onychomycoses are infectious, transmittable diseases, they represent a public health problem, not only because of their difficult treatment, but also because their frequency among the population.

**References**


