Saudi Journal of Medical and Pharmaceutical Sciences **ISSN 2413-4929 (Print)**
Scholars Middle East Publishers **ISSN 2413-4910 (Online)**
Dubai, United Arab Emirates
Website: https://saudijournals.com/

Onychomycosis in Patients Attending in Dermatology Clinics of the City of Rio de Janeiro, Brazil

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Original Research Article

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Article History
Received: 11.05.2018
Accepted: 24.05.2018
Published: 30.05.2018

DOI:
10.36348/sjmps.2018.v04i05.019

Abstract: Onychomycoses are fungal infections that affect hands and feet nails and are caused by several genera of filamentous and yeast fungi. The knowledge of the etiology is fundamental to establish the appropriate therapeutic conduct. Most fungi genera are free living, but some have adapted to parasitism and others depend on predisposing factors for implantation in the host. The objective of this research was to perform an epidemiological survey to identify the genera of fungi etiological agents of onychomycosis in patients attending in Rio de Janeiro dermatology clinics. After cleaning the nails with 70% alcohol, scraped scales with a sterilized scalpel were collected and seeded in Sabouraud-dextrose-agar and Mycosel culture media at room temperature. The colonies of filamentous fungi were identified by the cultural characteristics and the yeasturiformes by the staining of Gram method and biochemical tests. Among the 1290 cultures examined, 362 (28.06%) were positive for the following fungi: *Candida spp*. 335 (92.54%), *Trichophyton rubrum* 12 (3.31%), *Epidermophyton floccosum* 8 (2.21%), *Microsporum canis* 6 (1.66%) and *Aspergillus niger* 1 (0.28%). The fungal etiology of the nail lesions had an incidence of 28.06% of the samples from 1290 patients, being the genus *Candida* the most prevalent cause of onychomycosis

Keywords: Onycomycosis, *Candida*, Dermatophites, Fungi.

INTRODUCTION

Onychomycosis is an infection of the nails caused by fungal elements which infects and feed on keratin and other substances that compounds the nails.

The infections by dermatophyte fungi affects approximately 40% of the world population and accounts for 30% of all cutaneous mycotic infections, being the most common those affecting the skin and mucous membranes [1, 2]. Onychomycoses are the most frequent nail diseases, accounting for 18% to 40% of all onycopathies. The onychomycosis of the toenail is often associated with tinea interdigitalis or plantar tinea pedis [3, 4].

According to the clinical form, onychomycoses are distributed into the following classifications: 1 - subungual, which may be distal, lateral or proximal; 2- white superficial; 3 total dystrophic; 4-endonyx, whose clinical variety has been described in recent years [5, 6].

The prevalence of onychomycosis has increased in recent decades. Its incidence rate changes according to the studied population, but usually affects between 2% and 18% worldwide, and can reach up to 48% in adults over 70 years of age. This fungal infection may be related to certain professions as volleyball players, in which the incidence rates reach up to 89%. Onychomycoses are also associated with people with metabolic risk factors such as diabetes, autoimmune diseases, and some habits as the use of collective showers and swimming pools, use of tight shoes, among other conditions [7].

Onychomycoses represent infections that must be considered relevant as they damage population's health, since the patients' quality of life is impaired. Self-esteem can be reduced and functional capacity can be affected by interfering with daily activities. Onychomycoses may intensify other clinical infections, especially in elderly people, and may lead to lower limb amputations in patients with diabetes mellitus associated with onychomycosis. Diabetic patients have a higher prevalence of onychomycosis than non-

diabetics. The incidence of secondary infections by other microorganisms is higher in diabetic patients with onychomycosis than among diabetics who do not have this kind of infection [2, 8].

Interdigital fungal infections present a strong association with erysipelas or leg cellulitis [9]. Other causes which contribute to increase the number of people infected with dermatophytes are epidemic AIDS, the long-time use of corticosteroids and antibiotics, and the use of immunosuppressive drugs in transplant patients [10, 11]. A common mechanism for the transmission of fungal agents usually occurs among people frequenting gymnasium, as well as among athletes of collective sports activities. There are geographic differences in the epidemiology and etiology of onychomycosis, especially regarding the frequency of each fungal profile responsible for the infection. *Trichophytum rubrum* is the most frequent in England, Germany, Canada, the United States of America and India, while in Belgium, Saudi Arabia and Spain there is a high prevalence of *Candida* species infecting the nails [12].

One of the predisposing factors in the pathology of mycoses is related to the ambient. The population of the rural zone is often in contact with pathogens present in the environment or through a possible contact with animals which are hosts of superficial mycoses. Other aspects that should be considered for the predisposition to fungal agents are related to the use of public swimming pools, clothing without the correct hygiene measures, the use of inadequate footwear in warm climate places, which produce sweating of the feet and enables the implantation of onychomycosis fungi [13-15].

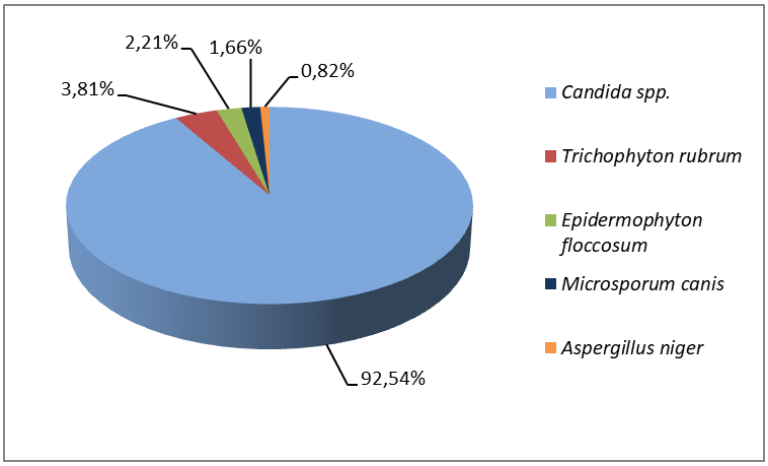
This research has the objective to survey the incidence and identify the etiology of the fungi which causes onychomycosis in the population of the city of Rio de Janeiro, Brazil.

MATERIALS AND METHODS

A total of 1290 nail samples from patients with suggestive lesions of onychomycosis were examined. After cleaning the nails with alcohol at 70%, scales scraped with a sterilized scalpel were collected. The material was seeded in Petri dishes containing Sabouraud-dextrose-agar and Mycosel culture media. The dishes were sealed with adhesive tape and kept at room temperature. The colonies of filamentous fungi were identified by the cultural and morphological characters and the yeasts by Gram staining method and biochemical tests.

RESULTS

Among the 1290 examined samples, 362 cultures (28.06%) were positive for the following fungi: *Candida* spp. 335 (92.54%), *Trichophyton rubrum* 12 (3.31%), *Epidermophyton floccosum* 8 (2.21%), *Microsporum canis* 6 (1.66%) and *Aspergillus niger* 1 (0.28%).



Graph-1: Distribution of the onychomycosis etiologic agents in patients attending in dermatologic clinics in the city of Rio de Janeiro by species:

DISCUSSION

Onychomycoses are the most common nail disease. These mycoses affect most frequently individuals with vascular diseases, with debilitating diseases such as carcimomatoses, smokers, immunosuppressed, immunodeficient and people with autoimmune diseases [16]. We also observed these characteristics in our research.

Studies on etiological agents of onychomycosis have been performed by researchers in several countries. These investigations use the direct research by the technique of whitening with potassium hydroxide for the observation of filamentous and yeast structures. However, for the final identification of the species it is mandatory to sow the nail scales in culture media suitable for the development of fungal species. Some authors claim that in their studies fungal infections that compromise nails are produced by

dermatophytes in 90% of cases [17, 18]. Our research involved the culture of the scales with the objective of not only detecting the incidence, but determining the prevalence through the identification of the fungal agents in the studied population.

During the years of 2013 and 2014, the researchers Mezzari *et al.*, attended 179 patients and collected 199 clinical samples from several anatomical sites. Mycological cultures showed that the most frequently isolated fungi were *Candida* species (24.9%), with the same incidence of *Trichophyton rubrum* (24.9%), followed by *Trichophyton mentagrophytes* (22.4%). After the mycological diagnosis, the patients were referred to the basic health unit in order to receive the appropriate treatment. Our results obtained with the mycological exams of people with onychomycosis in the city of Rio de Janeiro found that the fungal etiology of nail lesions by the genus *Candida* corresponded to 92.54% of the causes of onychomycosis [19].

Sav *et al.*, studied the sensitivity and enzymatic profile of *Candida* species isolated from onychomycosis. Samples were obtained from the nails of 1810 patients and analysed at the Mycology Laboratory of Kayseris Hospital, Istanbul, Turkey. The nail scrap material was clarified with potassium hydroxide solution for direct microscopy and part of the scraping was seeded on Sabouraud-dextrose-agar medium containing gentamicin and cycloheximide as inhibitors of bacterial growth. From the 50 samples of the genus *Candida*, the identified species were: 23 *C. parapsilos*is (46%), 13 *C. albicans* (26%), 4 *C. guillermondii* (8%), 4 *C. tropicalis* (8%), 2 *C. krusei* (2%) and 1 *C. sake* (2%) and 1 *C. kefir* (2%). In the cases of onychomycosis in patients from Rio de Janeiro, the genus *Candida* represented 92.54% among other species of filamentous fungi causing onychomycosis [20].

The clinical characteristics and epidemiological profile of onychomycosis among military were studied by Casanova-Claure & Navarrete-Mejía, who considered onychomycosis as frequent nail infections and constitutes a public health problem, due to its frequency and morbidity, demonstrating that this disease is closely related to the working conditions of the military. A total of 67 military personnel of both gender attended the Dermatology Clinic of the Military Hospital of the city of Lima, Peru, were examined. The mycological diagnosis was performed through the culture of the ungual scraping, showing a positivity for *Trichophyton rubrum* in 23.9% and *Candida parapsilosis* in 4.5% of the examined samples [21]. The results found by these authors differ from those obtained by other researchers, as well as in our research, where the most frequent etiological agents of onychomycosis were from the genus *Candida*. Regional differences of the incidence profile by species are reinforced by the researchers Gupta *et al.*, [12]. Environmental factors are also relevant. There are several factors that indicate military as a risk group for mycotic infections associated with the inherent conditions and activities of this population. Among these conditions, the use of collective showers, nail trauma during military exercises, heat and body humidity, the use of military occlusive boots in the campaign are favourable conditions for the implantation and development of fungi, specially species which causes onychomycoses [21, 22].

Mendez-Tovar *et al.*, performed a research that consisted in determining the frequency of onychomycosis in a series of patients with psoriasis who were treated at the Dermatology Service of the Specialties Hospital of the National Medical Center of the Mexican Social Security Institute. They collected scales from the compromised nails, performed direct examination and culture in the medium of Sabouraud-dextrose-agar. Filamentous fungi were identified by morphological and cultural characteristics and the yeasts through biochemical tests. From the 150 patients with psoriasis, 83 (55%) presented nail abnormalities and, among them, 42 were diagnosed with onychomycosis. The investigators isolated 22 species of fungi, 11 (50%) form yeasts of the genus *Candida*, and the most frequent filamentous fungus was *Trichophyton rubrum* [23]. In our research, fungi of the genus *Candida* followed by *Trichophyton rubrum* predominated, but the frequency of the genus *Candida* (92.54%) was higher than the results found by these authors.

Salas and Gross, and Segal *et al.*, considered that *Trichophytum rubrum* stands out in the high prevalence as etiological agent of onychomycosis. According to these authors, in studies conducted in Costa Rica [24] and Israel [25], the fungus most frequently isolated was *T. rubrum*, and the incidence increases along the age of the patient [25]. This same fungus was cited by Vasquez & Padilla as the most frequent onychomycosis agent, especially in male gender [26]. In India, in a survey involving 300 people with onychomycosis, *Trichophyton rubrum* was indicated as the most prevalent agent [17]. In our research, *T. rubrum* figures as the second species with the highest prevalence (3.31%), but much lower than the predom inance of species of the genus *Candida* (92.54%).

Our research in the city of Rio de Janeiro allowed us to determine the genus *Candida* as the most frequent agent of onychomycosis in this population. Investigations conducted in Iran by Soltani *et al.*, also point to the genus *Candida* as the main agent of onychomycosis in that country [27].

Onychomycoses caused by non-dermatophyte fungi were reported by Kaur *et al.*, among patients from the Northern India. These investigators examined 100 samples of nail scraping for direct research and cultivation on Sabouraud-dextrose-agar. Filamentous fungi were identified by morphological and cultural characteristics and the yeasts through biochemical tests. Onychomycoses on the hands nails (57%) were more frequent than on the toenails (43%). Onychomycoses were caused mostly by filamentous fungi followed by dermatophytes and, in fewer cases, by yeasts [28].

The researchers Khuraiya *et al.*, investigated 150 cases of onychomycosis in patients attended at a dermatology outpatient clinic in the city of Jodhpur, India. The ungual scrapings were processed by usual techniques of laboratorial mycology: direct research after whitening and culture in Sabouraud-dextrose-agar medium. The results showed that the incidence of onychomycosis was 1.8% among the studied population and the highest prevalence occurred in the age class between 20 and 40 years. Among the dermatophytes, the isolated species were: *Trichophyton rubrum*, *Trichophyton verrucosum* and *Trichophyton mentagophytes*, *Candida albicans* and fungi of the genus *Rhisopus*, *Aspergillus*, *Mucor*, *Curvalaria*, *Penicillium* and *Fusarium*. The authors reported that onychomycosis was more incidental in housewives, farmers and workers dealing with cattle. The results pointed to *Candida albicans* and *Trichophyton rubrum* as the most frequent onychomycosis agents in the Indian studied population [29]. A similar result was found in our research among patients with onychomycosis in the city of Rio de Janeiro, Brazil.

CONCLUSIONS

It was verified that the fungal etiology of the ungual lesions in City of Rio de Janeiro has the incidence of 28,06% among 1290 patients. Species of the genus *Candida* were the prevalent agent, with 92,54% of the cases of onychomycosis. As reported by several researchers, the profile of fungi causing onychomycosis differs according to the studied population, and the knowledge of the prevalent agents is important for epidemiological and preventive strategies.

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