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Epidemiological Aspects of Onychomycosis

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Abstract Background: Onychomycosis is a chronic, difficult-to-treat fungal infection of the nail unit, often associated with prolonged therapy and high recurrence rates. Accurate identification of causative pathogens is essential for optimizing treatment outcomes. Objective: To analyze the epidemiological and etiological structure of onychomycosis using molecular diagnostic methods combined with histopathological confirmation. Methods: A total of 110 patients with clinically diagnosed onychomycosis were included (2022–2025). Nail samples were analyzed using multiplex real-time polymerase chain reaction (PCR) and histopathological examination with Schiff staining. Pathogen identification included dermatophytes, non-dermatophyte molds, and yeasts. Statistical analysis was performed using odds ratios (OR) with 95% confidence intervals (CI); $p < 0.05$ was considered significant. Results: Dermatophytes accounted for 63.7% of confirmed cases, followed by non-dermatophyte molds (20.1%) and yeasts (6.2%). The most common pathogen was Trichophyton rubrum (54.3%), followed by Trichophyton mentagrophytes (6.5%), Aspergillus spp. (7.0%), and Fusarium spp. (4.5%). Women had a significantly lower risk of dermatophyte infection (−70%, $p < 0.001$) but higher rates of infections caused by non-dermatophytes (2-fold increase) and yeasts (+50%). With increasing age, the prevalence of T. mentagrophytes, non-dermatophytes, and yeasts increased, while T. rubrum decreased. Patients older than 50 years showed higher susceptibility to infections caused by Aspergillus, Acremonium, Scopulariopsis, and Candida spp. Non-dermatophyte fungi were more commonly associated with superficial forms of onychomycosis and less frequently with distal or dystrophic forms. Conclusion: A substantial proportion of onychomycosis cases are caused by non-dermatophyte pathogens, particularly in women and elderly patients. The use of molecular diagnostics (PCR) combined with histopathology improves pathogen identification and supports personalized, evidence-based treatment strategies.

Keywords Onychomycosis, Mycological, PCR studies

1. Introduction

Onychomycosis represents a difficult-to-treat form of fungal infection [1,4,7,12,13]. The management of this chronic, slowly progressive disease is complicated by prolonged courses of therapy (often exceeding 5–6 months) and a high recurrence rate (25–30%). The global prevalence of onychomycosis is estimated at approximately 4%, with disproportionately higher incidence observed among certain population groups [1,4,9].

Despite the wide range of available treatment options, including oral and topical antifungal agents, over-the-counter remedies, and laser therapies, complete cure rates (defined as eradication of fungal infection with restoration of normal nail appearance) remain unsatisfactory. This underscores the need for an individualized approach to treatment [3,5,8,9,11].

Onychomycosis is predominantly caused by dermatophyte fungi, with Trichophyton rubrum being the most commonly identified pathogen. However, infections caused by non-dermatophyte molds and yeasts are considered unfavorable prognostic factors due to differences in antifungal susceptibility profiles (e.g., terbinafine versus itraconazole as first-line therapies), as well as diagnostic challenges that may delay the initiation of appropriate treatment [2,3,6,9,11].

Conventional diagnostic methods based on fungal culture have significant limitations, including prolonged turnaround time (2–4 weeks) and low sensitivity. In cases involving non-dermatophyte fungi, repeated culture isolation is often required, which may further delay treatment initiation [7,8,9].

In recent years, molecular diagnostic methods based on polymerase chain reaction (PCR) have become available for identifying fungal pathogens [9,10]. In particular, multiplex PCR assays enable the simultaneous detection of multiple causative agents of fungal infections within 1–2 days.

The combined use of PCR and histopathological examination is recommended as a reliable diagnostic approach, as it allows pathogen identification through direct visualization of fungal invasion of the nail plate.

The present study aims to investigate the application of molecular diagnostics using multiplex real-time PCR in combination with histopathological examination.

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2. Materials and Methods

Nail plate samples from 110 patients with various clinical forms of onychomycosis were analyzed. Inclusion criteria comprised samples that underwent confirmatory testing using multiplex real-time polymerase chain reaction (PCR) and histopathological examination as prescribed by a dermatologist. DNA extraction and multiplex real-time PCR were performed according to standardized protocols. The detection panel included dermatophytes, non-dermatophyte molds, and yeasts. The dermatophyte identification panel included the *Trichophyton rubrum* complex, *Trichophyton mentagrophytes*, *Microsporum*, and *Epidermophyton*. The non-dermatophyte panel included *Acremonium*, *Aspergillus*, *Fusarium*, and *Scopulariopsis*. The yeast identification panel included *Candida albicans*, *Candida parapsilosis*, *Candida tropicalis*, *Cryptococcus*, *Malassezia*, and *Trichosporon*.

Nail samples were stained using periodic acid–Schiff (PAS) staining and examined by a dermatopathologist. Types of infection were defined as subungual (infiltration of fungal elements limited to subungual keratin), superficial (infiltration of fungal elements through the superficial keratin layers of the nail plate), and dystrophic (infiltration of fungal elements involving both the nail plate and subungual keratin). The quantity of fungal elements was classified as rare (sparse fungal elements, mainly in the subungual region, with minimal or no association with nail keratin), minimal (<10% involvement of nail keratin), moderate (10–80% involvement of nail keratin), and severe (>80% involvement of nail keratin).

A positive, mycologically confirmed diagnosis of onychomycosis was defined as a positive PCR result with histopathological confirmation of fungal invasion. For patients with multiple collected samples, only the first sample was considered. Analysis of histopathological findings included all samples. Mixed infections were not analyzed due to the inability to attribute histopathological findings to a specific detected fungal agent.

Data processing and analysis were performed using Microsoft Excel. Patient demographic characteristics (age, sex, etc.), PCR results, and histopathological findings were presented in tabular form. Associations between patient characteristics, pathogen identification results, and histopathological findings were quantitatively assessed using odds ratios (OR) with 95% confidence intervals (CI). A p-value <0.05 was considered statistically significant.

3. Results

The analysis of 110 patients with onychomycosis demonstrated that dermatophytes consistently accounted for the largest proportion of mycologically confirmed diagnoses, comprising 63.7% (range 55.4–69.0%), followed by non-dermatophyte molds at 20.1% (range 14.6–30.7%) and yeasts at 6.2% (range 4.5–6.8%).

The most frequently identified dermatophytes were representatives of the *Trichophyton rubrum* complex, followed by *Aspergillus* and *Fusarium*, with *Candida parapsilosis* detected less frequently.

In women, the likelihood of dermatophyte onychomycosis was 70% lower than in men ($p < 0.001$). In contrast, women had a twofold higher likelihood of infection caused by non-dermatophyte fungi ($p < 0.001$), and the probability of yeast-associated onychomycosis was 50% higher compared to men.

An age-dependent increase in the frequency of onychomycosis caused by the *Trichophyton mentagrophytes* complex, non-dermatophytes, and yeasts was observed. Among dermatophytes, patients older than 50 years had a 50% lower likelihood of infection caused by the *T. rubrum* complex compared to younger individuals (18–40 years). Conversely, in this age group, the likelihood of infection caused by the *T. mentagrophytes* complex increased nearly fourfold.

An increase in the frequency of onychomycosis caused by non-dermatophyte fungi with advancing age was also noted. Specifically, in patients older than 50 years, *Aspergillus*, *Acremonium*, and *Scopulariopsis* were detected more frequently, whereas *Fusarium* did not show a similar increase compared to younger patients.

Regarding yeasts, both *Candida albicans* and *Candida parapsilosis* were more frequently identified in individuals older than 50 years compared to younger patients. It should be noted that non-dermatophyte fungi and yeasts were more likely to cause superficial infections with characteristic clinical presentation and less likely to result in dystrophic forms of onychomycosis.

4. Discussion

In clinical practice, treatment decisions for patients with onychomycosis often do not take into account the identification of the causative pathogen [3,5,9]. This is partly due to the limited use of confirmatory diagnostic tests and existing methodological limitations. Although infections caused by non-dermatophyte molds and yeasts are less common in patients with onychomycosis, they are considerably more difficult to treat [2,5,11].

The present study provides a detailed analysis of the pathogen spectrum in patients with onychomycosis based on PCR diagnostics, with confirmation achieved through histopathological evidence of fungal invasion.

Overall, the findings of this study demonstrate that a substantial proportion of mycologically confirmed cases of onychomycosis are caused by less commonly recognized pathogens, which disproportionately affect female patients and older individuals. Contrary to current perceptions, women, who are often underrepresented in clinical studies, should not be considered a low-risk group for fungal infections.

Although older individuals are generally at higher risk for onychomycosis, they are less frequently affected by the

dominant dermatophyte pathogen (*T. rubrum* complex).

The primary goal of onychomycosis treatment should be the complete eradication of fungal infection. Rapid diagnostic methods, such as PCR, enable timely pathogen identification, while histopathological assessment allows characterization of the infection. Together, these approaches support clinicians in selecting individualized, evidence-based treatment strategies tailored to specific clinical conditions.

5. Conclusions

Dermatophytes remain the leading cause of onychomycosis; however, non-dermatophyte molds and yeasts account for a significant proportion of cases, particularly in women and older patients.

Multiplex PCR combined with histopathology enables rapid and accurate pathogen identification, supporting targeted therapy.

Implementation of molecular diagnostics can improve treatment outcomes and facilitate personalized, evidence-based management of onychomycosis.

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