Contrat doctoral – ED Galilée

Titre du sujet: Microbial and clinical epidemiology, drug susceptibility/resistance, gene mutations associated with antifungal resistance and molecular phylogeny of dermatophytes isolated from dermatophytosis in Iran and France.

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✓ **Discipline de doctorat :** Sciences de la vie et de la santé

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✓ **Mots clés :** Dermatophytosis ; *Trichophyton indotineae* ; Antifungal resistance ; France ; Iran

Hypothesis: The growing resistance of dermatophytes to terbinafine and azoles, driven by genetic mutations, contributes to increase treatment failure and complicates clinical management of dermatophytosis. Exploring the species distribution, drug resistance patterns, and genetic mutations of dermatophytes in two different geographical contexts, Iran and France, will provide insights for effective treatment protocols and prevention strategies.

Background: Superficial fungal infections, notably dermatophytosis (ringworm or tinea), are common in the community, and affect millions globally. Dermatophytes, particularly the genera *Trichophyton*, *Microsporum*, *Epidermophyton*, and *Nannizzia*, are the primary agents, leading to infections in keratinized tissues such as skin, hair, and nails. corporis, tinea pedis, tinea unguium, and tinea capitis. The most prevalent dermatophyte species responsible for human infections include *T. rubrum* and *T. mentagrophytes* complexes. Terbinafine, an allylamine antifungal drug, is a first-line oral treatment for dermatophytosis. It works by inhibiting the enzyme squalene epoxidase (SQLE), thereby blocking ergosterol synthesis and leading to fungal cell death. Recently, terbinafine-resistant dermatophytes, especially in the *T. mentagrophytes* complex, have emerged globally. Molecular studies have revealed that single nucleotide polymorphisms (SNPs) in the *SQLE* gene cause high resistance to terbinafine. Notably, a new genotype of *T. mentagrophytes* (genotype VIII), recently named *T. indotineae*, has emerged in India, displaying marked terbinafine resistance (1). The intercontinental transmission of this emerging pathogen is significant, as in the past five years, it has expanded to Europe (2), Americas and South-East Asia (). Terbinafine resistance in *T. rubrum* has also been increasingly reported globally, including in France and Iran, linked to mutations in the *SQLE* gene, responsible for substitutions (3).

Besides terbinafine, azoles, including imidazoles and triazoles, are widely used as antifungal agents for treating fungal infections like dermatophytosis. These drugs inhibit fungal cytochrome P450 14α-lanosterol demethylase, impairing ergosterol synthesis, a crucial component of fungal cell membranes. However, resistance to azoles has been increasingly observed in *T. rubrum* and other dermatophyte species. Various mechanisms contribute to this resistance, with the overexpression of efflux pumps being a key factor. Furthermore, azole resistance is more frequently reported in the emerging species *T. indotineae*. Studies show that reduced susceptibility of *T. indotineae* strains to azoles is primarily due to the overexpression of the *CYP51B* gene (*TinCYP51B*), which encodes a sterol 14α-demethylase.

This growing antifungal resistance represents a challenge for treating dermatophytosis, emphasizing the need for continued surveillance and development of alternative treatment options. The rapid global spread of terbinafine- and azole-resistant dermatophytes is a significant concern due to the limited alternative treatment options for resistant dermatophytosis. Thus, dermatophytosis pose a substantial public health

burden in both developed and developing regions, including countries such as Iran and France, with growing concerns about antifungal resistance and difficulties in ensuring effective treatment.

Main Objectives

1. Characterize dermatophyte species and determine frequency: identify and analyze the dermatophyte species isolated from patients in Iran and France to understand species distribution and prevalence. Conserve known isolates for further testing.
2. Evaluate antifungal susceptibility and resistance: assess the susceptibility and resistance profiles of isolated dermatophytes to commonly used antifungal drugs.
3. Detect genetic resistance patterns: identify genetic mutations associated with resistance to terbinafine and azoles in isolates obtained from patients in both countries.
4. Study phylogenetic relationships: explore the phylogenetic relationships of dermatophyte species identified in Iran and France to gain insights into their evolutionary connections.
5. Map the distribution of resistant Isolates.
6. Correlate species with resistance and clinical factors.

Methodology

1. Sample collection and identification: dermatophyte isolates will be collected from infected patients' skin, hair, and nails in Iran and France. Classical morphological identifications will be confirmed by molecular tools using ITS-PCR sequencing.
2. Antifungal susceptibility testing: Drug susceptibility of dermatophytes will be evaluated following the European Committee on Antimicrobial Susceptibility Testing (EUCAST) standards, with a focus on terbinafine and azoles resistance. Screening of resistance will be performed by terbinafine containing agar method (TCAM) and antifungal strips.
3. Genetic analysis of resistance: using Sanger sequencing of *SQLE* gene and NGS, mutations associated with antifungal resistance genetic mechanisms linked to terbinafine and azoles resistance will be identified in key resistance-associated genes.
4. Phylogenetic and population structure analysis: phylogenetic relationships and strain clustering will be analyzed to understand epidemiological links between Iran and France using MLST, microsatellite typing, New Generation Sequencing, and bioinformatics tools for population genetics.
5. Analysis of the geographic distribution and spread of antifungal-resistant strains between the two countries.
6. Analysis of the associations between dermatophyte species, antifungal resistance patterns, clinical presentations, and patient demographics.

References

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