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Short Communication

Single strains of *Trichophyton rubrum* in cases of tinea pedis

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Multiple colonies of *Trichophyton rubrum* were isolated from single skin specimens from 10 patients with tinea pedis and were typed using a PCR-based analysis of repeats in the rRNA intergenic spacer. In each case only a single strain type of *T. rubrum* was isolated, suggesting a monotypic aetiology of tinea pedis. This is in contrast to the multiple strains previously shown to be involved in many cases of onychomycosis.

Introduction

The vast majority of chronic dermatophyte infections of human skin are caused by *Trichophyton rubrum*, particularly tinea pedis (or athlete's foot), the most common dermatophyte infection worldwide (Rippon, 1988; Weitzman & Summerbell, 1995). Exposure to the causal fungus on the floors of communal bathing areas is known to be an important risk factor for developing tinea pedis and as the number of sports and leisure facilities increases, a larger number of individuals will become infected. In order to improve our understanding of the epidemiology of *T. rubrum* infections it is important to be able to distinguish between different strains. We have recently developed a typing method for *T. rubrum* (Jackson *et al.*, 1999, 2000) based on a rapid and sensitive PCR-based assay of the copy number of two tandemly repetitive subelements (TRS-1 and TRS-2) in the rDNA non-transcribed spacer (NTS) region. This technique has demonstrated that molecular diversity is detectable in a highly homogeneous species like *T. rubrum* (Jackson *et al.*, 2000).

In an initial study of isolates from patients with onychomycosis due to *T. rubrum*, we were surprised to find that in most cases nails were infected by multiple (⩽3) strains of *T. rubrum* (Yazdanparast *et al.*, 2003). Since onychomycosis is a relatively common sequela of untreated tinea pedis (Roberts *et al.*, 1990) we were interested in looking at the number of *T. rubrum* strains from patients with tinea pedis.

†Deceased

In the present study we determine whether tinea pedis in individual patients is caused by a single or multiple strains of *T. rubrum*.

Methods

Patients. This study involved 10 patients, from four cities in France, with tinea pedis and without onychomycosis, prior to antifungal therapy.

Isolation of dermatophytes. *T. rubrum* was isolated from samples of skin by culturing on Sabouraud agar at 27 °C. Up to five colonies of *T. rubrum* from each culture plate were individually subcultured on Sabouraud agar for 10 days at 27 °C to ensure purity.

DNA extraction. Individual *T. rubrum* isolates were cultured on the surface of Sabouraud broth in a Petri dish for up to 7 days at 27 °C. Mycelia were harvested with a sterile pipette tip and dried on filter paper. DNA was extracted from this fungal material as described previously (Jackson *et al.*, 2000).

PCR amplification. PCR amplification was performed as described by Jackson *et al.* (2000) using primers for both TRS-1 and TRS-2. The PCR products were electrophoresed in a 2 % agarose gel in the presence of ethidium bromide, visualized under UV light and photographed. Types were defined based on the size of the PCR product (Jackson *et al.*, 2000).

Results and Discussion

In this study all of the *T. rubrum* isolates from the one specimen taken from each patient were a single strain. DNA from each isolate was examined at two loci, TRS-1 and TRS-2, generating a combined type with an arabic numeral defining the TRS-1 type and a roman numeral defining the

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Table 1. Molecular types of isolates of *T. rubrum* from cases of tinea pedis

Patient no.	No. of isolates (colonies)	TRS-1 type	TRS-2 type
1	5	1	I
2	3	1	I
3	5	1	I
4	5	1	II
5	2	1	I
6	5	1	I
7	5	1	I
8	5	1	II
9	5	1	II
10	5	1	II

TRS-2 type, e.g. 1/II. Six out of the 10 of patients were infected with type 1/I and the rest of them were infected with 1/II (Table 1).

PCR-typing of *T. rubrum* on the basis of copy number of TRS-1 and TRS-2 is a simple and rapid method. The study of *T. rubrum* isolates from skin specimens in 10 patients showed the same PCR pattern in all the isolates from the same specimen, which suggests that a single strain of *T. rubrum* infects an individual. This is in contrast to our study on nail infections where six of 10 specimens yielded more than one strain when analysed in a similar way (Yazdanparast *et al.*, 2003).

While the previous study of strain diversity in onychomycosis was carried out on UK patients (Yazdanparast *et al.*, 2003), and the patients in this study were from France, the extent of geographic distribution of the patients in the two studies was similar and this difference was not thought likely to account for the difference in strain diversity observed. This difference in diversity between strains of *T. rubrum* causing onychomycosis and tinea pedis may reflect the duration of infection. However, data on the duration of infection in these patients was not available in this study or for the previous study of onychomycosis in order to corroborate this idea. Alternatively, this difference may be due to a fundamental difference in the pathogenesis of *T. rubum* infections in these two tissues. Further studies will be required to confirm these results, which have important implications for the numbers of isolates required for typing in epidemiological studies.

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