

Reference

202

Identification of clinical isolates *Trichophyton rubrum* using a rapid and accurate mass spectral analysis (MALDI-TOF ICMS)

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Keywords: Clinical Mycology, Dermatophytes, MALDI-TOF ICMS**Abstract**

Dermatophytes are keratinolytic fungi that are responsible for the commonest dermatologic condition called “ringworm” in man. The affinity for keratinized tissues by dermatophytes, implies in most of the cases, that the infection remains restricted to the nonliving cornified layers of the skin, nails, and hair [1]. Among dermatophytes, the species *Trichophyton rubrum* is of particular clinical interest for man because is the most common agent of human dermatophytoses. Macro and micro-morphological examination combined to physiological analysis of primary isolates grown in selective culture media are still the most used methods in routine laboratory. Besides their low specificity, an accurate diagnosis may take 3 to 4 weeks to be achieved. Modern identification methods involve molecular biology by using PCR technology based on differential sequence elements. It is gradually becoming clearer that microbial identification and authentication requires a polyphasic approach to generate quality data which are accurate and useful [2]. Microbial mass spectral analysis has been progressively more incorporated to the polyphasic approach to improve the accuracy of the microbial identification issue. Matrix Assisted Laser Desorption Ionization Time of Flight Intact Cell Mass Spectroscopy (MALDI-TOF ICMS) is becoming an alternative to DNA-dependent methods so it has been already successfully applied to the rapid identification and classification of microorganisms [3]. The aim of this work was to test the applicability of MALDI-TOF ICMS for identifying clinical isolates of *T. rubrum*. In this study twenty clinical isolates of *T. rubrum* were grown on Sabouraud culture medium. Plates were incubated for 7 days at 25 °C. All the isolates were identified both macroscopically and microscopically. From the same plate, a tiny sample (about 50 mg) was transferred to stainless steel templates. A 0.5 ml of dihydroxy-benzoic acid (DHB) matrix solution was added to the sample and air dried. Peak lists of individual samples were compared with the superspectra database generating a ranked list of matching spectra from SARAMIS software. All strains were accurately and consistently identified as *T. rubrum* by MALDI-TOF ICMS combined to SARAMIS database analysis. Spectral mass analysis proven to be a rapid method, as the analysis took only a few minutes to perform with the benefit of any laborious sample preparation procedures or any expensive chemical reagent was needed.

[1] Rippon, JW. (1988) Saunders (W.B.) Co Ltd. Philadelphia, USA.

[2] Keys, C.J., Dare, D.J., Sutton, H., Wells, G., Lunt, M., McKenna, T., McDowall, M., Shah, H.N. (2004) Infect Genet Evol, 4, 221-242.

[3] Santos, C., Paterson, R.M.R., Venâncio, A., Lima, N. J. Appl. Microbiol (In Press).