

Identification of *Malassezia* species isolated from the skin of patients with atopic dermatitis, psoriasis, and healthy volunteers by phenotypic and molecular typing methods

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Introduction & objectives

The yeasts of the *Malassezia* genus belong to the physiological cutaneous microflora of humans and other warm-blooded animals. These fungi have been associated with several dermatological pathologies of world-wide distribution, including pityriasis versicolor (PV), seborrheic dermatitis (SD), atopic dermatitis (AD), and psoriasis.

Currently, 14 *Malassezia* species are recognized, all but one being lipid-dependent species.

The aim of the study was to investigate the distribution of *Malassezia* spp. on the skin of healthy volunteers and patients with AD and psoriasis, by using a combination of conventional, phenotypic tests and molecular methods, namely PCR-restriction fragment length polymorphism (RFLP) analysis and sequencing analysis, both targeted towards the rDNA cluster of the *Malassezia* genome.

Materials & Methods

A total of 20 *Malassezia* sp. isolates, originating from 4 patients with AD, 4 patients with psoriasis, and 4 healthy volunteers were included in the study. The isolates were recovered from 1, 2 or 3 different body sites (scalp, face, chest, and back) in case of 5, 6, and 1 subjects, respectively. The yeasts were cultured on modified Dixon agar. Primary species identification was done with phenotypic methods, based on morphological and physiological criteria. Molecular identification involved 2 PCR-RFLP assays, targeting the internal transcribed

spacer 2 (ITS2) and partial 26S rRNA gene, along with sequence analysis of the ITS region and D1/D2 domains of the 26S rRNA gene.

Results

The most frequently identified species was *M. sympodialis*, having been cultured from 16 (80%) skin samples of 11 subjects. Two other species, namely *M. furfur* and *M. slooffiae* were isolated from 6 samples of 2 psoriatic patients, and 2 samples of a healthy subject, respectively. *M. sympodialis* was the only species to be observed among AD patients and the predominant species among healthy individuals. In psoriatic patients, *M. furfur* predominated over *M. sympodialis*. A co-culture of 2 species, i. e. *M. furfur* and *M. sympodialis* or *M. slooffiae* and *M. sympodialis* was demonstrated in 2 samples of psoriatic patients and 1 sample of a healthy subject, respectively. The results of phenotypic and molecular identification methods were in complete agreement in 70% (14/20 cases). The discrepant results included 3 isolates of *M. sympodialis*, initially identified as *M. slooffiae*, and 3 mixed-species isolates, formerly identified as one of 2 co-occurring species.

Conclusions

The most prevalent species, found in all subject groups, was *M. sympodialis*. All isolates from AD patients were *M. sympodialis*, whereas *M. furfur* was only observed in psoriatic patients. Whether this reflects any predilection of particular *Malassezia* species for certain clinical conditions needs further evaluation. For the identification of *Malassezia* spp., molecular methods should preferably be used, as their results are reliable and straightforward.