Original article

ISOLATION AND IDENTIFICATION OF MALASSEZIA SPP. IN PYTIRIASIS VERSICOLOR IN KASHAN, IRAN

Rezvan Moniri1, Mehdi Nazeri2, Shokouh Amiri3, Babak Asghari4

ABSTRACT

Objective: Pityriasis versicolor (PV) is a chronic superficial fungal disease caused by Malassezia spp. The incidence is as high as 30-40% in tropical climates. Epidemiological data suggest geographical variations in the rate of the isolated species from PV. Our aim was to identify Malassezia spp. from PV patients in Kashan, Iran.

Methodology: Isolates of Malassezia were collected from 118 PV patients (75 males and 43 females). A direct microscopy with KOH and methylene blue was carried out. Cultures were made in modified Dixon agar medium and the isolates were identified by macroscopic and microscopic features, physiological characteristics (catalase test) and biochemical criteria (esculin and lipid assimilation tests). Data were analyzed statistically by software SPSS (version 11) and Fischer’s exact and descriptive statistical tests.

Results: The average age of 118 patients in this study was 28.42±8.53 years. The percentages of patients in this study were 64.4 and 35.6 for men and women respectively. Hyperhidrosis was reported as the most important finding with 58.1%. Back (42.2%) and extremities (7.4%) were the highest and the lowest involved parts respectively. The isolates found were M. globosa (43.8%), Malassezia furfur (38.4%), M. obtusa (9.8%), M. sympodialis (6.3%), and M. slooffiae (1.7%).

Conclusion: From these findings it was suggested that M. globosa presents the main species implicated in the pathogenicity of PV and M. furfur as the second agent of importance.

KEY WORDS: Malassezia spp, Pityriasis versicolor.

INTRODUCTION

Pityriasis versicolor (PV) is a superficial infection of the stratum corneum caused by Malassezia spp.,1 pathological change occurred by inocu-
on the face increased for one month after birth, and started to decrease four months later in parallel with the amount of sebum. Until recently, *M. furfur*, *M. pachydermatis*, and *M. sympodialis* were the only members of the genus, but in 1996, Gueho et al. added four new species, *M. globosa*, *M. obtusa*, *M. restricta*, and *M. slooffiae*, based on their morphology, ultrastructure, physiology, and molecular biology. Recently, four new species were isolated, namely *M. dermatis*, *M. japonica*, *M. yamatoensis* and *M. nana* in Japan. Malassezia species are associated with PV, Malassezia folliculitis, seborrheic dermatitis (SD), dandruff, atopic dermatitis (AD), and CRP. Research attention has focused on the relationship between Malassezia species and skin disease. Until recently, PV was a superficial infection believed to be caused by *M. furfur*. Recent studies using morphological features and physiological tests indicate that *M. globosa* may actually be the causative agent, but the *M. globosa* rate of culture changed with reporters from 53 to 97%. On the other hand, Gupta et al. reported that *M. sympodialis* was detected at 60%. Nakabayashi reported that *M. globosa* was isolated from 55% of lesional skin specimens in PV, while other species were below 10% according to the method devised by Guillot et al. The present study was aimed to identify the Malassezia spp. causing pityriasis versicolor in a Kashan population in Iran.

**METHODOLOGY**

This descriptive study was conducted from August 2006 to October 2007. The samples were collected from patients with pityriasis versicolor in Kashan, Iran and processed in the mycology laboratory of the department of microbiology. Mycological evaluation by microscopic examination of KOH treated skin scrapings and methylene blue staining were done and 118 were cultured. The scales were inoculated into modified Dixon’s agar (mDixon’s agar) as described by Guillot et al and into Sabouraud dextrose agar containing 0.05% chloramphenicol and 0.05% cycloheximide (SDA). The tubes were incubated at 32°C for 3-4 days. The morphology of the yeast cells was studied by making Gram stained smears of the isolates from mDixon’s agar after one week incubation at 32°C. Ethical approval for the study was obtained from Research and Ethics Committee of the Kashan University of Medical Sciences.

**RESULTS**

The average age of 118 patients (76 males and 42 females) in this study was 28.42±8.53 years. Characteristics of patients with pityriasis versicolor in Kashan, Iran according to sex and age group shown in Table-I.

One hundred fourteen (94.2%) skin scrapings showed hyphae and spores in the KOH preparation. Growth was obtained on mDixon’s agar 112 out of 121(92.6%) skin scrapings. The isolates found were *M. globosa* (43.8%), *Malassezia furfur* (38.4%), *M. obtusa* (9.8%), *M. sympodialis* (6.3%), and *M. slooffiae* (1.7%). There was no statistically significant association between Malassezia spp. and demographic characteristics and clinical characteristics of the patients. Hyperhidrosis was reported as the most important finding with 58.1%. The frequency rate of patients with pityriasis versicolor according to site of lesion are shown in Table - II.

**DISCUSSION**

In our study, out of the 118 specimens that were inoculated, 92.6% yielded growth of *Malassezia* in culture. Out of this, the most frequently isolated species was *Malassezia globosa* (43.8%), followed by *M. furfur* (38.4%), *M. obtusa* (9.8%), *M. sympodialis* (6.3%), and *M. slooffiae* (1.7%).

**Table-I:** Characteristics of patients with pityriasis versicolor in Kashan, Iran according to sex and age group

<table>
<thead>
<tr>
<th>Age group</th>
<th>Female No. (%)</th>
<th>Male No. (%)</th>
<th>Total No. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>15 &lt; years</td>
<td>4(2.6)</td>
<td>2(4.8)</td>
<td>6 (3.4)</td>
</tr>
<tr>
<td>16-30</td>
<td>36(85.6)</td>
<td>45(59.2)</td>
<td>81 (68.6)</td>
</tr>
<tr>
<td>31-45</td>
<td>2(4.8)</td>
<td>23(30.3)</td>
<td>25 (21.2)</td>
</tr>
<tr>
<td>&gt;45</td>
<td>2(4.8)</td>
<td>6(7.9)</td>
<td>8 (6.8)</td>
</tr>
<tr>
<td>Total</td>
<td>42(100)</td>
<td>76(100)</td>
<td>118 (100)</td>
</tr>
</tbody>
</table>
42.2% of isolates, predominating in the back skin.

Crespo et al. reported that M. globosa was recovered from 97% of their patients, alone in 60% of them and associated with M. sympodialis in 29% and M. slooffiae in 7%. Crespo Erchiga et al. showed that in pityriasis versicolor, Malassezia globosa was found in 84% of cases, alone or associated with Malassezia sympodialis, which was by far the commonest species in normal skin (91.7% of isolates, predominating in the trunk skin). Kindo et al., described that out of 70 scrapings 48 (68.75%) showed growth on mDixon’s agar. The commonest isolate was M. sympodialis (28, 58%) followed by M. globosa (19, 40%) and one isolate was (2%) of M. restricta. Salah et al. reported that Malassezia globosa was the predominant species in lesional skin of PV (65%). It was isolated alone in 47% of cases and associated in 18% with M. furfur (13%) or M. sympodialis (5%). In healthy skin M. globosa was found alone in 7.77% and associated in 15.54%, respectively, with M. furfur (4.44%), M. sympodialis (4.44%), M. restricta (3.33%) and M. slooffiae (1.11%). Crespo-Erchig et al. indicated that Malassezia globosa is the predominant species found in the lesions of pityriasis versicolor, at least in temperate climates.

In a study by Krisanty et al. the isolates found were Malassezia furfur (42.9%), M. sympodialis (27.5%), M. globosa (13.3%), M. slooffiae (7.7%), M. obtusa (7.7%) and M. restricta (2.2%), and 7.14% specimens were unidentified. Karakaš et al. reported that 45.4% of the patients showed Malassezia spp. in culture and Malassezia globosa (47.7%) was the most commonly isolated species followed by Malassezia furfur (36.4%) and Malassezia slooffiae (15.9%). In this study, the most common isolated species in PV lesions was M. globosa, which is in agreement with the majority of studies worldwide. This was contrary to observation of Krisanty et al., which isolated M. furfur and M. sympodialis as the predominant species in PV lesions.

From these findings it was suggested that M. globosa presents the main species implicated in the pathogenicity of pityriasis versicolor and M. furfur as the second agent of importance, and predominating site of infection was in the back skin.

ACKNOWLEDGEMENTS

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REFERENCES