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ASPECTS ON ISOLATION AND IDENTIFICATION OF MICROSPORUM CANIS STRAINS FOUND IN DERMATOZOONOSIS

Adrian Buruiana[1] Aurel Ardelean[1,2]
[1]Faculty of Natural Sciences Western University of Arad,
[1,2] Institute of Life Sciences Western University of Arad

Abstract

In the practice of mycology laboratory diagnosis of skin fungal infections we identify the pathogenic species of dermatophytes from tinea lesions. Most of the identification is based on macro and microscopic view, but not all strains have as characteristic in the primary isolate the aspects which usually provide identification. A series of additional tests is needed to ensure proper identification of the species incriminated in skin pathology. From this point of view, at the genus Microsporum, the autoclavated rice gave excelent results to the six atipical strains obtained from the clinical isolates, acquiring thus a correct identification of the *M. canis* species.

Keywords: *Microsporum canis*, isolated atypical, autoclaved rice, organs of fructification.

Introduction

The species Microsporum canis belongs to the genus *Microsporum* of *Arthrodermataceae* family, in order to *Onygenales* in the kingdom Fungi, Ascomycota filumul. The teleomorph of the species is also known as *Arthroderma otae*.

It is a dermatophyte belonging to the zoophyle ecological niche, with a worldwide distribution, being for the first time isolated by Bodin in 1902. Its primary habitat is the cat (which may be saprophyte) but it can also be the dog. From these animals the human can easily be contaminated and the lessions that this keratolinic fungus produces are clinicaly represented by the tinea corporis (in children and adults) and by the tinea capitis (in children up to 14 years old). The isolation and identification of the lesions is the responsibility of the laboratory of medical mycology that can diagnose only by dermatozoonosis species identification. Phenotypic species identification approach in clinical isolates is not easy. As we will see further on, there is a series of phenotypic aspects atipical for this species. It is therefore advisable to use a series of tests to ensure correct identification of this species. We aimed to evaluate the prevalence of this and to apply some additional tests as well as to higlight their efficiency in identification. At the 6 atypical strains isolated we applied cultivation on autoclaved rice in order to obtain identification. The method gave good results. In all cases of atypical

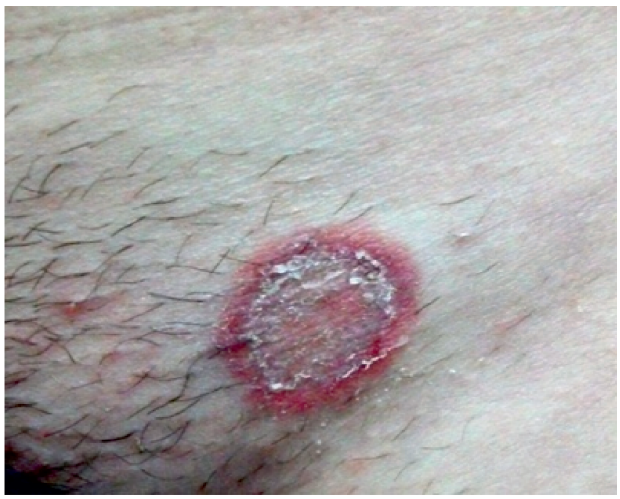

Fig. 1 Tinea corporis caused by Microsporum canis

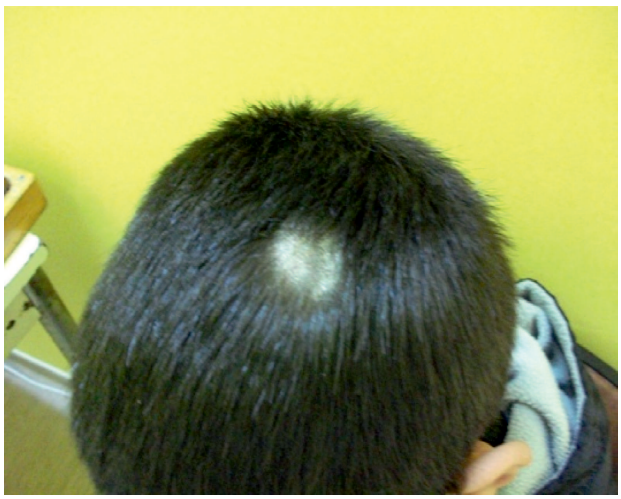

Fig. 2 Tinea capitis caused by Microsporum canis

strains results which allowed correct identification of species were obtained.

Materials and methods

50 strains were isolated issues that were characteristic of the species M. canis, 42 from tinea corporis lesions (Fig.1), 8 of tinea capitis (Fig.2).

Primary isolation was obtained for the Emmons modified Sabouraud medium. During development following macroscopic aspects of colony-texture, color, relief, pigments, and highlight the primary organs of fructification. The species identification was obtained from 44 strains macro and microscopic aspects had absolutely phenotypic characteristic summative play below:

Development time: slowly, in 4-5 days with characteristic appearance in 10-14 days.

Macroscopic appearance: fluffy colony, with long down the tube wall rises, easily removable medium, when the colony becomes grey. The dust colonies and young stars are part white with a smooth central area, mature colonies were textured with radial striations and woolly fringe issue, obverse and reverse of the colony is white to yellow to orange. (Fig.3a,b,c,d)

Microscopic appearance: macroconidia : are typically spindle-shaped, echinulate with 7-14 boxes with thick walls; microconidia are not constant when present pyriform to clavate or sessile are rare, placed laterally on hyphae, filaments with septated mycelia micelles in the missile issue, and chlamydospores in the spheroidal corpora missile (Fig. 4).

All strains have the ability to produce hair perforation.

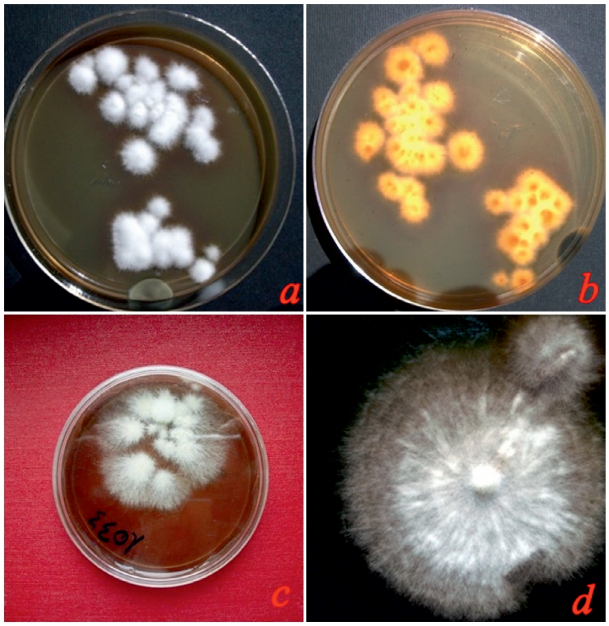

Fig. 3 Culture of Microsporum canis a.front view of culture; b. revers view of culture ; c. front view of culture d.close front view of culture, woolly aparence.

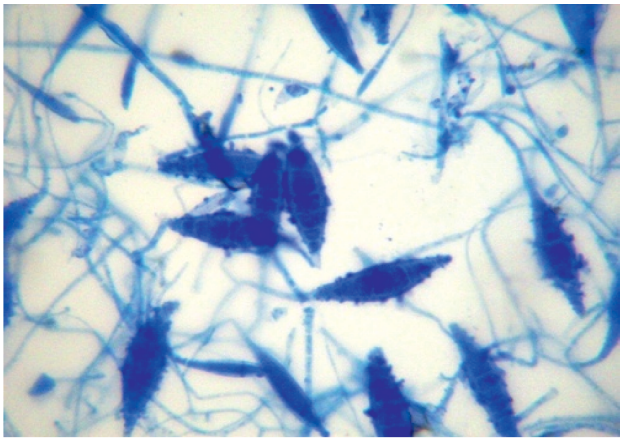

Fig.4 Microscopic appearance of M. canis macroconidia sharp at both ends, echinulate

Who had atypical strains have been six issues of which four were macroscopic but somehow lacked the characteristic fructification bodies and two that presented a glabous aspect that has not changed over time, and did not have bodies to identify the strain.

Glabrous colonies of this species are cited in the literature of specialists (Midley et al., 1997). It is noted that after a certain period of time, they revert to the typical form, the two cases also had such a colony we have not seen this, it is maintained as part glabros after 60 days of incubation.

Fig. 5 Glabrous variant coloy of M.canis secies

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Fig. 6 Dysgonic strain of M.canis secies

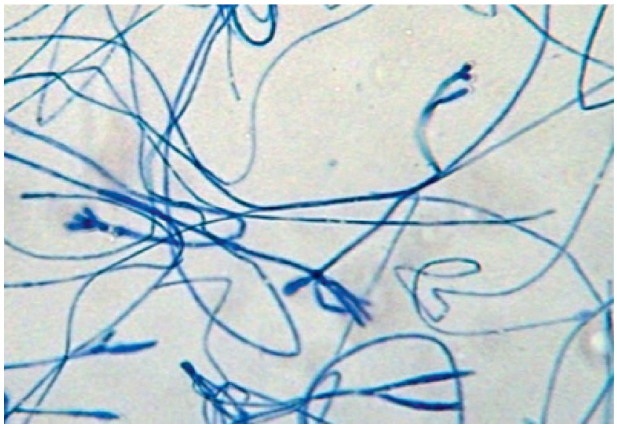

Fig. 7 Microscopic view of glabrous variant coloy of M.canis secies, macroconidia microconidia are often not produced 400X

From the microscopic point of view glabrous colonies were composed of sterile mycelium, sometimes dichotomously branched formations containing terminal-looking antlers, being totally absent of both macro and microconidiile (Fig. 7).

Rice cultivation is recommended especially when strains genus Microsporum doesn't get fructification bodies to ensure the identification of species (Ellis et al., 2007).

The six atypical strains were grown on autoclaved rice (8g rice in 25 ml distilled water, autoclave 120 ° C 15 minutes) to obtain and build upon all demonstrated after incubation at room temperature both macro and microconidii typical. In all six strains of rice, Microsporum canis grew lush and rich air mycelium producing a yellow pigment (Fig. 8).

Microscopic preparations made with fingerprinting method showed the presence of characteristic macro- and microconidia and extremely abundant (Fig.9).

Was obtained for the drilling and testing of the hair which showed all the organs perforatorii 6 cases of atypical cultures.

Fig. 8 Sub-cultures on autoclavated rice grains to stimulate sporulation

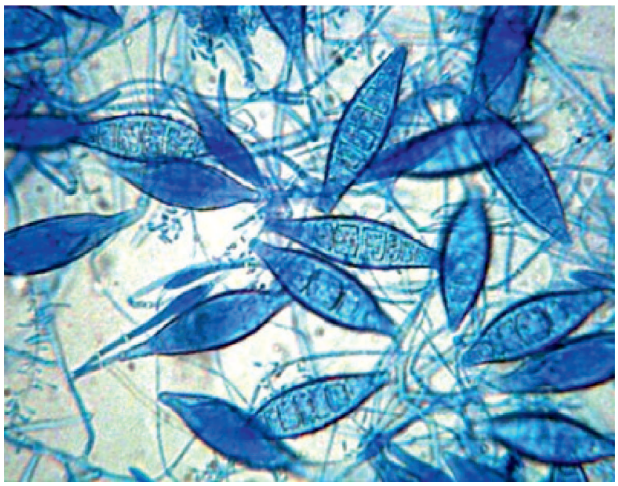

Fig. 9 Macroconidia are typically spindle-shaped on autoclavated rice

To highlight hair perforation bodies were used for human blond hair from prepubertar chaild which were placed directly on the colonies of dermatophytes, and examinated one week to highlight the hair perforation bodies that are formed. Hair lactofenol was placed in lactophenol aniline blue in preparation under the slide, and examined microscopically dry aperture of optical microscopy.

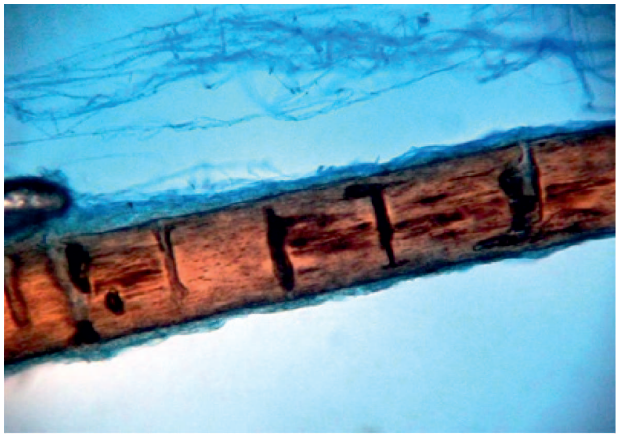

Fig.10 Positive hair perforation test for atipical strain of M. canis

Results and discussion

Identification of dermatophytes fall course in mycology laboratory task but the mere isolation medium Sabouraud not always ensures a correct identification of the species involved in pathology. Species of biolologic dermatophytes Diagnosis is based on macro and microscopic aspects characteristising anamorph dermatophytes. Differential diagnosis of biological species based on species belonging to the Microsporum genus, highlighting the characteristic microscopic macroconidiilor (Ellis et al., 2007). The absence of the macroconidia is found in antropofil species like Microsporum audouinii, which are usually only identified providing terminal chlamydospores (Cojocaru, 1979). Colonies of *M. canis* have some macro and microscopic characteristics, which ensures the identification mentioned above; there is variability from isolate to isolate which may be called and the adaptation to parasitism in saprofitism under cultivation in vitro. The absence of microscopic criteria makes possible the confusion with other species or species. Aspect glabrous colonies are considered by some authors to be unstable turning into normal colonies after a certain period of time (Midley et al., 1997). At the two presented glabrous strains we found this is irreversible and they remained constant after 60 days of incubation. Such atypical isolates of *M. canis* can be easily mistaken for *M. audouinii*. It is therefore needed the use of other methods to ensure the differential proper identification. Rice cultivation has given very good results and it is recommended to use it when there are difficulties in identification. It has a range of benefits: primarily the low cost of nutrient substrate and excellent results obtained with it. The main disadvantage is related to prolonged time to obtain the result, an additional week of incubation. Using the test drilling of the hair is a more expensive and laborious method, but also because non-specific and species can cause organ geofilă perforatorii M. gypseum. Production of organ hair perforation shows that these strains are pathogenic disgonic or, as shown in fig.10, the ability to perforate hair.

Conclusions

M.canis species is frequently involved in the pathology of cutaneous fungal strain identification but it also may encounter some problems. Using autoclaved rice is recommended when on primary isolation media not contains organs of fructification. It is not expensive and easily attainable in any mycology laboratory with modest equipment and its has satisfactory results recommend us to a common practice in medical mycology laboratory differential diagnosis of the genus *Microsporum*.

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