1985-2018 yeast-like filamentous fungi: molecular identification and in vitro susceptibility study

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Objectives

The taxonomy of yeast-like filamentous fungi of the genus *Geotrichum* has undergone several revisions over the years (*Blastoschizomyces*, *Dipodascus* etc.). According to the “one fungus, one name” principle, the name *Saprochaete clavata* (anamorphous of *G. clavatum*) has been changed to *Magnusiomyces clavatus* different from the species *M. capitatus* (*G. capitatum*) [1]. Recent reports in a region of northern Italy (Lombardia) of an increasing number of fungemia caused by *Geotrichum/Magnusiomyces* induced us to analyze clinical strains isolated from 1985 to 2018 with the aim of a correct identification by molecular biology and to test the susceptibility in vitro to antifungals.

Methods

For the molecular identification, the ITS region (ITS1-5.8S-ITS2) [2] and the Rpb2 gene were amplified and sequenced [1]. The sequences obtained were compared with those present in the CBS database. In addition, physiological tests for the utilization of cellobiose and salicin were performed to distinguish the isolates. In vitro susceptibility to 10 antifungals was tested by broth microdilution (EUCAST, CLSI and YeastOne method). Etest was also used to test the isavuconazole activity.

Results

Among the 48 strains, each isolated from a single patient, 28 were identified as *M. capitatus*, 18 as *M. clavatus* and 2 as *G. candidum*. Among the 24 isolates from blood culture, a prevalence of *M. clavatus* (18 strains, isolated mainly from haematological patients and in a period after 2003) compared to *M. capitatus* (6 strains, mainly isolated in the 1985-2003) was observed. *M. capitatus* was prevalent (22/28) among isolates from other biological materials, mainly respiratory secretions (Figure 1).

In vitro susceptibility confirmed the intrinsic resistance of these species to echinocandins. They showed a good susceptibility to itraconazole, posaconazole and voriconazole. About isavuconazole and flucytosine, different patterns of susceptibility between the two species have been highlighted: *M. clavatus* had lower MIC of flucytosine and higher MIC of isavuconazole than *M. capitatus* (in the table are reported the EUCAST data)

	NUMBER OF STRAINS WITH MIC (mg/L)													
	0,03	0,06	0,12	0,25	0,5	1	2	4	8	16	32	64	128	
ITRACONAZOLE		8	6	5	7	1	1				-	-	-	<i>M. capitatus</i>
		3	3	7	4	1					-	-	-	<i>M. clavatus</i>
VORICONAZOLE	5	3	10	5	3	1					-	-	-	
	1	3	3	6	4	1					-	-	-	
POSACONAZOLE	1	2	5	4	4	7	4				-	-	-	
			2	1	1	8	6				-	-	-	
ISAVUCONAZOLE	3	2	8	5	6	1	1	1			-	-	-	
				2	3		6	5	2		-	-	-	
FLUCONAZOLE	-	-	-			1	3	11	9	1	3			
	-	-	-					3	9	4	2			
FLUCYTOSINE	-	-	-	13		2		1	4	6	2			
	-	-	-	13	4	1								
AMFOTERICIN B		1	10	8	6	2					-	-	-	
			1	2	9	6					-	-	-	

Conclusion

With an increasing frequency, *Magnusiomyces/Geotrichum* infections are reported particularly in Southern Europe. However it's difficult to compare the etiology because of the difficulties in the identification. The analysis of our survey shows a different distribution of the two species (*M. clavatus* and *M. capitatus*) in the different biological materials and a considerable increase, in the recent years, of the *M. clavatus* in blood cultures, particularly in hematological patients. The intrinsic resistance of these species to echinocandins, antifungals of increasing use, could be the reason for the emergence of these rare infections.