Mould Identification in a Clinical Microbiology Laboratory using the Bruker Microflex Maldi-Tof MS with the Filamentous Fungi Library 1.0

S. Nys[1], L. Waumans[1], R. Cartuyvels[1], K. Vandebroek[2], E. Oris[2] and K. Magerman[1]
1. Jessa Hospital, Hasselt, Laboratory for Clinical Microbiology, Hasselt, Belgium
2. ZOL, Genk, Laboratory for Clinical Microbiology, Genk, Belgium

Introduction and objectives

- Identification of moulds is mostly based on microscopy or 18S sequencing.
- Both techniques are time consuming and strongly rely on well-qualified technicians.
- In this study, the use of Maldi-Tof MS and the Bruker Filamentous Fungi Library 1.0 for the identification of moulds isolated from clinical samples was evaluated.

Methods

- 65 reference strains (dermatophytes and others, mainly human pathogens) for the verification of the Filamentous Fungi Library 1.0.
- 2-day subculture in liquid sabouraud broth at room temperature on a rotor followed by two wash steps and ethanol-formic acid extraction.


18-48h cultivation


10 minutes precipitation


1 mL, pellet extraction


pellet resuspended


1µL

- a reduced cut-off value of >1.700 for correct species identification was used.

- 114 clinical isolates were prospectively included.
- cut-off value of >1.700 for correct species identification.
- 2-day liquid broth extraction protocol with Maldi-Tof compared to routine 7-14 day subculture followed by microscopy.

The Bruker Maldi-Tof MS combined with the Filamentous Fungi Library 1.0:

- is promising for the identification of moulds isolated in the clinical microbiology laboratory. However, the database needs to be expanded.
- can be used with a reduced cut-off value with minor impact on the rate of correct identifications.
- is much faster compared to identification using microscopy.

Vzw Jessa Hospital
Clinical Laboratory for Microbiology
Stadsomvaart 11,
3500 Hasselt, Belgium
+32 11 30 94 73
sita.nys@jessazh.be

Results

Reference strains

- Of the 65 reference strains 19 species are not included in the Bruker database (Table 1).
- Strains <u>included in the database:</u> 46 strains represent 36 different species.
  34 / 74% were identified correctly to genus level.
  31 / 67% were correctly identified to species level.
  0 were mis-identified on the genus level.
- Strains <u>not in the database:</u> 3 isolates showed a score value >1,700.
  3 isolates were correctly identified to the genus level.
  3 isolates were mis-identified on the species level.

Table 1: Refrence strains not included in the database

F. pedrosoi	
M. langeroni	*Hormographiella sp*
T. schoenleinii	*Auxarthron sp*
T. raubitschekii	*Sordaria sp*
T. verrucosum	*N. mangiferae*
T. soudanense	*H. werneckii*
E. jeanselmi	*Exophalia sp*
Mortierella sp	*S. hyalinum*
C. bertholletiae	*P. verrucosa*
T.viridae	*T. ajelloi*

Clinical strains

- 55 *Aspergillus* species, 28 *Penicillium* species and 30 other moulds (Table 2) were included (n = 113).
- 9 strains (8%) had a score value < 1,700
- Mis-identification, compared to microscopy, occurred on genus level in 1 (1%) and on species level in 5 (5%) of the strains.
- Results are presented in Table 3.

Table 2: Clinical strains other *than Aspergillus sp* and *Penicillium sp*

Strain	# isolates
T. rubrum	11
T. mentagrophytes	6
T. intedigitele	1
F. solani	2
Geosmithia sp	1
Chrysosporium sp	2
S. brevicaulis	1
Curvularia sp	1
R. microspurus	1
L. corymbifera	1
A. alternata	1
Scedosporium sp	1
Chaetomium sp	1

Table 3: Results for the clinical strains

	Microscopy		Maldi-Tof >1,700			
	Genus	Species	Genus	Microscopy confirmed species	Additional species	Wrong species
Asp. species	52	44 (84%)	52 (100%)	42 (81%)	8 (15%)	2 (4%)
Pen. species	24	0	24 (100%)	0	24 (100%)	0
Other moulds	27	17 (63%)	26 (96%)	13 (48%)	10 (37%)	3 (11%)
Total	104	62 (60%)	103 (99%)	56 (54%)	42 (40%)	5 (5%)