EVALUATION OF MATRIX-ASSISTED LASER DESORPTION IONIZATION-TIME OF FLIGHT MASS SPECTROMETRY (MALDI-TOF MS) FOR ACCURATE AND FAST IDENTIFICATION OF YEASTS ISOLATED FROM BLOODSTREAM INFECTIONS

N. Karabicak[1], O. Karatuna[2], I. Akyar[2]

[1]Mycology Reference Laboratory, Public Health Institution of Turkey, Ankara, Turkey ; [2]Department of Medical Microbiology and Acibadem Labmed Clinical Laboratories, Acibadem University School of Medicine, Istanbul, Turkey

Objectives: Fast and accurate identification of yeast isolates from invasive infections at the species level is clinically important because of the varying intrinsic antimicrobial resistance profiles among different yeast species and the newly introduced species-specific clinical breakpoints. We evaluated the performance of matrix-assisted laser desorption ionization-time of flight mass spectrometry (MALDI-TOF MS) for the identification of yeast isolates recovered from bloodstream infections in comparison to phenotypic methods. Methods: Yeast isolates (n = 443, 16 species in 4 genera) obtained from blood cultures of invasive candidiasis patients from various intensive care units in Turkey between 2010-2012 and eight reference strains (8 species in 3 genera: *Candida dubliniensis* ATCC MYA-583, *Candida krusei* ATCC 6258, *Candida parapsilosis* ATCC 22019, *Candida orthopsilosis* ATCC 96139, *Candida metapsilosis* ATCC 96144, *Candida albicans* ATCC 10231, *Cryptococcus neoformans* ATCC 32268 and *Trichosporon asahii* ATCC 90039) were included in the study. Organisms were identified to the species level by germtube test, API ID 32 C (bioMérieux, France) test strips and morphologic examination on corn meal agar with Tween 80 at the Public Health Institution of Turkey, Mycology Reference Laboratory. MALDI-TOF MS analyses were performed using the Bruker Autoflex III instrument, Biotyper software v. 3.0 and database v. 3.1.2 (Bruker Daltonik, Germany) according to manufacturer's instructions at Acibadem Labmed Clinical Laboratories. Yeast isolates were processed by formic acid extraction procedure and spotted in duplicate onto a reusable 96-well polished steel target plate. The confidence of MALDI-TOF MS identification results were assessed with the scores generated by the software (>=2.000 confident species identification, 1.700–1.999 confident genus identification, <1.700 no reliable identification). Discrepancies between the morphological identification and MALDI-TOF MS were resolved by internal transcribed spacer (ITS) 1 and 2, and for rare yeasts, D1/D2 sequencing. Results: All reference strains (n = 8) were accurately identified at the species level by MALDI-TOF MS. For clinical isolates (n = 443), accurate identification rates at the species and genus levels were 90.3% (n = 400) and 97.7% (n = 433), respectively. Although the scores obtained for ten clinical isolates were lower than the confidence limit (scores <1.700), the identifications were in concordance with morphological identification results. In addition, MALDI-TOF MS presented some advantages over phenotypic methods by accurately identifying uncommon *Candida* species, differentiating *C. parapsilosis* from *C. metapsilosis* and *C. orthopsilosis*, distinguishing between *C. albicans* and *C. dubliniensis*, and identifying a *Magnusiomyces capitatus* isolate which also was confirmed with DNA-sequencing (Table 1). Conclusion: MALDI-TOF MS enabled fast and accurate identification of yeast bloodstream isolates with low consumable costs. The addition of new spectra to expand the database for the identification of rare and phenotypically similar species and optimization in identification confidence scores are required to further improve its utility.

Table 1. Performance of MALDI-TOF MS in identification of reference and clinical yeast isolates

Yeast species (final identification)	No. tested	Confidence level of identification		
		Not reliable Score <1.7	Genus level Score ≥1.7 - <2.0	Species level Score ≥2.0
Reference strains	**8**			
Candida albicans ATCC 10231	1			1
Candida dubliniensis ATCC MYA 583	1			1
Candida krusei ATCC 6258	1			1
Candida parapsilosis ATCC 22019	1			1
Candida orthopsilosis ATCC 96139	1			1
Candida metapsilosis ATCC 96144	1			1
Cryptococcus neoformans ATCC 32268	1			1
Trichosporon asahii ATCC 90039	1			1
Clinical isolates	**443**			
Candida albicans	144	1	7	136
Candida parapsilosis	98	3	3	92
Candida tropicalis	58	1	5	52
Candida glabrata	53		8	45
Candida krusei	28			28
Candida kefyr	19		4	15
Candida lusitaniae	16	3	0	13
Candida pelliculosa	12		4	8
Candida guilliermondii	4	1	0	3
Candida orthopsilosis	1			1
Candida metapsilosis	1		1	
Candida dubliniensis	1			1
Trichosporon asahii	4	1		3
Trichosporon mucoides	2			2
Cryptococcus neoformans	1		1	
Magnusiomyces capitatus	1			1
Total	**451**	**10**	**33**	**408**