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Rapid, Accurate Identification of *Candida auris* by Using a Novel Matrix-Assisted Laser Desorption Ionization–Time of Flight Mass Spectrometry (MALDI-TOF MS) Database (Library)

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The newly emerging multidrug-resistant yeast *Candida auris* can cause serious infections and may be underrepresented, as it can be misidentified as other species (e.g., *Candida haemulonii*, *Candida duobushaemulonii*, or *Saccharomyces cerevisiae*) by some biochemical-based testing systems (1–4). *Candida auris* can be identified using research use only (RUO) libraries on matrix-assisted laser desorption ionization–time of flight (MALDI-TOF) platforms, such as the Biotyper platform (Bruker, Billerica, MA), but may need labor-intensive full-tube extraction procedures (3–5). Our laboratory uses the Biotyper equipped with both FDA-approved and RUO libraries. The RUO library contains three *C. auris* entries, but the identification of *C. auris* remains a challenge, and some isolates are never identified by the system. To improve the identification, a novel database, "CMdb," was developed and evaluated on our two Biotyper systems.

The CMdb was created using internationally collected yeasts from the CDC (6) and one in-house clinical *C. auris* isolate. Bruker's protocol was used for database creation, and the direct on-plate extraction method was used for target preparation (7). The CMdb was evaluated on 23 clinical *C. auris* isolates, 20 CDC strains, 52 isolates of 10 other yeast species, and 28 isolates of 16 bacterial species.

TABLE 1 Identification of *Candida auris* and its closely related yeasts using the novel CMdb database on a Bruker Biotyper MALDI-TOF mass spectrometer[a]

Organism (no. of isolates)	CMdb alone		RUO library		EPdbs	
	No. of IDs (%)	Avg score	No. of IDs (%)	Avg score	No. of IDs (%)	Avg score
Candida auris (33[b])	33 (100)	2.50	13 (39%)	1.76	33 (100)	2.51
Candida haemulonii (4)	4 (100)	2.43	4 (100)	1.99	4 (100)	2.38
Candida duobushaemulonii (5)	5 (100)	2.52	4 (80)	2.12	5 (100)	2.52
Candida krusei (11)	11 (100)	2.42	7 (64)	1.85	11 (100)	2.40
Kodameae ohmeri (1)	1 (100)	2.40	1 (100)	1.75	1 (100)	2.40
Saccharomyces cerevisiae (5)	0 (0)	1.32	4 (80)	2.03	5 (100)	2.00
Candida parapsilosis (5)	0 (0)	1.05	5 (100)	2.23	5 (100)	2.15
Candida lusitaniae (3)	0 (0)	1.26	3 (100)	1.87	3 (100)	2.04
Candida guilliermondii (6)	0 (0)	1.15	6 (100)	2.35	6 (100)	2.33
Candida famata (2)	0 (0)	1.27	2 (100)	1.96	2 (100)	1.98
Candida catenulata (3)	0 (0)	1.34	3 (100)	2.02	3 (100)	2.04
Candida kefyr (2)	0 (0)	1.27	1 (50)	1.93	2 (100)	2.09

[a]EPdbs, a combination of the RUO library and the CMdb. IDs, identifications.

[b]Includes 23 clinical isolates (11 submitted from New York, 4 from New Jersey, 6 from Connecticut, and 2 from Florida). The sources were as follows: blood (n = 9), tissue (n = 3), body fluids (n = 2), urine (n = 1), and unknown (n = 8).

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TABLE 2 Antifungal resistance results of the clinical *C. auris* yeasts[a]

Antifungal agent	No. of susceptible isolates	No. of S-DD isolates	No. of isolates that were resistant or NS (no. of patients)
Anidulafungin	21	0	2 (1)
Micafungin	21	0	2 (1)
Caspofungin	21	0	2 (1)
5-Flucytosine	23	0	0
Posaconazole	23	0	0
Voriconazole	19	4	0
Itraconazole	5	18	0
Fluconazole	0	0	23 (15)
Amphotericin B	5	0	18 (11)

[a]Twenty-three isolates were tested. The assays were conducted by using the Sensititre YeastOne panel according to the manufacturer's instructions (Trek Diagnostic System, Independence, OH). S-DD, susceptible-dose dependent; NS, nonsusceptible.

The new CMdb database contains 22 mean spectrum projections from *C. auris* and 4 other related species (*C. haemulonii*, *C. duobushaemulonii*, *Candida krusei*, and *Kodameae ohmeri*). When we used the CMdb, all 23 clinical *C. auris* isolates plus the 10 CDC strains were correctly identified (100%) (Table 1); of these isolates, 22 had their identification confirmed by sequencing of the internal transcribed spacer region of their ribosomal DNA (rDNA) (one isolate was lost to contamination, and 22 additional isolates were identified during preparation of the manuscript and are not listed here). The identification log score was consistently greater than 2.40, with an average of 2.50. In comparison, 13 (39%) *C. auris* isolates were identified by the RUO database, with an average log score of 1.76 ($P < 0.001$ [R-project.org]), and the rest had log scores below the identification level (1.7). The 4 closely related non-*C. auris* species were correctly identified by using the same CMdb database. No misidentification was observed with the CMdb when tests were run on other yeasts and bacteria.

When the spectrum-producing targets were counted from *C. auris* and the 4 other species, all spectra were correctly identified with isolates in the CMdb (100%), while 32% of the spectra were identified with isolates in the RUO library. The low identification rate from the RUO library might have been caused by losing some proteins during its full-tube extraction database creation. *Candida auris* was well identified from 3 different culture media (Sabouraud dextrose agar [SDA], Columbia nalidixic acid agar, and Trypticase soy agar with sheep blood) but had the highest log scores from isolates grown on SDA. *Candida auris* was reliably identified on SDA at 20 to 30°C for 3 days.

All 23 clinical isolates (100%) were highly resistant to fluconazole (>256 μg/ml), and two isolates expressed multidrug resistance (Table 2).

The identification of *C. auris* with this novel CMdb was accurate (100%), quick, and easy, with significantly higher log scores, which will benefit patient care and public health interventions.

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There are no conflicts of interest to report.

REFERENCES

1. Centers for Disease Control and Prevention. 21 August 2017. *Candida auris*. Centers for Disease Control and Prevention, Atlanta, GA. https://www.cdc.gov/fungal/diseases/candidiasis/candida-auris.html.
2. Kordalewska M, Zhao Y, Lockhart SR, Chowdhary A, Berrio I, Perlina DS. 2017. Rapid and accurate molecular identification of the emerging multidrug-resistant pathogen *Candida auris*. J Clin Microbiol 55:2445–2452. https://doi.org/10.1128/JCM.00630-17.
3. Mizusawa M, Miller H, Green R, Lee R, Durante M, Perkins R, Hewitt C, Simner PJ, Carroll KC, Hayden RT, Zhang SX. 2017. Can multidrug-resistant *Candida auris* be reliably identified in clinical microbiology laboratories? J Clin Microbiol 55:638–640. https://doi.org/10.1128/JCM.02202-16.
4. Kathuria S, Singh PK, Sharma C, Prakash A, Masih A, Kumar A, Meis JF, Chowdhary A. 2015. Multidrug-resistant *Candida auris* misidentified as *Candida haemulonii*: characterization by matrix-assisted laser desorption

ionization–time of flight mass spectrometry and DNA sequencing and its antifungal susceptibility profile variability by Vitek 2, CLSI broth microdilution, and Etest method. J Clin Microbiol 53:1823–1830. https://doi.org/10.1128/JCM.00367-15.
5. Girard V, Mailler S, Chetry M, Vidal C, Durand G, van Belkum A, Colombo AL, Hagen F, Meis JF, Chowdhary A. 2016. Identification and typing of the emerging pathogen *Candida auris* by matrix-assisted laser desorption ionisation time of flight mass spectrometry. Mycoses 59:535–538. https://doi.org/10.1111/myc.12519.
6. CDC. 2017. Isolates currently available. *Candida auris* panel. CDC, Atlanta, GA. https://www.cdc.gov/drugresistance/resistance-bank/currently-available.html.
7. Bruker Daltonics. 2015. MALDI Biotyper CA system user manual. Bruker Daltonics, Bremen, Germany.