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Multicenter Study Evaluating the Vitek MS System for Identification of Medically Important Yeasts

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The optimal management of fungal infections is correlated with timely organism identification. Matrix-assisted laser desorption ionization–time of flight (MALDI-TOF) mass spectrometry (MS) is revolutionizing the identification of yeasts isolated from clinical specimens. We present a multicenter study assessing the performance of the Vitek MS system (bioMérieux) in identifying medically important yeasts. A collection of 852 isolates was tested, including 20 *Candida* species (626 isolates, including 58 *C. albicans*, 62 *C. glabrata*, and 53 *C. krusei* isolates), 35 *Cryptococcus neoformans* isolates, and 191 other clinically relevant yeast isolates; in total, 31 different species were evaluated. Isolates were directly applied to a target plate, followed by a formic acid overlay. Mass spectra were acquired using the Vitek MS system and were analyzed using the Vitek MS v2.0 database. The gold standard for identification was sequence analysis of the D2 region of the 26S rRNA gene. In total, 823 isolates (96.6%) were identified to the genus level and 819 isolates (96.1%) were identified to the species level. Twenty-four isolates (2.8%) were not identified, and five isolates (0.6%) were misidentified. Misidentified isolates included one isolate of *C. albicans* ($n = 58$) identified as *Candida dubliniensis*, one isolate of *Candida parapsilosis* ($n = 73$) identified as *Candida pelliculosa*, and three isolates of *Geotrichum klebahnii* ($n = 6$) identified as *Geotrichum candidum*. The identification of clinically relevant yeasts using MS is superior to the phenotypic identification systems currently employed in clinical microbiology laboratories.

As the number of patients with profound immunosuppression (such as those with solid-organ and hematopoietic stem cell transplants) continues to rise, the morbidity and mortality burdens attributed to invasive fungal infections are increasing (1–6). In the case of invasive fungal infections, expedient identification of the offending organism is essential for optimal patient management and the best clinical outcomes. As the antifungal susceptibility profiles for many fungi (both yeasts and molds) are predictable, organism identification frequently is sufficient to expedite appropriate empirical antifungal therapy. This has been demonstrated both to reduce the overall length of hospitalization and to maximize favorable clinical outcomes (7–10). Conversely, the rapid exclusion of overt pathogenic or intrinsically resistant species can be used to narrow therapy and/or to prevent treatment with potentially toxic antifungal agents, thereby reducing negative clinical outcomes and costs.

The methods for identification of yeasts in the diagnostic clinical microbiology laboratory have improved significantly over the past several decades (11, 12), with methods ranging from simple manual biochemical assays to automated biochemical methods to sophisticated nucleic acid-based assays (11, 12). While these advancements in methodology have greatly enhanced our ability to identify yeasts, the limitations of these methods include cost, turnaround time, and, in some instances, the need for considerable expertise. Additionally, the accuracy of identification for some less-common species is not optimal for some of the methods (13–17).

A technology that is poised to revolutionize the rapid identification of yeasts isolated in the clinical microbiology laboratory is

matrix-assisted laser desorption ionization–time of flight (MALDI-TOF) mass spectrometry (MS). MALDI-TOF MS-based microbial identification relies on the generation of an organism-specific mass spectrum or “protein fingerprint” that is examined against a reference database to provide an organism identification (18). The objective of this multicenter study was to assess the performance of the Vitek MS MALDI-TOF mass spectrometer (bioMérieux) in conjunction with the Vitek MS v2.0 database for the identification of yeasts isolated in diagnostic clinical microbiology laboratories.

(This work was presented in part as an abstract at the 113th General Meeting of the American Society for Microbiology, Denver, CO, 18 to 21 May 2013.)

MATERIALS AND METHODS

Isolates used in this study. Yeasts isolated and identified from clinical specimens obtained from five diagnostic clinical microbiology laboratories, located at geographically distinct sites in North America, were included in the study. The study sites were Barnes-Jewish Hospital (St. Louis, MO), the Cleveland Clinic (Cleveland, OH), the UCLA Health System (Los Angeles, CA), the North Shore LIJ Core Laboratory (Lake

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TABLE 1 Performance characteristics of the Vitek MS system in identifying clinically relevant *Candida* species

Organism	No. (%) of isolates				
	Total	Identified correctly to genus	Identified correctly to species	Unidentified	Misidentified
<i>Candida albicans</i>	58	57 (98.3)	57 (98.3)	0 (0)	1 (1.7) ^a
<i>Candida dubliniensis</i>	34	34 (100)	34 (100)	0 (0)	0 (0)
<i>Candida famata</i>	29	29 (100)	28 (96.6)	0 (0)	0 (0)
<i>Candida glabrata</i>	62	62 (100)	62 (100)	0 (0)	0 (0)
<i>Candida guilliermondii</i>	36	35 (97.2)	35 (97.2)	1 (2.8)	0 (0)
<i>Candida haemulonii</i>	12	12 (100)	12 (100)	0 (0)	0 (0)
<i>Candida inconspicua</i>	23	23 (100)	23 (100)	0 (0)	0 (0)
<i>Candida intermedia</i>	7	7 (100)	7 (100)	0 (0)	0 (0)
<i>Candida kefyr</i>	30	30 (100)	30 (100)	0 (0)	0 (0)
<i>Candida krusei</i>	53	53 (100)	53 (100)	0 (0)	0 (0)
<i>Candida lambica</i>	9	9 (100)	9 (100)	0 (0)	0 (0)
<i>Candida lipolytica</i>	28	28 (100)	28 (100)	0 (0)	0 (0)
<i>Candida lusitanae</i>	33	30 (90.9)	29 (87.9)	3 (9.1)	0 (0)
<i>Candida norvegensis</i>	30	29 (96.7)	29 (96.7)	1 (3.3)	0 (0)
<i>Candida parapsilosis</i>	73	72 (98.6)	72 (98.6)	0 (0)	1 (1.4) ^b
<i>Candida pelliculosa</i>	33	33 (100)	33 (100)	0 (0)	0 (0)
<i>Candida rugosa</i>	6	6 (100)	6 (100)	0 (0)	0 (0)
<i>Candida tropicalis</i>	54	51 (94.4)	49 (90.7)	3 (5.6)	0 (0)
<i>Candida utilis</i>	8	8 (100)	8 (100)	0 (0)	0 (0)
<i>Candida zeylanoides</i>	8	8 (100)	8 (100)	0 (0)	0 (0)
Total	626	616 (98.4)	612 (97.8)	8 (1.3)	2 (0.3)

^a Isolate misidentified as *C. dubliniensis*.^b Isolate misidentified as *C. pelliculosa*.

Success, NY), and the Massachusetts General Hospital (Boston, MA). In total, the collection tested was composed of 852 yeast isolates obtained from the five trial sites (508 isolates) and the bioMérieux stock collection (344 isolates). The collection included 20 *Candida* species (Table 1), *Cryptococcus neoformans*, and 10 species in the genera *Geotrichum*, *Kodamaea*, *Malassezia*, *Rhodotorula*, *Saccharomyces*, and *Trichosporon* (Table 2).

Of the 344 isolates from the bioMérieux stock collection, 96 were used in the development of the database. These isolates represent rare taxa, such that it would not have been possible to evaluate them exclusively via prospective collection.

Cultivation of yeast isolates. The isolates were obtained from frozen stocks or were tested fresh from clinical cultures. Strains that were stored frozen were subcultured on Sabouraud dextrose agar (SDA; Remel, Le-

nexa, KS) twice before mass spectrometric analysis. Freshly collected isolates were subcultured on SDA to assess purity before testing, or, if a pure culture was observed on the primary SDA plate, it was tested directly. All isolates were analyzed within 72 h after visible growth at 35°C. In only four instances, isolates were taken from media other than SDA, including one isolate taken from CHROMagar *Candida* (Becton, Dickinson, Sparks, MD), one isolate taken from Mueller-Hinton II agar (Becton, Dickinson), and two isolates taken from tryptic soy agar with sheep's blood (Remel). In the four instances where SDA was not used to cultivate the strain for MS analysis, the MS identification matched the reference identification method.

Sample preparation. The yeast isolates were prepared for mass spectrometric analysis using a direct, on-target, extraction method (19).

TABLE 2 Performance characteristics of the Vitek MS system in identifying clinically relevant non-*Candida* yeast species

Organism	No. (%) of isolates				
	Total	Identified correctly to genus	Identified correctly to species	Unidentified	Misidentified
<i>Cryptococcus neoformans</i>	35	35 (100)	35 (100)	0 (0)	0 (0)
<i>Geotrichum capitatum</i>	32	30 (93.8)	30 (93.8)	2 (6.3)	0 (0)
<i>Geotrichum klebahnii</i>	6	0 (0)	0 (0)	3 (50)	3 (50) ^a
<i>Kodamaea ohmeri</i>	11	10 (90.9)	10 (90.9)	1 (9.1)	0 (0)
<i>Malassezia furfur</i>	7	6 (85.7)	6 (85.7)	1 (14.3)	0 (0)
<i>Malassezia pachydermatis</i>	8	3 (37.5)	3 (37.5)	5 (62.5)	0 (0)
<i>Rhodotorula mucilaginosa</i>	35	35 (100)	35 (100)	0 (0)	0 (0)
<i>Saccharomyces cerevisiae</i>	42	41 (97.6)	41 (97.6)	1 (2.4)	0 (0)
<i>Trichosporon asahii</i>	32	30 (93.8)	30 (93.8)	2 (6.3)	0 (0)
<i>Trichosporon inkin</i>	9	9 (100)	9 (100)	0 (0)	0 (0)
<i>Trichosporon mucoides</i>	9	8 (88.9)	8 (88.9)	1 (11.1)	0 (0)
Total	226	207 (91.6)	207 (91.6)	16 (7.1)	3 (1.3)

^a Isolates were misidentified as *G. candidum*.

Briefly, a portion of a single colony was applied directly to a disposable target slide (product no. 410893; bioMérieux, Marcy l'Etoile, France) composed of a polypropylene carrier with a stainless steel layer, using a 1- μ l loop (product no. 861567010; Sarstedt, Newton, NC), and was lysed by direct application of 0.5 μ l formic acid (25% [vol/vol], product no. 411072; bioMérieux) to the isolate immediately after application on the target plate. Immediately after the formic acid overlay was allowed to dry at room temperature, 1 μ l of matrix solution (3.1% [wt/vol] α -cyano-4-hydroxycinnamic acid, product no. 411071; bioMérieux) was applied and allowed to dry at room temperature prior to mass spectrometric analysis. Isolates were prepared for mass spectrometric analysis at the Vitek MS preparation station, and the isolate information was transferred to the Vitek MS acquisition station using Myla v2.4 middleware. The total sample preparation time was approximately 1 min per isolate.

MALDI-TOF MS. Following sample preparation, samples were analyzed with the Vitek MS MALDI-TOF mass spectrometer in linear positive-ion mode, across the mass-to-charge ratio range of 2,000 to 20,000 Da. Each spot was irradiated with 500 laser shots at 50 Hz. Target plates were calibrated and quality controlled both before and after data acquisition by using *Escherichia coli* ATCC 8739. Additionally, a *Candida glabrata* isolate (*C. glabrata* ATCC MYA-2950) and a sample containing matrix only (negative control) were assayed for quality control purposes. After the acquisition of spectra, data were transferred from the Vitek MS acquisition station to the Vitek MS analysis server, and identification results were displayed using Myla v2.4 middleware. The total processing and data analysis time was approximately 20 min for a single isolate; this time increased by approximately 1 min for each subsequent sample. Each operator participating in the study was required to analyze a proficiency panel successfully prior to beginning to test isolates for this investigation.

Data analysis. The Vitek MS identification system is based on comparison of the characteristics of the spectra obtained with the Vitek MS v2.0 database. This database was built using spectra for known strains for each claimed species. Based on this representative data collection, a weight is assigned to each peak for each species according to its specificity. As part of the identification process, the software compares the spectrum obtained with peak weights defined for each claimed species. The resulting quantitative value, the confidence value, is calculated and expresses the similarity between the unknown organism and every organism or organism group in the database. A single identification is displayed, with a confidence value from 60.0 to 99.9, when one significant organism or organism group is retained. "Low-discrimination" identifications are displayed when more than one but not more than four significant organisms or organism groups are retained. In this case, the sum of confidence values is equal to 100. When more than four organisms or organism groups are found, or when no match is found, the organism is considered unidentified.

Molecular identification of yeast isolates. The molecular identification of all isolates in the test collection was performed by MIDI Labs (Newark, DE). The isolates were identified by sequencing the D2 region of the 26S rRNA gene (12) using the MicroSeq D2 LSU rDNA fungal identification kit (Applied Biosystems, Foster City, CA) (20). Briefly, yeast genomic DNA was extracted and the D2 region was amplified by PCR; the resultant PCR product was sequenced and compared with fungal sequences in the MicroSeq D2 fungal library and other public databases, including GenBank (<http://www.ncbi.nlm.nih.gov/GenBank>).

RESULTS

Overall performance of the Vitek MS system. A collection of 852 yeast isolates, comprising 31 different species obtained primarily from clinical microbiology laboratories located in five different geographical regions in North America, was used to challenge the Vitek MS v2.0 database (bioMérieux). Of the 852 isolates included in the collection, 823 (96.6%) were identified to the genus level, while 819 (96.1%) were identified to the species level. In total, 24

isolates (2.8%) were not identified and five isolates (0.6%) were misidentified.

Performance of the Vitek MS system in identifying *Candida* species. A total of 626 *Candida* isolates representing 20 different species, including 58 *Candida albicans*, 62 *C. glabrata*, and 53 *Candida krusei* isolates, were analyzed (Table 1). Of the 626 isolates, 616 (98.4%) were identified to the genus level and 612 (97.8%) were identified to the species level. Only eight isolates (1.3%) were unidentified and two isolates (0.3%) were misidentified. The isolates that were misidentified included one isolate of *C. albicans* that was misidentified as *Candida dubliniensis* and one isolate of *Candida parapsilosis* that was misidentified as *Candida pelliculosa*.

When the isolates from the bioMérieux stock collection were excluded, 16 species of *Candida* were represented. Of these 404 isolates, 396 (98.0%) were identified correctly to the genus level and 393 (97.3%) to the species level (Table 3).

Performance of the Vitek MS system in identifying non-*Candida* yeast isolates. A total of 226 isolates representing 11 different species, including 35 *C. neoformans* isolates, 50 *Trichosporon* isolates, and 35 *Rhodotorula mucilaginosa* isolates, were analyzed (Table 2). The number of isolates identified to both the genus and species levels was 207 (91.6%), with all 35 (100%) *C. neoformans* isolates correctly identified to the species level. The number of isolates that were misidentified (three isolates [1.3%]) was low. The three misidentified isolates were *Geotrichum klebahnii* isolates that were identified as *Geotrichum candidum*. The proportion of isolates that were not identified in this group (16 isolates [7.1%]) was greater than the proportion of isolates that were not identified in the *Candida* species group.

When the isolates from the bioMérieux stock collection were excluded from this group of organisms, nine species of non-*Candida* yeast isolates remained. Of the 104 isolates, 99 (95.2%) were correctly identified to both the genus and species levels (Table 4).

Quality control. The *C. glabrata* quality control organism and the negative control sample (matrix only) were tested by the Vitek MS every day that yeast isolates were assayed and with every new lot of target slides, formic acid, and matrix. During the trial, the quality control organism was tested 141 times and acceptable results were obtained 139 times (98.6%). Two quality control tests yielded no identification upon initial testing. In both instances, however, the correct identification was obtained upon repeat testing on the same day. In all instances, the negative control yielded no identification.

DISCUSSION

Although the identification of yeast isolates has greatly improved over the past several decades, the manual and automated biochemical methods commonly used to identify contemporary yeast isolates are time-consuming and may result in low-discrimination identifications that require additional testing (12, 21). Nucleic acid-based identification techniques, such as DNA sequencing of yeast, have high accuracy but are expensive, might have prolonged turnaround times, and require technical expertise and equipment that may not be available to all laboratories. MALDI-TOF MS offers a balance between speed and highly accurate yeast identifications.

While fewer studies evaluating MALDI-TOF MS identification of yeasts than bacteria have been published to date, the theme of the existing literature is that the performance of MALDI-TOF MS in identifying fungi, both yeasts and molds, is comparable or su-

TABLE 3 Performance characteristics of the Vitek MS system in identifying *Candida* species recovered from clinical specimens

Organism	No. (%) of isolates				
	Total	Identified correctly to genus	Identified correctly to species	Unidentified	Misidentified
<i>Candida albicans</i>	58	57 (98.3)	57 (98.3)	0 (0)	1 (1.7) ^a
<i>Candida dubliniensis</i>	24	24 (100)	24 (100)	0 (0)	0 (0)
<i>Candida famata</i>	2	2 (100)	2 (100)	0 (0)	0 (0)
<i>Candida glabrata</i>	62	62 (100)	62 (100)	0 (0)	0 (0)
<i>Candida guilliermondii</i>	25	24 (96)	24 (96)	1 (4)	0 (0)
<i>Candida haemulonii</i>	4	4 (100)	4 (100)	0 (0)	0 (0)
<i>Candida inconspicua</i>	1	1 (100)	1 (100)	0 (0)	0 (0)
<i>Candida kefyr</i>	15	15 (100)	15 (100)	0 (0)	0 (0)
<i>Candida krusei</i>	46	46 (100)	46 (100)	0 (0)	0 (0)
<i>Candida lambica</i>	1	1 (100)	1 (100)	0 (0)	0 (0)
<i>Candida lipolytica</i>	4	4 (100)	4 (100)	0 (0)	0 (0)
<i>Candida lusitanae</i>	30	27 (90)	26 (87)	3 (0.1)	0 (0)
<i>Candida parapsilosis</i>	72	72 (100)	72 (100)	0 (0)	0 (0)
<i>Candida pelliculosa</i>	5	5 (100)	5 (100)	0 (0)	0 (0)
<i>Candida rugosa</i>	2	2 (100)	2 (100)	0 (0)	0 (0)
<i>Candida tropicalis</i>	53	50 (94)	48 (91)	3 (6)	0 (0)
Total	404	396 (98.0)	393 (97.3)	7 (1.7)	1 (0.2)

^a Isolate misidentified as *C. dubliniensis*.

rior to that of conventional and nucleic acid-based identification methods (11, 12, 19, 22–29). The major advantages of MALDI-TOF MS identification of yeasts, compared with conventional methods, are the marked decreases in cost and time to identification (30). Antifungal susceptibility profiles generally are predictable from the species identification (8) and, of note, the four species of yeast that account for the vast majority of infections, i.e., *C. albicans*, *C. glabrata*, *C. krusei*, and *C. parapsilosis*, have distinct susceptibility profiles (8). Therefore, rapid, highly accurate identification of yeast isolates using MALDI-TOF MS is poised to enhance patient care drastically and to reduce hospital-associated costs due to fungal infections.

In this study, we evaluated the performance characteristics of the Vitek MS with the v2.0 database for identification of medically important yeast species. This study has a number of strengths. The first is that this was a multicenter evaluation; therefore, a large number of independent operators were able to demonstrate the interlaboratory accuracy of this method. Isolates were recovered from geographically distinct areas across North America, enrich-

ing the collection for strain heterogeneity. In addition, this study included a large number of isolates, and the identification of all isolates was verified using sequence analysis as a gold standard. Finally, this is the first study to date to evaluate the performance characteristics of the Vitek MS v2.0 database for identification of clinically relevant yeast species.

The results of the multicenter study indicate that, independent of the laboratory and the geographical origin of the isolates, the Vitek MS demonstrated an overall species identification rate comparable or superior to those for both traditional biochemical and nucleic acid-based yeast identification systems (11, 12) but with a significant reduction in the time to identification. This method is technically facile and, once the laboratory has recovered the capital investment for the instrument purchase, the ongoing cost of consumables is low.

In our study, 24 (2.8%) and 5 (0.6%) isolates were not identified and were misidentified, respectively. Overall, we identified >96% of the 852 isolates in this study to the species level. This is comparable to the findings of other studies evaluating MALDI-

TABLE 4 Performance characteristics of the Vitek MS system in identifying non-*Candida* yeast isolates recovered from clinical specimens

Organism	No. (%) of isolates				
	Total	Identified correctly to genus	Identified correctly to species	Unidentified	Misidentified
<i>Cryptococcus neoformans</i>	29	29 (100)	29 (100)	0 (0)	0 (0)
<i>Geotrichum capitatum</i>	3	3 (100)	3 (100)	0 (0)	0 (0)
<i>Kodamaea ohmeri</i>	1	1 (100)	1 (100)	0 (0)	0 (0)
<i>Malassezia furfur</i>	1	0 (0)	0 (0)	1 (100)	0 (0)
<i>Malassezia pachydermatis</i>	1	0 (0)	0 (0)	1 (100)	0 (0)
<i>Rhodotorula mucilaginosa</i>	26	26 (100)	26 (100)	0 (0)	0 (0)
<i>Saccharomyces cerevisiae</i>	28	27 (96)	27 (96)	1 (4)	0 (0)
<i>Trichosporon asahii</i>	11	9 (82)	9 (82)	2 (18)	0 (0)
<i>Trichosporon mucoides</i>	4	4 (100)	4 (100)	0 (0)	0 (0)
Total	104	99 (95.2)	99 (95.2)	5 (4.8)	0 (0)

TOF MS identification of yeasts using other instrumentation platforms or spectral databases; Yaman and coworkers identified 94% of 265 yeast isolates correctly using the Bruker Biotyper (29), Bader and colleagues identified >95% of 1,192 isolates correctly using both the Bruker Biotyper and the Saramis instruments (26), Dhiman and colleagues identified >96% of 138 “common” yeasts and 84.5% of 103 “uncommon” yeasts to the species level using the Bruker Biotyper (27), and Iriart et al. identified 184 of 188 yeast isolates (97.9%) tested using the Vitek MS (19). In contrast to the current study, the study by Iriart et al. (19) evaluated the Vitek MS v1.0 database and included primarily *Candida* isolates from a medical center in France, and sequencing was not the reference method for the study.

For the isolates that were misidentified in the current study, the incorrect identifications would be unlikely to lead to adverse clinical outcomes. Two of the five incorrectly identified isolates were *Candida* species, including an isolate of *C. albicans* misidentified as *C. dubliniensis* and an isolate of *Candida parapsilosis* misidentified as *C. pelliculosa*. The clinical impact of misidentifying *C. albicans* as *C. dubliniensis* is likely to be minimal, although it has been suggested that the development of fluconazole resistance is more likely for *C. dubliniensis* than for *C. albicans* (31). *C. parapsilosis* exhibits higher MICs for the echinocandins than do most other *Candida* species (8, 32); therefore, misidentification might be clinically significant. However, data on the susceptibility profile of *C. pelliculosa* are sparse, and it is not obvious what empirical therapy might be initiated based on this identification. Although few isolates were not identified in this study, three (9.1%) of the *Candida lusitanae* isolates tested were not identified. This is of minor importance, compared with the overall performance characteristics of this method, but this finding is of note in light of the fact that this species can be resistant to amphotericin B, a trait unusual for *Candida* species (8).

The three other misidentified isolates were *Geotrichum klebahnii* identified as *G. candidum*. *G. klebahnii* is in the current database. While this error is unlikely to be clinically significant, bioMérieux indicated that future database and software updates will result in reporting of these two species as *G. candidum/klebahnii* rather than specific species-level identification, to circumvent this misidentification event (bioMérieux, personal communication).

In contrast to the “direct colony” methods typically used for MALDI-TOF MS identification of bacterial isolates, the majority of studies to date evaluating MALDI-TOF MS methods for identification of yeasts have suggested the use of a more labor-intensive formic acid/organic solvent extraction method. This method involves a series of centrifugation steps and is thought to be necessary for reliable identification of these organisms, because of the thick, chitin-containing cell walls of yeasts (26, 27, 29, 33, 34). These additional steps significantly increase the hands-on time required for analysis and negatively affect turnaround times. For example, using the full extraction method for sample preparation, one study reported an average of 5.1 min of hands-on time and a total turnaround time of 38.4 min per isolate (27). A recent study conducted by Theel and coworkers evaluated a direct on-plate extraction preparation method using 70% formic acid, and 73 of 90 isolates (81.1%) were identified to the species level using this method (35). The performance of the on-plate direct extraction method demonstrated in this study and by Theel et al. (35) represents improvements in both turnaround times and workflow for MALDI-TOF MS identification of yeasts. However, one point of

caution when using a direct plate extraction preparation method is that the early growth of some thermally dimorphic fungi, such as *Histoplasma capsulatum* and *Coccidioides immitis/posadasii*, might resemble yeast-like colonies. Therefore, clinical laboratories should be mindful of growth rates and colony morphology when using this method for yeast identification.

Despite the promising results reported in this study, there are some limitations to our data. All except four of the isolates were grown on SDA for MALDI-TOF MS analysis; therefore, the performance characteristics of this methodology for yeast grown on other types of media are unknown. For the 852 yeast isolates tested in this study, all of the species identified are included in the Vitek MS v2.0 database. It is not known if unusual taxa not represented in the database would be misidentified or simply not identified if tested with this system. Finally, no isolates of *Cryptococcus gattii*, an emerging fungal pathogen (36), were included in the study. Thus, the ability of the Vitek MS to differentiate *C. neoformans* from *C. gattii*, which might be of epidemiological and clinical importance, is not known. Previous studies using other platforms suggest that MALDI-TOF MS methods do have the potential for species resolution of *Cryptococcus* species by permitting the addition of mass spectra to the reference database (28). The Vitek MS IVD system evaluated in this study does not permit user modifications, such as the addition of spectra to the database.

In conclusion, we present the results of a multicenter study evaluating the Vitek MS system for identification of clinically relevant yeasts. Identification of yeasts using the Vitek MS is faster and more accurate than phenotypic identification systems currently employed in clinical microbiology laboratories and affords accuracy comparable to that of more laborious and costly molecular methods. Implementation of this methodology should streamline yeast identification in the laboratory, positively affect patient care, and reduce health care-associated costs.

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