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Comparison of the Quantum II, API Yeast Ident, and AutoMicrobic Systems for Identification of Clinical Yeast Isolates

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The Quantum II Yeast Identification System (Abbott Laboratories) is a microprocessor-based spectrophotometric system for identification of clinical yeast isolates within 24 h. We compared the Quantum II system with the API Yeast Ident (Analytab Products) and the AutoMicrobic System Yeast Biochemical Card (AMS-YBC; Vitek Systems, Inc.) for the identification of 221 clinical yeast isolates, including 120 common clinical isolates (*Candida albicans*, *C. tropicalis*, *C. parapsilosis*, *Torulopsis glabrata*, and *Cryptococcus neoformans*) and 101 relatively uncommon clinical isolates. The API 20C (Analytab) was used as the reference system. The Quantum II and AMS-YBC systems correctly identified 181 (82%) and 184 (83%) isolates, respectively, whereas the Yeast Ident system correctly identified 132 (60%) isolates. Of the 120 common clinical isolates, 113 (94%) were correctly identified by Quantum II, 103 (86%) were correctly identified by AMS-YBC, and 83 (69%) were correctly identified by Yeast Ident. Of the 101 uncommon clinical isolates tested, 68 (67%) were correctly identified by Quantum II, 81 (80%) were correctly identified by AMS-YBC, and 49 (49%) were correctly identified by Yeast Ident. The overall accuracy of the Quantum II, AMS-YBC, and API Yeast Ident was not sufficient to recommend any of these systems for routine use in the clinical microbiology laboratory without substantial expansion of the respective data bases.

Diagnosis and therapy of fungal infection in immunocompromised patients has become increasingly important in the last several years. Because colonization of patients and medical devices with organisms such as *Candida albicans*, *C. tropicalis*, and *C. parapsilosis* frequently precedes the development of invasive disease with the same organisms (8, 10, 22,26, 28), it is important for the clinical laboratory to have the capability to rapidly and accurately identify both common and uncommon yeast isolates.

The inherent slowness of the standard methods for identifying clinical yeast isolates has greatly hindered the clinical usefulness of yeast identification. Conventional methods can require as long as 14 to 28 days for completion of biochemical tests (1, 12, 14, 16). Some of the newer miniaturized systems such as the API 20C (Analytab Products, Plainview, N.Y.), the Minitek Yeast System (BBL Microbiology Systems, Cockeysville, Md.), and the Uni-Yeast-Tek (Flow Laboratories, Inc., McLean, Va.) may provide biochemical testing in 3 to 7 days, but despite this improvement, the results are frequently of retrospective interest only. A system that accurately identifies clinical yeast isolates in 24 h or less is sorely needed to provide the information necessary for optimal care of patients at risk for fungal infections.

One of the more recent advances in the rapid identification of yeast isolates is the use of chromogenic substrates to assess preformed enzyme activity. By detecting preformed enzymes, these systems provide a growth-independent means of identifying clinically important yeasts within 4 h of inoculation (3, 4, 24). The API Yeast Ident system (Analytab) is the first commercially available product to utilize this approach for the identification of clinically important yeasts (24). In the Yeast Ident system the yeast enzymes cleave chromogenic components from specific chromogen-attached substrates. Color changes in test wells are manually read and interpreted and indicate a positive enzyme test.

The application of instrumentation to the various yeast identification systems allows automated or semi-automated test reading and interpretation, thus providing the potential for improved speed, standardization, accuracy, and reproducibility (6, 7, 13–15, 17, 19). Although several large automated or semi-automated systems are available for yeast identification (6, 7, 13, 14, 19), these instruments frequently do not fulfill the needs of smaller laboratories, many of which would benefit from rapid automated test reading and interpretation but do not have the necessary laboratory space or funds. The recent adaptation of the Abbott Yeast Identification System (6) to the Quantum II instrument (Abbott Laboratories, Irving, Tex.) appears to be a possible solution for laboratories which would like to progress beyond the slower manual yeast identification systems but stop short of the larger, more expensive automated systems. The Quantum II Yeast Identification System automatically reads and interprets the results of biochemical tests within 24 h of inoculation by using a photometer provided by the manufacturer. Although studies comparing the Quantum II with rapid manual yeast identification systems are necessary (11, 25, 27), it is equally important to compare it with other identification systems offering automated test reading and interpretation such as the AutoMicrobic System (AMS; Vitek Systems, Inc., Hazelwood, Mo.). In an effort to evaluate the usefulness of the various yeast identification systems in the clinical laboratory, we compared the Quantum II, the API Yeast Ident, and the AMS plus Yeast Biochemical Card and accompanying software (AMS-YBC) with the widely used API 20C for the identification of common and uncommon clinical yeast isolates.

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MATERIALS AND METHODS

Test organisms. A total of 221 clinical yeast isolates, encompassing nine genera and 21 species, were tested in this study. The test organisms included 120 common clinical isolates (*Candida albicans*, *C. tropicalis*, *C. parapsilosis*, *Torulopsis glabrata*, and *Cryptococcus neoformans*) and 101 relatively uncommon clinical isolates. Although *Geotrichum* and *Prototheca* species are not yeast-fungi, they are included in yeast identification systems, and we have elected to place them in the category of uncommon yeast isolates. All test organisms were clinical isolates obtained from patients hospitalized at either the University of Iowa Hospitals and Clinics or the University of Virginia Medical Center. The isolates were stored as suspensions in sterile water or on agar slants and were subcultured at least twice onto Sabouraud dextrose agar (Difco Laboratories, Detroit, Mich.) before being tested. Each isolate was tested in the Quantum II, the API Yeast Ident, the AMS-YBC, and the API 20C yeast identification systems. For this study, the correct identification was defined as that given by the API 20C. Discrepancies between the individual test systems and API 20C were arbitrated by performing conventional assimilation, fermentation, and morphologic testing (1, 12, 14, 16). Quality control for each identification system was performed with stock isolates of *C. albicans*, *C. tropicalis*, and *Cryptococcus neoformans* which had been identified by conventional methods. The quality control strains were correctly identified by all of the test systems.

API 20C. The API 20C system consists of a plastic strip containing dehydrated reagents for the performance of 19 biochemical tests. The tests were performed as specified by the manufacturer. The strips were incubated at 30°C, and results were recorded after 24, 48, and 72 h of incubation. Identifications were made by referring to the API Analytical Profile Index. An isolate identification was considered final when the first choice listed was described as excellent, very good, or acceptable. In addition, a designation of low selectivity was accepted if confirmed by additional tests as recommended by the manufacturer.

Quantum II Yeast Identification System. The Quantum II system consists of a 20-chamber disposable plastic cartridge containing lyophilized biochemical substrates, a multipunch cartridge perforator, and a dual-wavelength photometer that measured colorimetric changes in the individual cartridge chambers. The readings for the individual biochemical tests are automatically interpreted by the photometer and compared with an established probability matrix by an internal microcomputer. The biochemical results, biocode number, most likely identification, and additional test information (percent likelihood of identification and supplemental tests) are automatically printed out.

The Quantum II tests were performed exactly as specified by the manufacturer. The test inoculum was prepared by suspending a portion of growth from an agar plate in a vial containing 0.5% Noble agar base medium (Difco) and adjusting the turbidity to match that of a no. 0.5 McFarland standard. The top of the 20-chamber cartridge was perforated, and 0.2 ml of the inoculum suspension was dispensed into each reaction chamber. The top of the cartridge was sealed with an adhesive strip, and the cartridge was gently agitated to ensure adequate mixing of the inoculum and reagents in each chamber. The cartridges were incubated at 30°C for 24 h and placed in the Quantum II analyzer. A germ tube test was performed on all isolates, and the results were entered manually into the analyzer. The Quantum II analyzer then interpreted the individual reactions, compared the test results with the stored data base, and printed the identification. An identification was considered final when the likelihood of the first choice was at least 80% or when additional tests suggested on the printout were completed.

API Yeast Ident system. The Yeast Ident system consists of a disposable plastic strip containing 20 microcupules. Each microcupule contains dehydrated substrates or nutrient media or both. Enzymatic tests are carried out with the rehydration of the cupules. All Yeast Ident procedures were conducted as directed by the manufacturer. Yeasts were grown for 48 h at 30°C on Sabouraud dextrose agar and transferred to 3 ml of sterile distilled water to prepare a 2+ Wickerham suspension. Each microcupule was rehydrated with approximately 0.1 ml of the inoculum suspension, and the strip was incubated for 4 h at 35°C in a plastic tray. After 4 h of incubation, the reactions in microcupules 1 through 9 were manually read and recorded. The formation of β-naphthylamides was detected in microcupules 10 to 20 following the addition of one drop of cinnamaldehyde reagent (provided by the manufacturer). A positive color reaction (pink, red, or purple) was read manually at the end of a 3-min incubation period. The color reactions were interpreted by following the indicator color guidelines listed in the API procedure supplement. Numerical values assigned to each microcupule were calculated, and a seven-digit biocode was generated. Yeast identifications were made by referring to the API Yeast Ident Analytical Profile Index included in the procedure supplement. Numbers not listed were called into the API computer service center for reference to their data base. An isolate identification was considered final when the first choice listed was described as excellent, very good, or acceptable. A designation of good likelihood/low selectivity was accepted if confirmed by additional tests as recommended by the manufacturer.

AMS-YBC. The YBC is a 30-well plastic card containing 26 conventional biochemical tests and 4 negative controls. The YBC is used in conjunction with the AMS. Included in the AMS is a programmed computer, a reader/incubation unit, a filling module, and a printer. The automated portion and data management system of the AMS have been described in detail in previous publications (2, 7, 13, 14, 17, 19).

All AMS-YBC procedures were conducted as directed by the manufacturer. An inoculum suspension was prepared by suspending a portion of a 48-h-old culture in 1.8 ml of 0.5% NaCl to a density equal to a no. 2 McFarland standard. The suspension was inoculated into the card via the filling module, incubated at 30°C for 24 h, and read in the reader incubator module. The reading cycle is approximately 1 h. During this time the biochemical patterns are analyzed by the AMS computer and the results are printed. The identification consists of individual biochemical test results, a nine-digit biocode, and a list of one, two, or three of the most likely identifications with their percent probabilities. An identification was considered acceptable if the likelihood of the first choice was greater than or equal to 80%.

Statistical methods. Statistical analysis was performed by chi-square testing with the Yates correction (5).

RESULTS

A total of 221 clinical yeast isolates including 120 common isolates and 101 uncommon isolates were tested with the Quantum II, Yeast Ident, and AMS-YBC yeast identification systems (Table 1). The Quantum II and AMS-YBC systems correctly identified 181 (82%) and 184 (83%) isolates, respec-

TABLE 1. Identification by the Quantum II, API Yeast Indent, and AMS-YBC systems

Organism	No. tested	No. correctly identified by:		
		Quantum II	Yeast Ident	AMS-YBC
Candida albicans[a]	23	23	20	22
Candida tropicalis[a]	39	34	31	32
Candida parapsilosis[a]	24	24	21	20
Candida ciferrii	1	1	1	1
Candida guilliermondii	6	6	5	6
Candida humicola	1	0	0	0
Candida krusei	9	9	8	8
Candida lambica	4	3	1	0
Candida lipolytica	2	1	2	2
Candida lusitaniae	9	9	5	8
Candida paratropicalis	15	1	2	11
Candida pseudotropicalis	2	1	1	2
Candida rugosa	6	4	5	6
Torulopsis glabrata[a]	25	24	4	21
Torulopsis candida	3	0	1	2
Cryptococcus neoformans[a]	9	8	7	8
Cryptococcus laurentii	3	3	2	1
Geotrichum spp.	13	9	2	11
Hansenula sp.	1	0	0	1
Prototheca wickerhamii	1	1	1	0
Rhodotorula spp.	7	5	1	7
Saccharomyces cerevisiae	7	7	3	6
Trichosporon beigelii	10	8	9	8
Trichosporon pullulans	1	0	0	1

[a] Common isolate.

tively ($P > 0.05$), whereas the Yeast Ident system correctly identified 132 (60%) isolates ($P < 0.05$ for the comparison of Yeast Ident with Quantum II and AMS-YBC). Of the 120 common clinical isolates (*C. albicans*, *C. tropicalis*, *C. parapsilosis*, *T. glabrata*, and *Cryptococcus neoformans*), 113 (94%) were correctly identified by Quantum II, 103 (86%) were correctly identified by AMS-YBC ($P > 0.05$ for the comparison of Quantum II with AMS-YBC), and 83 (69%) were correctly identified by Yeast Ident ($P < 0.05$ for the comparison of Yeast Ident with Quantum II and AMS-YBC). Of the 101 uncommon clinical isolates tested, 68 (67%) were correctly identified by Quantum II, 81 (80%) were correctly identified by AMS-YBC ($P > 0.05$ for the comparison with Quantum II), and 49 (49%) were correctly identified by Yeast Ident ($P < 0.05$ for the comparison with Quantum II and AMS-YBC).

Of the 40 misidentifications by Quantum II, 15 were at the genus level and 21 were at the species level, and in 4 instances no identification was assigned to organisms contained within the data base of the Quantum II. The majority of the misidentifications with this system were due to multiple different false-positive and false-negative biochemical test results. Three groups of isolates, *C. paratropicalis*, *C. tropicalis*, and *Geotrichum* spp., accounted for 60% of the misidentifications. *C. paratropicalis*, an organism not included in the Quantum II data base, was the most frequently misidentified organism (14 isolates), followed by *C. tropicalis* and *Geotrichum* spp. (5 isolates each). Although *C. paratropicalis* is not included in the Quantum II data base, the system assigned an incorrect identification to 14 of 15 isolates rather than reporting no identification. Of the 14 misidentified isolates of *C. paratropicalis*, 11 were identified as *C. stellatoidea*. The biochemical profile obtained for these isolates was consistent with that obtained for *C. paratropicalis* when using both conventional biochemicals and the API 20C System.

Of the 89 misidentifications by the Yeast Ident system, 49 were at the genus level and 37 were at the species level and in 3 instances no identification was assigned to organisms contained within the data base of the Yeast Ident system. The misidentifications involved virtually every biochemical test, and the majority were due to multiple false-positive and false-negative test results. The organisms most frequently misidentified were *T. glabrata* (21 isolates), *C. paratropicalis* (13 isolates), *Geotrichum* spp. (11 isolates), *C. tropicalis* (8 isolates), and *Rhodotorula* spp. (8 isolates). These five organisms accounted for 69% of all misidentifications with the Yeast Indent system. In each case the misidentifications included between three and six different yeast species. As was seen with the Quantum II, *C. paratropicalis* was one of the yeasts most commonly misidentified by the Yeast Ident system. The majority (61%) of the misidentified *C. paratropicalis* isolates were identified as *C. krusei* on the basis of false-positive α-D-glucopyranoside, indoxyl acetate, and L-leucyl-glycine-β-naphthylamide reactions.

Of the 37 misidentifications by AMS-YBC, 13 were at the genus level and 18 were at the species level, and in 6 instances no identification was assigned to organisms contained within the data base of AMS-YBC. AMS-YBC correctly classified 8 of 9 isolates of *C. lusitaniae* and 11 of 15 isolates of *C. paratropicalis* as not contained in the data base; however, several isolates including *C. humicola*, *C. lambica*, *C. lusitaniae*, *C. paratropicalis*, and *Prototheca wickerhamii*, which were not included in the AMS-YBC data base, were assigned incorrect identifications. As was observed with the Quantum II and Yeast Ident systems, the misidentifications were due to multiple false-positive and false-negative biochemical tests. The organisms most frequently misidentified were *C. tropicalis* (7 isolates), *C. parapsilosis* (4 isolates), *C. lambica* (4 isolates), *C. paratropicalis* (4 isolates), and *T. glabrata* (4 isolates). These five yeasts accounted for 62% of all misidentifications by AMS-YBC.

DISCUSSION

The results obtained in the present study with the Quantum II Yeast Identification System were similar to those reported by previous investigators (6, 11, 25, 27). Cooper et al. (6) demonstrated an accuracy of 96% for common clinical isolates and 88% for uncommon clinical isolates compared with either API 20C or Uni-Yeast-Tek. Similarly, Salkin et al. (25) reported that Quantum II was 92% accurate in identification of common isolates and only 73% accurate for uncommon isolates when compared with the API 20C. Studies by Kiehn et al. (11) and Sekhon et al. (27) likewise demonstrated accuracies of 80 and 86%, respectively, for Quantum II versus API 20C and conventional identification methods. Using the API 20C results as the correct identification, we found that Quantum II was 94% accurate in identification of common isolates but only 67% accurate for uncommon clinical isolates.

Similarly to Kiehn et al. (11), we did not find any clustering of misidentifications or individual test discrepancies, with the exception of the misidentification of *C. paratropicalis* as *C. stellatoidea* We agree with the previous investigators that the Quantum II data base must be expanded before it is an acceptable system for use in the clinical laboratory. Although Salkin et al. (25) reported several physical problems with the Quantum II system, we did not

encounter problems with the inoculation, cartridge handling, or loading mechanism of the instrument.

The API Yeast Ident was significantly less accurate than either Quantum II or AMS-YBC, regardless of whether common (69%) or uncommon (49%) isolates were considered. Like Salkin et al. (24), we found the data base to be quite limited and the tests involving aminopeptidase substrates to be very difficult to read and interpret. In addition, a large number of the biocodes were not listed in the API Analytical Profile Index, and an identification could be obtained only by calling the API computer service center. Although the Yeast Ident provided an identification in 4 h and hence was the most rapid of the three systems evaluated, it still required a delay of 48 h beyond primary isolation to obtain sufficient growth for inoculum preparation. Overall, the problems with the limited data base and test interpretation make the Yeast Ident system unacceptable for use in the clinical microbiology laboratory.

Previous studies comparing AMS-YBC with the API 20C or conventional identification systems have reported accuracies ranging from 84.0 to 99.0% (7, 13, 14, 19). Land et al. (13) have described their experience with AMS-YBC following an expansion of the YBC data base and improvements in the data analysis scheme and construction of the taxonomic keys. In the first phase of their study, they found a 98.8% correlation between AMS-YBC and API 20C with a panel of 934 yeast isolates; however, a second randomized trial revealed an accuracy of 88% for the identification of varieties of less common perfect and imperfect yeasts. We found AMS-YBC to be somewhat less accurate than did Land et al. (13) and more comparable to the earlier results of Oblack et al. (19) and Hasyn and Buckley (7). These investigators reported overall accuracies of 84.9 and 84.0%, respectively, when compared with Uni-Yeast-Tek and API 20C. The overall accuracy of AMS-YBC (83%) in the present study was comparable to that of Quantum II (82%) and significantly better than that of the API Yeast Ident system (60%). Although AMS-YBC was less accurate than Quantum II in identifying common clinical isolates (86 versus 94%; $P > 0.05$), it was slightly better in identifying the less common isolates (80 versus 67%; $P > 0.05$). As with Quantum II, we did not observe any clustering of misidentifications or individual test discrepancies with AMS-YBC. On the basis of the data obtained in the present study, we believe that additional expansion of the AMS-YBC data base, particularly with the more common yeast isolates, is necessary before this system can be recommended for routine use.

In addition to data base limitations, the accuracy of the Quantum II, Yeast Ident, and AMS-YBC systems may be affected by the inoculum preparation. Organisms which are highly filamentous or encapsulated may pose particular problems in this regard owing to the difficulty in producing a standardized inoculum. Although we did not formally evaluate the effect of inoculum size in the present study, we did observe false-positive and false-negative test results in all systems when the inoculum size was too heavy or too light, respectively. Careful standardization of the inoculum preparation by using a spectrophotometer or hemacytometer may be necessary to ensure optimal performance with these and other yeast identification systems.

A major advantage of the Quantum II and AMS-YBC systems is the use of computer-based instrumentation which can objectively read and interpret the biochemical tests and automatically provide a listing of the most likely identifications without requiring the microbiologist to refer to code books or other listings. Both systems provide results in 24 h. They are also extremely versatile and are capable of identifying bacteria as well as clinical yeast isolates (2, 15, 17, 18, 23). In addition to identifying yeasts and gram-negative bacilli, the Quantum II instrument, with appropriate software, is capable of performing a variety of serologic determinations, including microbial antigen detection (9, 20). The AMS can also quantitate and identify common urinary tract pathogens directly from urine specimens, identify gram-positive organisms, and perform both gram-negative and gram-positive susceptibility testing (15, 17, 21).

In summary, the Quantum II Yeast Identification System provided accurate identification of common clinical yeast isolates in a 24-h time frame but did not perform acceptably in identifying clinically uncommon isolates. The overall accuracy of the Quantum II (82%), AMS-YBC (83%) and API Yeast Ident (60%) systems was not sufficient to recommend any of these systems for routine use in the clinical microbiology laboratory without substantial expansion of their data bases.

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