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NOTES

Rapid Identification of *Candida albicans* by Using Albicans ID and Fluoroplate Agar Plates

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Two commercially available agar media, Albicans ID and Fluoroplate, that use a chromogenic or a fluorogenic substrate for the detection and identification of *Candida albicans* were evaluated. From 1,006 clinical samples containing 723 yeast strains, 352 *C. albicans* strains were detected with either of the two media. The sensitivity of each of the two media was 93.8% and the specificity was 98.6%, with five false-positive reactions for *Candida tropicalis* and no false-negative reactions.

Candida albicans is the most frequently isolated yeast pathogen, and its incidence is increasing because of the development of therapeutic modalities for the treatment of cancer, the use of corticosteroids as immunosuppressants, and the increase in the prevalence of immunocompromising diseases (3). In addition, the number of strains resistant to antifungal agents is increasing (2, 8), so it is important that *C. albicans* species be identified rapidly and reliably in routine clinical microbiological practice.

The use of conjugated substrates to measure enzyme activities has been described previously for the rapid identification of medically important yeasts (1, 13), including *C. albicans* (9, 10, 14). Albicans ID (bioMérieux, Marcy l'Etoile, France) and Fluoroplate (Merck, Damstadt, Federal Republic of Germany) are commercially available media for the rapid identification of *C. albicans*. They contain, respectively, a chromogenic substrate and a fluorogenic substrate hydrolyzed by the hexosaminidase of *C. albicans*, leading to a rapid identification of *C. albicans* on the basis of the colors of the colonies. The aim of the study was to evaluate the capacities of these two media to detect and rapidly identify *C. albicans*.

A total of 1,006 clinical samples (424 stools, 44 urine, 45 vagina, 11 vulva, 5 penis, 27 urethra, 32 mouth, 25 tongue, 37 throat, 49 sputum, and 73 bronchoalveolar fluid and 134 miscellaneous samples such as wounds, eye, and skin) were collected from patients at hospital clinics (Hôpital de l'Antiquaille, Lyon, France) and community outpatient clinics (Institut Pasteur de Lyon, Lyon, France).

Every nonfluid sample was suspended in 1 ml of 0.85% physiologic saline, and then 0.01 ml of this suspension was plated onto Albicans ID and Fluoroplat agar plates. Equivalent amounts of each fluid sample were directly applied onto the media.

Albicans ID is a recently commercialized ready-to-use medium (4) (bio-Thione, 2 g; yeast extract, 6 g; sodium hydrogenophosphate, 0.5 g; chromogenic substrate [hexosamine], 0.05 g; *N*-2-acetamidoimino-diacetic acid monosodium salt buffer, 0.5 g; gentamicin sulfate, 0.1 g; chloramphenicol, 0.05 g; agar, 14 g; completed to 1 liter with distilled water [pH 6.6]) which allows, after hydrolysis by the corresponding enzyme, the specific identification of *C. albicans* colonies on the basis of their blue color and smooth appearance.

Fluoroplate is also a commercially available medium (7) [peptone, 10 g; D-(+)-glucose, 20 g; yeast extract, 2 g; malt extract, 1 g; agar, 17 g; gentamicin, 0.1 g; chloramphenicol, 0.05 g; 4 methylumbellifeyl-*N*-acetyl-β-D-galactosaminide, 0.05 g; completed to 1 liter with distilled water (pH 6.8)] which allows, after hydrolysis by the corresponding enzyme, the specific identification of *C. albicans* colonies on the basis of their fluorescence when excited by 365-nm UV light and their smooth appearance. All other yeasts should appear as white colonies on both Albicans ID and Fluoroplate media.

For the two agars all of the morphologically different colonies were picked and immediately identified by conventional methods (5, 6): germ tube induction in foal serum (Diagnostics Pasteur) at 37°C for 2 to 4 h, chlamydospore formation on rice-agar-Tween medium (bioMérieux), and if necessary, assimilation pattern with the Auxacolor yeast identification panel (Diagnostics Pasteur) tested previously (12). In the case of poor colony isolation, replating of the corresponding colonies was done on the same medium.

The reading of the plates and interpretation of the results were conducted after 24, 48, and 72 h of incubation at 30°C. In order to prevent prejudice in reading the results, the determination of the color and the number of different grown colonies on Albicans ID and Fluoroplate agar plates were performed by two different readers.

Among the 1,006 clinical samples tested, 723 yeast strains belonging to 13 species were detected (Table 1), among which a total of 352 *C. albicans* strains were identified (Table 2). No significant differences in the detection of any other yeast or yeastlike species other than *C. albicans* were noted between the two media (data not shown).

On Albicans ID agar plates, the 330 strains found to have distinguishable smooth blue colonies were confirmed to be *C. albicans* by conventional methods. Five other strains which appeared in 48 or 72 h as colonies with clear blue centers and

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TABLE 1. Yeast and mold strains isolated from 1,006 clinical samples

Yeast and mold species	No.
Candida albicans	352
Geotrichum candidum	179
Candida (Torulopsis) glabrata	87
Saccharomyces cerevisiae	30
Candida tropicalis	15
Candida krusei	14
Candida kefyr	14
Candida parapsilosis	13
Rodotorula rubra	10
Candida inconspicua	4
Trichosporon beigelii	2
Candida famata	2
Candida rugosa	1
Total	723

TABLE 3. Discordances in detection of *C. albicans* on Albicans ID and Fluoroplate agar plates: nature of eventual associated yeast or yeastlike species

Medium on which *C. albicans* was identified	No. of isolates	Associated yeast or yeastlike species
Albicans ID	9	None
	7	*Candida (Torulopsis) glabrata*
	3	*Geotrichum candidum*
	2	*Saccharomyces cerevisiae*
	1	*Geotrichum candidum* + *Saccharomyces cerevisiae*
Fluoroplate	16	None
	4	*Geotrichum candidum*
	1	*Saccharomyces cerevisiae*
	1	*Geotrichum candidum* + *Candida kefyr*

white outlines were further identified by conventional methods to be *Candida tropicalis*.

On Fluoroplate agar plates, 335 strains were found to have smooth fluorescent colonies. Among these 335 fluorescent colonies, 330 were confirmed to be *C. albicans* by conventional methods. The five remaining fluorescent strains were identified as *C. tropicalis*. When they were streaked onto Albicans ID agar plates they appeared as colonies with clear blue centers and white outlines, showing that there were five false-positive results (1.5%) on both Albicans ID and Fluoroplate agar plates. No *C. albicans* isolates were identified among the noncolored colonies on Albicans ID and Fluoroplate agar plates, showing that there were no false-negative results on the two media.

A few discordances in the detection of *C. albicans* between Albicans ID and Fluoroplate agar plates were noted in both monomicrobial and plurimicrobial samples (Table 3). In the 24 cases of monomicrobial samples, the discordances always corresponded to samples containing one to three colonies of *C. albicans* and might be a consequence of the random seeding. Seven *C. albicans* isolates found in association with *Candida (Torulopsis) glabrata* on Albicans ID agar plates were missed on Fluoroplate agar plates (13% of the total number of this combination). The 44 discordant samples corresponded to 24 stools, 3 urine, 3 vagina, 4 throat, 2 sputum, and 8 miscellaneous samples which were equally distributed between the two media.

After 48 h of incubation, all strains of *Geotrichum candidum* showed a diffuse and weak fluorescence on Fluoroplate agar plates and gave pale blue colonies on Albicans ID agar plates. However, colonies of *G. candidum* appeared as typical downy and large colonies and were easily distinguishable from those of *C. albicans*.

Our results obtained with clinical samples are in concordance with previous data conducted with reference strains or clinical isolates, showing the superior performance of methods that use hexosaminidase substrates for the detection and rapid identification of *C. albicans* (7, 9, 14). The technical time is decreased because of the direct identification of *C. albicans* by reading the results for colonies on the primary culture. This property is especially useful when *C. albicans* is associated with other yeasts in clinical samples. In addition, the nature of the clinical samples is of no relevance for the detection and rapid identification of *C. albicans* on these two media. Moreover, Albicans ID and Fluoroplate agar plates allow for the reliable detection and identification of 5% germ tube-negative strains of *C. albicans* that we found in the study and that are described in the literature (11). The sensitivities of the Albicans ID and Fluoroplate agar plates are equal (93.8%), with Albicans ID performing better at 24 h. The specificities of both media were 98.6% because of the presence of false-positive reactions for *C. tropicalis*. However, the appearance of *C. tropicalis* colonies on Albicans ID agar plates is different from that of *C. albicans* and can be distinguished by an experienced technician, while their appearances are not different on Fluoroplate agar plates. On Fluoroplate agar plates, a diffusion of the fluorescent product to the neighboring colonies was often observed after 48 h of incubation. This diffusion enhanced the difficulty of detecting associated yeasts and may lead to an incorrect determination of the real number of associated species. Although *C. (Torulopsis) glabrata* does not seem to inhibit *C. albicans* growth on Fluoroplate agar plates, *C. albicans* may be missed because of a combination of a weak fluorescence and the similar macroscopic features of *C. (Torulopsis) glabrata*. The blue color of *C. albicans* colonies on Albicans ID agar plates does not diffuse into the medium. With Albicans ID agar plates *C. albicans* detection does not require any additional equipment compared with the fluorescence technique. Interference caused by the growth of *G. candidum* was not disturbing because of the specific macroscopic characteristics of this organism.

The Albicans ID chromogenic medium and the Fluoroplate fluorogenic medium permit good detection and rapid macroscopic identification of *C. albicans*. The present study also indicates that Albicans ID agar plates, which are equivalent to Fluoroplate agar plates in sensitivity and specificity, presented several advantages in terms of the ease, rapidity, and reliability that *C. albicans* colonies could be read. Although further studies should be conducted to evaluate the growth of other yeasts on these media, they may be useful in clinical laboratories for the detection and rapid identification of *C. albicans*.

TABLE 2. Detection of *C. albicans* on the two media

Incubation time (h) at 30°C	Cumulative no. (%) of *C. albicans* isolates detected		
	Conventional identification	Albicans ID	Fluoroplate
24		197 (56)	165 (46.9)
48		320 (90.9)	316 (89.8)
72	352	330 (93.8)	330 (93.8)

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