Comparison of HardyCHROM™ Candida with the Becton Dickinson CHROMagar ™ Candida for the Identification of Commonly Isolated Yeast Pathogens

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ABSTRACT

The identification of the most commonly encountered yeast pathogens in the clinical microbiology laboratory has been expedited with the use of chromogenic agars. The chromogenic substrates allow for the differentiation of *Candida albicans, C. tropicalis* and *C. krusei* based on differences in their colony morphologies and color. In addition, the HardyCHROM can identify *C. glabrata* when used in conjunction with a rapid trehalose fermentation test.

In this study, 84 strains of yeast were inoculated onto HardyCHROM™ Candida (HC, Hardy Diagnostics, Santa Maria, CA) and BBL™ CHROMagar™ Candida (CA, BD Diagnostic Systems, Sparks, MD) plates using suspected yeast colonies from primary blood agar plates inoculated with clinical specimens that included 61 urines, 12 wounds and 11 blood cultures. Both chromogenic plates were incubated in a dark, plastic storage box protected from the light, in a non-CO2 incubator at 33° ± 2°C and read at 24 and 48 hours. A total of 43 isolates of *Candida albicans*, 6 isolates of *C. tropicalis* and 2 isolates of *C. krusei* were identified using both chromogenic agar plates. In addition, 10 isolates of *C. glabrata* were identified by selecting smooth pink colonies with darker pink centers from the HC plates and performing a rapid three carbohydrate fermentation test (GlabrataQuick™, Hardy Diagnostics, Santa Maria, CA). All *C. glabrata* isolates were confirmed with the Vitek Yeast Biochemical Card (YBC). A total of 14 other yeast species that were non-chromogenic on both HC and CA were identified using the YBC as *C. parapsilosis* (10), *C. lusitaniae*(3) and *1 C. kefyr* (1). Nine other yeast that were non-chromogenic on HC and CA were not identified further since they were considered non-significant mixed cultures. All *C. albicans*, *C. tropicalis* and *C. krusei* were correctly identified using both of the chromogenic agars read at 24 and 48 hours. The HC performed better than the CA in that the chromogenic reactions showed a darker, more intense color, as well as better colony growth at 24 hours with HC. In addition, 10 isolates of *C. glabrata* were identified using a combination of appearance on HC and the GlabrataQuick kit.

INTRODUCTION

The last decade has shown a dramatic, worldwide increase in fungal infections, which can be attributed to factors such as an increase in the number of patients in intensive care units, cancer patients receiving chemotherapy, HIV positive patients, organ transplant patients and other immunocompromised patient conditions. In addition, the rise of yeast isolates parallels the widespread use of broad-spectrum antibiotics.

Although *Candida albicans* still remains the most frequently isolated yeast pathogen, other *Candida* species such as *Candida glabrata*, *Candida tropicalis, Candida parapsilosis, Candida krusei* and others are recovered in clinical samples.

Due to the increase of fungal infections, the process of identifying *Candida* species in the clinical microbiology laboratory has become more important. In order to accurately and rapidly prescribe the proper antifungal treatment regimens, the laboratory must ensure correct and timely identification of the *Candida* species.

The development of chromogenic agars for *Candida* species has allowed for the rapid differentiation of several *Candida* species. HardyCHROM™ Candida and BBL™ CHROMagar™ Candida are selective and differential media that can identify the most common yeast isolates based on differences in colony morphology and color in pure as well as mixed yeast cultures.

METHODS

This study compared 84 clinical strains of yeast that were isolated from primary blood agar plates and plated to HardyCHROM™ Candida and BD™ CHROMagar™ Candida. Plates were incubated in dark, plastic storage boxes protected from the light, in a non-CO_2 incubator at 33° ± 2° C. Both agar plates were read at 24 and 48 hours. The colony pigments were recorded as green for *Candida albicans*, blue for *Candida tropicalis* and dry, pink colonies for *Candida krusei*. In addition, the HardyCHROM™ agar plates were observed for smooth, pink colonies, having a dark mauve colored center consistent with *Candida glabrata*. These isolates were also tested with the GlabrataQuick™ carbohydrate fermentation test (Hardy Diagnostics) and additionally were set to a Vitek™ Legacy Yeast Biochemical Card (YBC), bioMerieux ™ Inc., Durham, NC. Isolates that were completely non-chromogenic or showing no pigment were identified using the YBC. Nine isolates that were non-chromogenic were not identified further to species level since they were considered non-significant mixed cultures.

RESULTS

Table 1. Comparison of Chromogenic Agars

Yeast ID (No.)	Hardy-CHROM™	BBL™ CHROM-agar™	Additional Tests
Candida albicans (43)	Green-dark green 	Light green to green (fig 1)	
Candida tropicalis (6)	Blue to dark blue 	Light blue to blue (fig 1)	
Candida krusei (2)	Dry, pink 	Dry, pink (fig 1)	
Candida glabrata (10)	Smooth, pink with dark mauve center 	Light pink (no ID, no manufacturer recommend-ation)	Trehalose + Glabrata-Quick™ and confirmed with YBC
Candida parapsilosis (10)	Light purple	Buff	YBC
Candida lusitaniae (3)	Buff to light pink	Buff	YBC
Candida kefyr (1)	Smooth, pink	Buff	YBC
Yeast (9)	White to buff	White to buff	No further ID

RESULTS

Table 2. Identification of Yeast at 24 and 48 hours on Two Chromogenic Agars

Organism	No.	Hardy CHROM™ No. (%) ID at:		BBL™ CHROM-agar™ No. (%) ID at:		Comments
		24 h	48 h	24 h	48 h	
C. albicans	43	43 (100)	-	43 (100)	-	HC had darker, more intense color and larger colonies than CA
C. tropicalis	6	3 (50)	3 (50)	3 (50)	2 (33)	1 isolate did not turn blue on CA until 72 h
C. krusei	2	2 (100)	-	2 (100)	-	
C. glabrata	10	5 (50)	5 (50)	No claim for this organism	No claim for this organism	Presumptive ID of C. glabrata on HC

CONCLUSIONS

Both chromogenic agars provide good clinical results and identified the most commonly encountered yeast isolates. HardyCHROM™ Candida performed better that the BBL CHROMagar™Candida in that the chromogenic pigments showed a darker, richer more intense color, as well as better colony growth at 24 hrs. In addition, HardyCHROM™ Candida has an indication for *Candida glabrata* when used in conjunction with the GlabrataQuick™ kit.The chromogenic media are easy to use, accurate and a cost effective method to use in the laboratory in identifying the most commonly encountered *Candida* species in clinical specimens

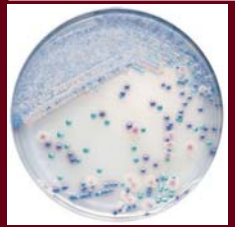

Fig 1. Appearance of Yeast on BBL™ CHROM-agar™
C. albicans – light green to green
C. tropicalis – light blue to blue
C. krusei – Dry pink