Research Article

Identification of various Candida species by using CHROMagar Candida: A rapid screening method

Amit Bhosale[1*], S J Ghosh[2], P G Shadija[3]

[1]Assistant Professor, Department of Microbiology, Bharati Vidyapeeth Deemed University Medical College and Hospital, Sangli, Maharashtra, INDIA.
[2]Associate Professor, [3]Professor and HOD, Department of Microbiology, D Y Patil Medical College, Kasaba Bawda, Kolhapur, Maharashtra, INDIA.
Email: amitbhosale12345@gmail.com

Abstract

Introduction: CHROMagar Candida is a differential culture medium that allows identification of yeasts like *Candida albicans*, *C. tropicalis*, *C. krusei and C. glabrata.* Identification of various candida species by colour of colony on CHROMagar Candida is a rapid and reliable method of speciation of Candida. **Materials and methods:** A total of 172 Candida strains were isolated from various clinical specimens like oral swab, vaginal swab, urine, sputum, bronchial aspirate and other samples. Species identification was first done by using conventional methods by using germ tube test, morphology on cornmeal agar, urease test and growth at 45ºC. All that isolates were again inoculated on CHROMagar Candida and identified by colour and morphology of colony. The results of CHROMagar Candida were compared with the results of conventional methods. **Results:** CHROMagar Candida can easily identify four species of Candida i.e. *Candida albicans*, *C. tropicalis*, *C. krusei and C. glabrata.* Other species were not identified by using CHROMagar Candida. **Conclusion:** CHROMagar Candida is useful for rapid and reliable method for speciation of various clinically important Candida species.

Keywords: CHROMagar Candida, Candida speciation, immunocompromised

[*]**Address for Correspondence:**
Dr. Amit Bhosale, Assistant Professor, Department of Microbiology, Bharati Vidyapeeth Deemed University Medical College and Hospital, Sangli, Maharashtra, INDIA.
Email: amitbhosale12345@gmail.com

Received Date: 22/07/2015 Revised Date: 02/10/2015 Accepted Date: 10/10/2015

Access this article online	
Quick Response Code:	Website: www.statperson.com
	DOI: 16 October 2015

INTRODUCTION

Fungal infections are known to affect the immunocompromised hosts. The advent of HIV and pandemic of AIDS have greatly increased the number of immunocompromised individuals susceptible to a wide variety of infections including mycoses like Candida[1]. The common species found to cause human infections are C. albicans, C. tropicalis, C. parapsilosis, C. glabrata, C. krusei, C. lusitaniae, C. kefyr, C. gullermondii and C. dubliniensis. Candida albicans is the most frequently reported causative agent associated with various lesions in immunocompromised individuals. The frequency of infections caused by non albicans Candida (NAC) is also increasing. The increased frequency of these non albicans species is probably secondary to an alteration in the flora induced by the use of systemic azoles[2]. These species are also shown to have decreased susceptibility to antifungal agents. C. krusei and C. glabrata have innate resistance to fluconazole[3,4]. Hence, identification of Candida species is very important in the laboratory. Identification of species has therapeutic significance, allowing use of appropriate antifungal agents and preventing emergence of drug resistance. Routinely for identification of various Candida species tests like germ tube test, urease test, Growth on cornmeal agar, fermentation and assimilation tests, growth at 45°C are used. These all tests are cumbersome methods of identification. These methods are time consuming also. There are some chromogenic media containing chromogenic substrates, which can be used for the rapid identification of various Candida species. CHROMagar Candida is a selective and differential medium, which facilitates rapid isolation of yeasts from mixed cultures and allows differentiation of Candida species namely C.albicans, C.krusei, C.tropicalis and C.glabrata on the basis of colouration and colony morphology[5]. On this medium results are obtained within

How to site this article: Amit Bhosale, S J Ghosh, P G Shadija. Identification of various Candida species by using CHROMagar Candida: A rapid screening method. *International Journal of Recent Trends in Science and Technology.* October 2015; 16(3): 596-598. http://www.statperson.com (accessed 18 October 2015).

48 hours and it is useful for the rapid and presumptive identification of common yeasts in Mycology and Clinical Microbiology Laboratory. Interpretation of results is easy, whole procedure requires less time and it is cost effective[6]. Chloramphenicol suppresses the accompaning bacterial flora. C.albicans appear as light green coloured smooth colonies, C.tropicalis appear as blue to metallic blue coloured raised colonies. C.glabrata colonies appear as cream to white smooth colonies, while C. krusei appear as purple colonies. Chromogenic medium facilitates the detection of specimens containing mixture of yeast species[6,7].

The present study was done to study the usefulness of CHROMagar Candida as compared to routine conventional methods for identification of various Candida species.

MATERIALS AND METHODS

A total of 172 Candida strains were isolated from various clinical specimens like oral swab, vaginal swab, urine (midstream), sputum, bronchial aspirate and blood. All these isolates grown on Sabouraud dextrose agar. These isolates were identified by using conventional methods of identification. The methods used were urease test, germ tube test, growth on cornmeal agar, growth at 45ºC. All these isolates were inoculated on CHROMagar Candida (Hichrome agar, Himedia, Mumbai, India). These CHROMagar plates were incubated at 37ºC for 48 hrs. Culture plates were observed after 48hrs of incubation. There was luxuriant growth of Candida isolates on CHROMagar Candida. Different isolates produced distinctive coloured colonies. The size, colour, texture of colonies were noted and depending upon that identification of various Candida species were done. The interpretation of colony morphology was done using standard references. The isolates producing light green coloured colonies were identified as C. albicans, producing dark green colonies were identified as C. dubliniensis, producing blue colour were identified as C. tropicalis, producing cream to white coloured colonies were identified as C. glabrata and producing purple colonies were identified as C. krusei. Few isolates gave different morphology by which they could not be identified.

RESULTS

All strains showed luxurious growth on Chrome agar and most of the isolates showed distinctive colony morphology after 48 hrs of incubation. A total of 172 strains were isolated from various clinical specimens. We recovered 62 (36%) isolates from oral swab, 37 (22%) from vaginal swab, 33 (19%) from sputum samples, 23 (13%) from urine samples, 14(8%) from tracheal aspirate and 3 (2%) from pus samples.

Table 1

Specimen	No. of isolates	Percentage
Oral swab	62	36%
Vaginal swab	37	22%
Sputum	33	19%
Urine	23	13%
Tracheal aspirate	14	8%
Pus	03	2%

Out of total 172 isolates, 168 isolates were identified by using colour and colony morphology on chrome agar. Morphology of various Candida species observed on chrome agar Candida is shown in figure 1. Rest four isolates couldn't be identified by using chrome agar. Out of four unidentified isolates, one C. albicans gave off white coloured colonies, one C. tropicalis gave dark pink colonies, one C. glabrata and one C. dubliniensis could not be identified by colour of colony. Chrome agar showed good identification in 99.1% C.albicans isolates, 97.5% in C. tropicalis, 100% in C. krusei, 75% in C. dubliniensis and 90% in C. glabrata.

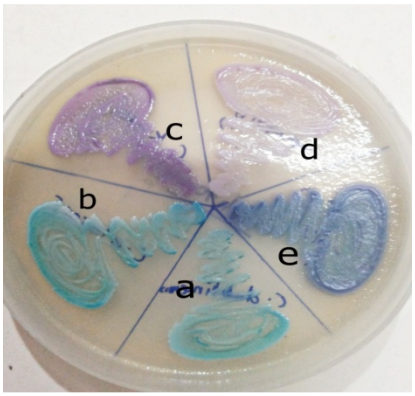

Figure 1: Different Candida isolates grown on Chrome agar Candida

a. **Candida albicans**: Light green colonies.
b. **Candida dubliniensis**: Dark green colonies.
c. **Candida krusei**: Purple colonies.
d. **Candida glabrata**: cream to white coloured colonies.
e. **Candida tropicalis**: blue coloured colonies.

Table 2

Species	Identified by CHROMagar	Identified by conventional methods
C. albicans	111	112
C. tropicalis	40	41
C. krusei	05	05
C. dubliniensis	03	04
C. glabrata	09	10

DISCUSSION

CHROMagar candida is a chromogenic medium which gives rapid presumptive identification of some clinically important Candida species. This medium contains substrates with which the different enzymes produced by Candida species react to form a specific colour. This medium easily and accurately identifies the important Candida species namely C. albicans, C. tropicalis, C. glabrata, C. dubliniensis and C. krusei based on the colour and morphological features.

C. albicans gives light green coloured colonies, C. dubliniensis gives dark green coloured colonies, C. tropicalis gives blue coloured colonies, C. krusei gives pink coloured colonies and C. glabrata gives cream to white coloured colonies on chrome agar.

In our study, Chrome agar showed good identification in 99.1% C.albicans isolates, 97.5% in C. tropicalis, 100% in C. krusei, 75% in C. dubliniensis and 90% in C. glabrata. Out of four unidentified isolates, one C. albicans gave off white coloured colonies, one C. tropicalis gave dark pink colonies and one C. glabrata showed poor growth, one C. dubliniensis could not be identified by colour of colony.

Comparison of percentage of Candida species correctly identified in various studies

Species	Our study	Sanjeev kumar *et al.*[6]	Sayyada ghufrana nadeem *et al.*[8]	Odds and Bernaerts[9]
C. albicans	99.1	100	99	100
C. tropicalis	97.5	100	98	>99
C. krusei	100	100	100	100
C. glabrata	90	100	94	-

The findings in our study matches with various studies as shown in table. There is accurate differenciation between in the three species C. albicans, C. tropicalis and C. krusei. Also the accuracy in identification of C. glabrata was 90%. C. dubliniensis could be identified correctly in 75% isolates, so for identification of these two species it is a good screening method.

Identification of various Candida species also reduces the cost per test, so it reduces the financial burden of poor patients[8]. It identifies the three species i.e. C. albicans, C. tropicalis, C. krusei accurately and can identify majority of C. glabrata, C. dubliniensis isolates. It reduces time required to identify various identification tests.

Rapid identification of C. krusei and C. glabrata isolates with chromogenic media has a special importance because C. glabrata is less sensitive than other species to ketoconazole and fluconazole and C. krusei exhibits innate resistance to fluconazole Hence, rapid identification helps in proper treatment. Thus, Chrome agar can be used in routine diagnostic laboratories for identification of various Candida species.

CONCLUSION

Identification Candida species with CHROMagar Candida is a rapid, reliable, affordable method to identify various commonly isolated Candida species within 48 hrs. Clinicians can choose proper antifungal depending on the identification of Candida species. Identification by this method also reduces the cost per culture thus reduces financial burden on patient. Rapid identification reduces patient mortality and morbidity and because of proper treatment helps in controlling rise in antifungal drug resistance. Thus, CHROMagar Candida medium can be used for rapid identification in routine diagnostic laboratories.

REFERENCES

1. Mandell GL, Bennett JE, Dolin R. Mandell, Cipugias and Bennett's Principles and practice of infectioun diseases. 4th ed. Vol. II. Churchill Livingtone, Edindurgh: 1995
2. McCreary C. Bergin C. Pilkington R, Kelly G, Muicahy F. Clinical parameters associated with recalcitrant oral candidosis in HIV infection: a preliminary study. Int. J. STD and AIDS. 1995, 6 : 204-207.
3. Chander J. A text book of Medical mycology, Candidiasis 3rd edition. New Delhi: Mehta Publishers; 2009. Pp. 266-90.
4. John H Rex JH, Pfaller MA, Walsh TI. Antifungal Susceptibility testing: Practical aspects and Current challenges. Clinical Microbiol Rav 2001; 14: 643-58.
5. Baradkar VP, Mathur M, Kumar S. Hichrome Candida agar for identification of Candida species. Indian Journal Of Pathology and Microbiology, 2010;53(1):93-95.
6. Sanjeev Kumar et.al. Application of CHROMagar Candida for identification of clinically important candida species and their antifungal susceptibility pattern. Internation Journal of Biological and Medical Research. 2013; 4(4): 3600-3606.
7. Yucesoy M, Marol S. Performance of CHROMagar Candida and Biggy agar for identification of yeast species. Annals Clin Microbiol Antimicrobiol 2003;2:1-8
8. Sayyada Ghufrana Nadeem, Shazia Tabassum Hakim and Shahana Urooj Kazmi. Use of CHROMagar Candida for the presumptive identification of Candida species directly from clinical specimens in resource limited settings. Libyan J med 2010, 5:2144.
9. Frank C. Odds and Ria Bernarts. CHROMagar candida, a new differential isolation medium for presumptive identification of clinically important Candida species.

Source of Support: None Declared
Conflict of Interest: None Declared