

# BD<sup>™</sup> Sabouraud GC Agar / CHROMagar<sup>™</sup> Candida Medium (Biplate)

## INTENDED USE

**BD Sabouraud GC Agar / CHROMagar Candida Medium (Biplate)** is used for the selective isolation of fungi and for the isolation and identification of *Candida albicans, C. tropicalis* and *C. krusei* from clinical specimens.

## PRINCIPLES AND EXPLANATION OF THE PROCEDURE

Microbiological method.

Sabouraud Agar with glucose is a widely used medium which, due to its low pH and the high glucose concentration, is partially selective for fungi. As many bacteria tolerate the low pH and the high glucose concentration and will grow on Sabouraud agar, especially during the prolonged incubation period often necessary for fungal isolation, several formulations containing antibacterial inhibitors have been developed. Antimicrobials like penicillin, chloramphenicol, aminoglycosides, or combinations of these have been shown to be effective in inhibiting bacteria without affecting fungal growth.<sup>1-6</sup>

<u>In Sabouraud GC Agar</u> the peptones are sources of nitrogen. Glucose (=dextrose) is an energy source for the growth of fungi. Chloramphenicol and gentamicin are broad-spectrum antibiotics inhibitory to a wide range of gram-negative and gram-positive bacteria.

**<u>CHROMagar Candida Medium</u>** is a selective and differential medium for the isolation of fungi. With the inclusion of chromogenic substrates in the medium, the colonies of *C. albicans, C. tropicalis* and *C. krusei* produce different colors, thus allowing the direct detection of these yeast species on the isolation plate.<sup>7-12</sup> Colonies of *C. albicans* appear light to medium green, *C. tropicalis* colonies appear blue-greenish to metallic-blue, and *C. krusei* colonies appear light rose with a whitish border. Other yeast species may develop either their natural color (cream) or appear rose or light to dark mauve [e.g., *Candida (Torulopsis) glabrata* and other species]. An additional advantage of the medium is the easy detection of mixed yeast cultures due to their colony appearance in different colors.<sup>7,9-12</sup>

Specially selected peptones supply the nutrients in **CHROMagar** Candida Medium. The proprietary chromogen mix consists of artificial substrates (chromogens), which release differently colored compounds upon degradation by specific enzymes. This permits the differentiation of certain species, or the detection of certain groups of organisms, with only a minimum of confirmatory tests. Chloramphenicol inhibits most bacterial contaminants. Titanium dioxide has been added to differentiate this medium from the Sabouraud GC Agar.

**CHROMagar** Candida Medium was developed by A. Rambach and is sold by BD Diagnostic Systems under a licensing agreement with CHROMagar, Paris, France.

## REAGENTS

Formulas\* Per Liter Purified Water

Sabouraud Agar with Gentamicin and Chloramphenicol		CHROMagar Candida Medium	
Pancreatic Digest of Casein	5.0 g	Chromopeptone	10.0 g
Peptic Digest of Animal Tissue	5.0	Glucose	20.0
Glucose	40.0	Chromogen Mix	2.0
Agar	15.0	Chloramphenicol	0.5
Gentamicin	0.04	Titanium dioxide	0.35
Chloramphenicol	0.4	Agar	15.0
pH 5.6 +/- 0.3		pH 6.0+/- 0.3	

\*Adjusted and/or supplemented as required to meet performance criteria.

## PRECAUTIONS

IVD . For professional use only. 🛞

Do not use plates if they show evidence of microbial contamination, discoloration, drying, cracking or other signs of deterioration.

Consult **GENERAL INSTRUCTIONS FOR USE** document for aseptic handling procedures, biohazards, and disposal of used product.

## STORAGE AND SHELF LIFE

On receipt, store plates **in the dark** at 2 to 8° C, in their original sleeve wrapping until just prior to use. Avoid freezing and overheating. The plates may be inoculated up to the expiration date (see package label) and incubated for the recommended incubation times.

Plates from opened stacks of 10 plates can be used for one week when stored in a clean area at 2 to 8° C.

Minimize exposure to light before and during incubation, since light may destroy the chromogens.

## USER QUALITY CONTROL

Inoculate representative samples with the following strains (for details, see **GENERAL INSTRUCTIONS FOR USE** document). Incubate the plates aerobically for 20 to 48 hours at 35 +/- 2° C

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Strains	Sabouraud GC Agar	CHROMagar Candida Medium
Candida albicans	Growth good to excellent;	Growth good to excellent; light
ATCC™ 60193	white colonies	to medium green colonies
Candida krusei	Growth good to excellent;	Growth good to excellent; light
ATCC 34135	white to creme, flat colonies	rose to pink, flat colonies with a whitish border
Candida tropicalis	Growth good to excellent;	Growth good to excellent; gray
ATCC 1369	white to creme colonies	blue to blue-greenish or metallic blue colonies with or without violet halos in the surrounding medium
Pseudomonas aeruginosa ATCC 27853	Inhibition partial to complete	Inhibition partial to complete
*Aspergillus niger ATCC 16404	Growth good to excellent	Growth good to excellent
Uninoculated	Light amber, transparent	Milky white, opaque

\* may be incubated for up to 4 days

## PROCEDURE

#### Materials Provided

**BD Sabouraud GC Agar / CHROMagar Candida Medium** (90 mm **Stacker™** biplates). Microbiologically controlled.

For differentiation of the two media of this biplate, <u>CHROMagar Candida Medium</u> contains titanium dioxide (see formula) which renders the medium milky white and opaque while Sabouraud GC Agar is transparent.

#### Materials Not Provided

Ancillary culture media, reagents and laboratory equipment as required.

#### Specimen Types

The media in this biplate are used for the isolation of fungi and for isolation and identification of *Candida albicans, C. tropicalis, and C. krusei* from all types of clinical specimens (see also **PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE**).

#### **Test Procedure**

Streak the specimen or culture for isolation onto the surface of each medium. If the specimen is cultured from a swab, roll the swab gently over a small area of each surface at the edge, then streak from these areas with a loop. Incubate plates aerobically at 35 +/- 2° C. Minimize exposure to light both before and during incubation. Read <u>CHROMagar Candida Medium</u> after 42 to 48 h. As some slow-growing filamentous fungi may require a longer incubation, return the plate to the incubator until day 4 or longer. After this time, inspect <u>Sabouraud GC Agar</u> for additional isolates not yet found on **CHROMagar** Candida Medium, but do not read **CHROMagar** Candida Medium again after the extended incubation. Isolates on Sabouraud GC Agar must be further differentiated for complete identification.<sup>3-6</sup> Occasional isolates, such as *Cryptococcus neoformans* and filamentous fungi, will require a longer incubation time and possibly a lower incubation temperature. Therefore, it is recommended to inoculate a **BD Sabouraud Agar with Gentamicin and Chloramphenicol** plate with the specimen and to incubate this plate at 25 to 30° C, if fungi requiring a lower incubation temperature are expected.

## Results

After sufficient incubation, examine <u>Sabouraud GC Agar</u> for fungal colonies exhibiting typical color and morphology. Biochemical tests and microscopical and serological procedures should be performed for complete identification of isolates.<sup>3-6</sup>

**CHROMagar** Candida Medium: It is recommended to read this medium on a white background. If *Candida* species are present, colonies will appear light to medium green (*C. albicans*), light rose to pink with a whitish border (*C. krusei*), or blue-greenish to metallic blue with or without violet halos (*C. tropicalis*). Other *Candida* species and other yeasts appear light to dark mauve (rose to violet) or, if none of the chromogenic substrates is utilized, will assume their natural colony color (cream to white).

Data from various studies indicate that further identification tests are not necessary for *Candida albicans, C. tropicalis,* and *C. krusei.*<sup>7,9-11</sup>

Colonies that appear light to dark rose or mauve to violet, or appear in their natural cream color must be identified using standard methods.<sup>7-11</sup>

# PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE

**BD Sabouraud GC Agar / CHROMagar Candida Medium (Biplate)** is used for the selective isolation of fungi (Sabouraud GC Agar) and for the isolation and identification of *C. albicans, C. krusei, and C. tropicalis* (CHROMagar Candida Medium).

<u>Sabouraud GC Agar</u> is a conventional medium, widely used for the selective isolation of fungi. Isolates from this medium must be further differentiated using the classical procedures for fungal identification.<sup>1-6</sup>

*Nocardia* and *Actinomyces* are filamentous bacteria (no fungi!) and, therefore, do not grow on Sabouraud media containing bacterial inhibitors.

Consult appropriate references for detailed information and recommended procedures for the identification of isolates.<sup>3-6,8</sup>

Use of <u>CHROMagar Candida Medium</u> for direct identification of *C. albicans, C. krusei, and C. tropicalis* has been documented in several studies and manuals which can also be consulted for additional information on the recommended procedures.<sup>7,9-12</sup> Results of a recent performance evaluation of **BD CHROMagar Candida Medium** are reported by Jabra-Rizk and coworkers.<sup>11</sup>

*Candida (Torulopsis) glabrata* usually produces mauve to dark mauve colonies on this medium.<sup>9</sup> However, it is recommended that organisms appearing in this color should be confirmed with additional biochemical tests since this colony color can be produced by a variety of yeast species.

Colonies that appear rose or light to dark mauve, or appear in their natural cream color on this medium must be identified using standard methods.<sup>3-5</sup>

Fungi other than yeasts can also be isolated on this medium if incubated at a temperature and for a time appropriate for these organisms.

Since filamentous fungi may metabolize the chromogenic substrates, the colors exhibited by these organisms on **CHROMagar** Candida Medium may differ from those exhibited on other fungal media. Do not use the appearance of growth of filamentous fungi on this medium for traditional morphological identification.

It has been reported that *C. dubliniensis* produces a distinctive dark green color on primary isolation with **CHROMagar** Candida Medium.<sup>13-15</sup> However, this property may not be retained in subculture. Additional phenotypic and genotypic tests are necessary for confirmation of *C. dubliniensis*. Simple phenotypic tests, e.g., growth of the isolate at 45° C (*C. dubliniensis*: negative; *C. albicans*: positive) may be used for the differentiation of the two species.<sup>12</sup>

Before using **BD CHROMagar Candida Medium** for the first time, we recommend to train the typical colony appearance with defined strains of *C. albicans, C. krusei, and C. tropicalis*, e.g., the strains mentioned under **USER QUALITY CONTROL**.

Several filamentous fungi require lower incubation temperatures than those necessary for **BD Sabouraud GC Agar / CHROMagar Candida Medium (Biplate)**. Incubation of this biplate at temperatures lower than 35° C, however, may delay the chromogenic reactions on **CHROMagar** Candida Medium.

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## PACKAGING/AVAILABILITY

#### BD Sabouraud GC Agar / CHROMagar Candida Medium (Biplate)

Cat. No. 254515	Ready-to-use Plated Media, cpu 20
Cat. No. 257663	Ready-to-use Plated Media, cpu 120

#### FURTHER INFORMATION

For further information please contact your local BD representative.



#### **Becton Dickinson GmbH**

Tullastrasse 8 – 12 D-69126 Heidelberg/Germany Phone: +49-62 21-30 50 Fax: +49-62 21-30 52 16 Reception\_Germany@europe.bd.com

http://www.bd.com http://www.bd.com/europe/regulatory/

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