

Candida Selective Supplement

PRODUCT INFORMATION

- C028-5g - Chloramphenicol, Powder, 5g
- C028-25g - Chloramphenicol, Powder, 25g
- C028-100g - Chloramphenicol, Powder, 100g
- C028-500g - Chloramphenicol, Powder, 500g
- C028-1kg - Chloramphenicol, Powder, 1kg
- C090-10ml - Chloramphenicol, Solution, 10ml

DESCRIPTION

Chromogenic Candida Agar with Candida Selective Supplement is a selective differential medium for the rapid isolation and identification of clinically important *Candida* species.

BACKGROUND

Chloramphenicol is a bacteriostatic antimicrobial. It is considered a prototypical broad-spectrum antibiotic, alongside the tetracyclines. Chloramphenicol is effective against a wide variety of Gram-positive and Gram-negative bacteria, including most anaerobic organisms.

Mechanism of action

Chloramphenicol is bacteriostatic. It is a protein synthesis inhibitor, causing inhibition of peptidyl transferase activity of the bacterial ribosome, binding to A2451 and A2452 residues in the 23S rRNA of the 50S ribosomal subunit, preventing peptide bond formation. While chloramphenicol and the macrolide class of antibiotics both interact with ribosomes, chloramphenicol is not a macrolide. It directly interferes with substrate binding; macrolides sterically block the progression of the growing peptide.

APPLICATION IN CHROMOGENIC CANDIDA AGAR

Chromogenic Candida Agar allows the differentiation of *Candida albicans* and *Candida tropicalis* from other species of *Candida* within 48 hours. This is important because rapid identification has been shown to have an impact on the morbidity, mortality, and duration of

hospitalization. However, conventional identification of *Candida* spp. is based on a time-consuming and extensive series of tests, e.g. carbohydrate fermentation and assimilation, growth at 37°C and 42°C, colony and cell morphology and the ability to form germ tubes.

Serious infections due to *Candida* species are becoming increasingly prevalent. This poses particular problems because of the increasing incidence of non-*albicans* spp. and the emergence of non-*albicans* isolates resistant to both amphotericin B and the newer azoles. *Candida* species are the fourth most commonly encountered nosocomial pathogens in bloodstream infections in the United States, and candidiasis is associated with mortality rates as high as 60% in immuno-suppressed patients.

Of the *Candida* spp. encountered in clinical practice, *Candida albicans* is the most common, and this species is usually susceptible to the azole group of antifungal agents. However, it is the shift toward the isolation of more azole-tolerant species, such as *Candida glabrata*, *Candida tropicalis*, and *Candida krusei*, due to the increasing use of itraconazole and fluconazole as the antifungal drugs of first choice for candidiasis, which is causing greatest concern. Rapid identification of the *Candida* spp. causing infection is, therefore, critical for the clinician in determining the correct antifungal therapy.

For *Candida* species involved in bloodstream infections on ICUs, it was shown by Ibrahim et al. that initial therapy was inadequate in 95% of the cases because no antifungal agent was administered. Due to this inadequacy in the initial treatment, a mortality rate of about 60% was observed in the patient group with *Candida* infections. Hence, early recognition of a *Candida* infection would help a clinician to select proper treatment. Combined with rapid identification of the causative organism, this treatment could be optimized to include a non-azole group anti-fungal agent, if required, at an early stage of the infection.

Chromogenic Candida Agar incorporates two chromogens that indicate the presence of the target enzymes:

- X-NAG (5-bromo-4-chloro-3-indolyl N acetyl β -D-glucosaminide) detects the activity of hexosaminidase.
- BCIP (5-bromo-6-chloro-3-indolyl phosphate p-toluidine salt) detects alkaline phosphatase activity.

The typical enzyme patterns of *Candida* spp. are shown

in Table 1. An opaque agent has been incorporated into the formulation to improve the colour definition on the agar. The broad-spectrum antibacterial agent, chloramphenicol, is added to the agar at 500mg/l to inhibit bacterial growth on the plates.

Chromogen:	X-NAG	BCIP	Typical colony appearance
Enzyme:	Hexosaminidase	Alkaline phosphatase	
<i>C. tropicalis</i>	+		Dark blue
<i>C. albicans</i>	+		Green ‡
<i>C. dubliniensis</i>			
<i>C. krusei</i>		+	Dry, irregular pink-brown
<i>C. glabrata</i>		Variable	Beige/yellow/brown †
<i>C. kefyr</i>			
<i>C. parapsilosis</i>			
<i>C. lusitanae</i>			

Table 1 - Table of expected reactions on Chromogenic Candida Agar

Notes:

‡ The green colour of *Candida albicans* and *Candida dubliniensis* is caused by the same chromogenic reaction as the dark blue colour of *Candida tropicalis*. However, other reactions caused by the medium cause the colonies to appear green.

† *Candida glabrata*, *Candida kefyr*, *Candida parapsilosis* and *Candida lusitanae* appear as a variety of beige/brown/yellow colours, due to the mixture of natural pigmentation and some alkaline phosphatase activity. Experienced users may be able to differentiate these species by colour and colony morphology.

Content concentrations

Typical Formula*	mg/litre
Chromogenic Candida Agar	
Peptone	4
Chromogenic mix	13.6
Agar	13.6
Final pH 6.0 ± 0.2 @ 25°C	
Candida Selective Supplement	
Chloramphenicol	500
* Adjusted as required to meet performance standards	

Table 2 Typical Formula for Chromogenic Candida Agar and Candida Selective Supplement

METHOD

Preparation

Suspend appropriate amount of Chromogenic Candida Agar in distilled water. Add Candida Selective Supplement reconstituted as directed. Mix well and bring to the boil with frequent agitation. DO NOT

AUTOCLAVE. Cool the medium to 45°C and pour into sterile Petri dishes.

Protocol

Good laboratory practices for the appropriate collection and transport of specimens should be followed. Clinical specimens should be inoculated directly onto the agar. Incubate plates aerobically at 30°C. Inspect for the growth of *Candida* spp. at 24, 48 and 72 hours.

Quality control

Positive control:

Candida albicans ATCC®10231: Good growth; green colonies

Candida krusei ATCC®6258: Good growth; dry, irregular pink-brown colonies

Negative control:

Escherichia coli ATCC® 25922: Inhibited

REFERENCES

1. Sheehan, D. J. et al. (1999) Current and Emerging Azole Antifungal Agents *Clinical Microbiology Reviews*, 12(1): 40-79
2. Odds, F. C. (1988) *Candida and candidosis*, 2nd ed. Baillière Tindall, London, England.
3. Ibrahim E.H. et al. (2001) The influence of inadequate antimicrobial treatment of bloodstream infections on patient outcomes in the ICU setting. *Chest*, 118(1):146-55