

INSTRUCTIONS FOR USE – READY-TO-USE PLATED MEDIA

BD[™] Cepacia Medium • **BD™ OFPBL Agar**

INTENDED USE

BD Cepacia Medium and BD OFPBL Agar are selective differential media for the isolation of Burkholderia cepacia from clinical specimens.

PRINCIPLES AND EXPLANATION OF THE PROCEDURE

Microbiological method.

Burkholderia (formerly Pseudomonas) cepacia is well recognized as a nosocomial pathogen usually causing infections associated with contaminated equipment, disinfectants, and medications. Infections include bacteremia, urinary and respiratory tract infections, and others.¹ Additionally, the organism is an important pathogen in patients with cystic fibrosis (CF), also called mucoviscidosis, ¹⁻³ and in patients with chronic granulomatous disease.⁴ Recovery of the organism on routine media, such as blood agar or MacConkey Agar is sometimes difficult because other organisms such as Staphylococcus or Pseudomonas aeruginosa frequently overgrow and mask its presence.^{1,2,5}

BD OFPBL (Oxidation/Fermentation-Polymyxin-Bacitracin-Lactose) Agar is used for the isolation of *B. cepacia*, and is reported to be slightly less selective than **BD Cepacia Medium**.^{1,6} **BD OFPBL Agar** is based on O/F Medium (Oxidation/Fermentation Medium). The pH indicator bromthymol blue changes from blue to yellow when lactose is fermented to acid products. Phosphate is added to stabilize the pH. Bacitracin and polymyxin B act as inhibitors of accompanying bacteria.

BD Cepacia Medium is also used for the isolation of B. cepacia. Peptones and ammonium sulfate supply nitrogen. Pyruvate is the carbon source. Phenol Red is used as a pH indicator. During the metabolism of pyruvate, the sodium ions accumulate, raising the pH. This results in a color change of phenol red from yellow-orange to pink or red around B. cepacia colonies and intensifies to pink in areas of dense growth. Bile salts, crystal violet, ticarcillin, and polymyxin B act as inhibitors to suppress normal flora and other pathogens. The phosphates are included to maintain the pH stable while magnesium and iron are growth factors for many nonfermenters formerly included into the genus Pseudomonas.

Both media are recommended for the isolation of *B. cepacia* from clinical specimens.^{1,7,8}

Formulas* Per Liter Purified Water			
BD Cepacia Medium		BD OFPBL Agar	
Peptones	8.0 g	Pancreatic Digest of Casein	2.0 g
Ammonium Sulfate	1.0	Sodium Chloride	5.0
Sodium Pyruvate	5.0	Dipotassium Hydrogen Phosphate	0.3
Magnesium Sulfate	0.2	Bromthymol Blue	0.03
Ferrous Ammonium Sulphate	0.01	Lactose	10.0
Potassium Dihydrogen Phosphate	4.35	Agar	15.0 g
Disodium Hydrogen Phosphate	1.42	Polymyxin B	300000 U
Phenol Red	0.02	Bacitracin	200 U
Bile Salts	0.5	pH 6.8 +/- 0.2	
Agar	12.0		
Ticarcillin	0.1		
Polymyxin B	300000 U		
Crystal Violet	1.0 mg]	
pH 6.3 +/-0.2			

REAGENTS

*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.

USER QUALITY CONTROL

Inoculate the plates with the strains mentioned below. Incubate for 20 to 24 hours aerobically at 30 to 35° C or at 35 to 37° C.

Strains	BD Cepacia Medium	BD OFPBL Agar
Burkholderia cepacia	Growth fair to excellent;	Growth good to excellent;
ATCC™ 25416	colonies pale yellowish to rose,	colonies transparent to yellow;
	rose to pink-red medium	yellow medium
Escherichia coli ATCC 25922	Inhibition complete	Inhibition complete
Pseudomonas aeruginosa	Inhibition partial to complete	Inhibition complete
ATCC 27853		
Stenotrophomonas maltophilia	Inhibition partial to complete	Inhibition partial to complete
ATCC 13637		
Staphylococcus aureus	Inhibition partial to complete	Inhibition complete
ATCC 25923		
Uninoculated	Yellow to orange	Green

PROCEDURE

Materials Provided

BD Cepacia Medium or **BD OFPBL Agar**, both provided in **Stacker™** plates. Microbiologically controlled.

Materials Not Provided

Ancillary culture media, reagents and laboratory equipment as required.

Specimen Types

These media are used for the isolation from clinical specimens such as respiratory tract, urinary tract, an others (see also **PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE**). Specimens from CF patients include bronchoalveolar lavage fluid (preferred), sputum, nasolaryngeal aspirates, and oropharyngeal swabs.

Test Procedure

Inoculate one of these media with the specimen after it arrives in the laboratory. Streak for isolation. In addition to **BD Cepacia Medium** or **BD OFPBL Agar**, inoculate **BD Columbia Agar** with 5% Sheep Blood and **BD MacConkey II Agar** with the specimen, in order to isolate all pathogens involved in the infection. Incubate **BD Cepacia Medium** or **BD OFPBL Agar** aerobically at 30 to 35° C or at 35 to 37° C for 18 to 24 hours or longer, if necessary. Some strains of *B. cepacia* prefer the lower temperature range or will need an incubation of 72 hours for full growth. Incubate the other media as appropriate.

Results

After the incubation, typical colonies of *B. cepacia* on **BD Cepacia Medium** are pale yellowish to rose, surrounded by rose to pink-red zones. On **BD OFPBL Agar**, *B. cepacia* colonies are transparent to yellow, surrounded by yellow zones.

PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE

BD Cepacia Medium and **BD OFPBL Agar** are recommended for the isolation of *Burkholderia cepacia* from clinical specimens suspected to contain contaminating normal flora, such as (but not limited to) respiratory and urinary tract specimens.^{1,7,8}

Gilligan et al., who developed Pseudomonas Cepacia Agar as a selective differential medium, reported isolation of *B. cepacia* from 35 CF patients' respiratory secretions on this medium, while only 21 isolates were obtained on MacConkey Agar.⁵

Occasionally, other bacteria that are resistant to the selective agents will grow on these media. *Burkholderia gladioli* which has been shown to occur in respiratory tract specimens of CF patients, will grow on OFPBL Agar and may resemble *B. cepacia.*⁹

Further tests such as a Gram stain and a complete biochemical identification are necessary to confirm the presence of *B. cepacia*.¹

REFERENCES

- Gilligan, P.H., G. Lum, P.A.R. Vandamme, and S. Whittier. 2003. Burkholderia, Stenotrophomonas, Ralstonia, Brevundimonas, Comamonas, Delftia, Pandoraea, and Acidovorax. In: Murray, P. R., E. J. Baron, J.H. Jorgensen, M. A. Pfaller, and R. H. Yolken (ed.). Manual of clinical microbiology, 8thed. American Society for Microbiology, Washington, D.C.
- Gilligan, P.H. 1991. Microbiology of airway disease in patients with cystic fibrosis. Clin. Microbiol. Rev. 4: 35-51.
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- 9. Christenson, J.C., et al. 1989. Recovery of *Pseudomonas gladioli* from respiratory tract specimens of patients with cystic fibrosis. J. Clin. Microbiol. 27: 270-273.

PACKAGING/AVAILABILITY

BD Cepacia Medium

Cat. No. 256180 Ready-to-use Plated Media, cpu 20

BD OFPBL Agar

Cat. No. 254481

Ready-to-use Plated Media, cpu 20

FURTHER INFORMATION

For further information please contact your local BD representative.



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