

Technical Information

Burkholderia Cepacia Agar Base

Product Code: DM 2640

Application: - Burkholderia Cepacia Agar Base is a selective medium recommended for isolation of *Burkholderia cepacia* from the respiratory secretions of patients with cystic fibrosis and other non-clinical specimens

Composition**

Ingredients	Gms / Litre
Peptone	5.000
Yeast extract	4.000
Sodium pyruvate	7.000
Potassium dihydrogen phosphate	4.400
Disodium hydrogen phosphate	1.400
Bile salts	1.500
Ammonium sulphate	1.000
Magnesium sulphate	0.200
Ammonium ferrous sulphate	0.010
Phenol red	0.020
Crystal violet	0.001
Agar	12.000
Final pH (at 25°C)	6.2±0.2

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

Burkholderia cepacia is an important opportunistic pathogen and causes pulmonary infection among individuals with cystic fibrosis (CF). The organism may lead to *Burkholderia cepacia* syndrome, a neutralizing pneumonia associated with fever that culminates in to a rapid and fatal clinical deterioration (1). *B. cepacia* is difficult to isolate on routinely used laboratory media like MacConkey Agar, since *B. cepacia* is a slow grower and therefore it is usually outgrown by the faster growing *Escherichia coli*, *Staphylococcus aureus*, and *Pseudomonas aeruginosa*. Burkholderia Cepacia Agar is based on PC medium, which was originally devised by Gilligan (2). This medium was found to be superior to MacConkey Agar for growth of *B. cepacia*. The medium is made selective for *B. cepacia* by the incorporation of bile salts, crystal violet and antibiotics. The antibiotics included are Polymyxin B, Gentamycin, Ticarcillin in the form of freeze dried supplement (FD). Peptone and yeast extract in the medium supplies the nitrogenous, vitamin B source and other essential nutrients. Crystal violet, bile salts and antimicrobial agents are used as selective agents. Crystal violet and bile salts inhibits gram-positive cocci including Enterococci and Staphylococci. The antibiotics (FD) namely ticarcillin, polymixin B and gentamycin prevent gram- negative bacteria. *B. cepacia* metabolizes pyruvate forming alkaline end products. These end products elevate the pH of the medium. The phenol red indicator changes colour from pink orange to pink red in alkaline pH.

Inoculate the plate with the specimen so as to obtain isolated colonies. The plates should be incubated for a period of 4 days to allow *B. cepacia* to grow and form colonies and subsequent colour change (3, 4). The medium is not selective only for *B. cepacia*. Other organisms forming similar colonies may also grow on this medium. Therefore results obtained on this media should not be the sole criteria for identification of *B. cepacia* (5).

Methodology

Suspend 18.26 grams of dehydrated media in 500 ml distilled water. Mix thoroughly & heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 50°C and aseptically add the rehydrated contents of 1 vial of Burkholderia Selective Supplement (MS2232). Shake well before pouring in sterile Petri plates.

Quality Control

Appearance

Light yellow to pink homogeneous free flowing powder

Gelling

Firm, comparable with 1.2% Agar gel.

Colour and Clarity

Orange coloured clear to slightly opalescent gel forms in Petri plates

Reaction

Reaction of 3.65% w/v aqueous solution at 25°C. pH : 6.2±0.2

pH Range

6.00-6.40

Cultural Response

DM2640: Cultural characteristics observed, with addition of Burkholderia Selective Supplement (MS2232), after an incubation at 35-37°C for 48-72 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony
Cultural Response				
<i>Burkholderia cepacia</i> ATCC 25608	50-100	good-luxuriant	>=50%	sage green colonies with bright pink medium
<i>Pseudomonas aeruginosa</i> ATCC 9027	>=10 ³	inhibited	0%	-

Storage and Shelf Life

Dried Media: Store below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label.

Prepared Media: 2-8° in sealable plastic bags for 2-5 days.

Further Reading

1. Whitby P. W., 1998, J. Clin. Microbiol., 36:1642 1645
2. Gilligar, Gage, Bradshaw, schidlow and Deciscco, 1985, J. Clin. Microbiol., 22:5.
3. MacDonald Gilligan, Welch, Reller and Menegus, 1994, Vol. 5:1, Cystic Fibrosis Foundation, Washington, D.C.
4. Gilligan, 1996. Clin. Microbiol. Newsl. 18:83.
5. Christensen et al, 1980, J. Clin. Microbiol., 27:270.

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