Dehydrated Culture Media
Bases / Media Supplements

Technical Information

Burkholderia Cepacia Selective Agar Base

Product Code: DM 3089U

Application: Recommended for selective medium used for isolation of Burkholderia cepacia from pharmaceutical products, the respiratory secretions of patients with cystic fibrosis and other non-clinical specimens in accordance with USP.

Composition**

Ingredients	Gms / Litre
Casitose #	10.000
Lactose	10.000
Sucrose	10.000
Sodium chloride	5.000
Yeast extract	1.500
Phenol red	0.080
Crystal violet	0.002
Agar	14.000
pH after sterilization (at 25°C)	6.8± 0.3

**Formula adjusted, standardized to suit performance parameters
casein peptone

Principle & Interpretation

Burkholderia cepacia is an important opportunistic pathogen and causes pulmonary infection among individuals with cystic fibrosis (CF). Burkholderia cepacia species are gram negative, rod shaped bacteria. 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). Burkholderia cepacia species may cause severe infection in individuals with cystic fibrosis and immunosuppressed individuals. 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. Burkholderia cepacia have the potential of overcoming antimicrobial preservative systems and antiseptics, and can grow in preserved aqueous oral liquids and topical products. This medium is recommended for detection of Burkholderia cepacia in pharmaceutical products (3).

Casitose and yeast extract in the medium provides the carbonaceous, nitrogenous, long chain amino acids, vitamin B source and other essential nutrients. Crystal violet and antimicrobial agents are used as selective agents. Crystal violet and vancomycin inhibits gram-positive cocci including Enterococci and Staphylococci. The antibiotics namely polymyxin B and gentamicin inhibits gram-negative bacteria.

B. cepacia metabolizes pyruvate forming alkaline end products. Sucrose and Lactose are the fermentable carbohydrate. The phenol red indicator changes colour from pink orange to pink red in alkaline pH. Test procedure: The sample is initially enriched in Soyabean Casein Digest Medium and then plated on Burkholderia cepacia Selective Agar.

Type of specimen

Clinical samples: Respiratory secretions, Pharmaceutical samples

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (4,5). For pharmaceutical samples follow appropriate techniques for handling specimens as per established guidelines (3). After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

In Vitro diagnostic use. For professional use only. Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling clinical specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Other bacteria resistant to the selective agents may grow on this media.
2. Further biochemical characterization is necessary for complete identification.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Methodology

Suspend 50.58 grams in 1000 ml purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C and aseptically add the rehydrated contents of one vial of BCSA Selective supplement (MS2361). Mix well and pour in sterile Petri plates.

Quality Control

Appearance

Light yellow to pink homogeneous free flowing powder

Gelling

Firm, comparable with 1.4% Agar gel.

Colour and Clarity of prepared medium

Orange coloured clear to slightly opalescent gel forms in Petri plates

Reaction

Reaction of 5.06% w/v aqueous solution at 25°C . pH : 6.8 ±0.3

pH

6.50 -7.10

Cultural Response

Cultural response was observed after an incubation at 30-35°C for 48-72 hours. Recovery rate is considered as 100% for bacteria growth on Soybean Casein Digest Agar.

Growth promoting properties

Growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating 100 cfu (at 30-35°C for <=48 hours).

Indicative properties

Colonies are comparable in appearance and indication reaction to those previously obtained with previously tested and approved lot of medium occurs for the specified temperature for a period of time within the range specified inoculating <=100 cfu (at 30-35°C for 48-72 hours).

Inhibitory properties

No growth of the test microorganism occurs for the specified temp for not less than longest period of time specified inoculating >=100cfu (at 30-35°C for >= 72 hours).

Organism	Inoculum (CFU)	Growth	Recovery	Colour of Colony
Burkholderia cepacia ATCC 25416	50-100	good-luxuriant	>=50%	greenish brown colonies w/yellow halo or white colonies surrounded by pink red zone
Burkholderia cenocepacia ATCC BAA-245	50-100	good-luxuriant	>=50%	greenish brown colonies w/yellow halo or white colonies surrounded by pink red zone
Burkholderia multivorans ATCC BAA-247	50-100	good-luxuriant	>=50%	greenish brown colonies w/yellow halo or white colonies surrounded by pink zone
^*Pseudomonas paraeruginosa* ATCC 9027 (00026*)	>=10^3	inhibited	0%	
Staphylococcus aureus subsp.*aureus* ATCC 6538 (00032*)	>=10^3	inhibited	0%	

Additional Microbiological tesing

Burkholderia cenocepacia ATCC 25608	50-100	good-luxuriant	>=50%	greenish brown colonies w/yellow halo or white colonies surrounded by pink red zone

Key: (*) Corresponding WDCM numbers ^ Formerly known as *Pseudomonas aeruginosa*

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (4,5).

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. The United States Pharmacopoeia, 2022, Microbial exmination of nonsterile products- Tests for *Burkholderia cepacia* complex. The United States Pharmacopoeial Convention. Rockville, MD.
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
5. Jorgensen, J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Disclaimer :

- User must ensure suitability of the product(s) in their application prior to use.
- The product conforms solely to the technical information provided in this booklet and to the best of knowledge research and development work carried at **CDH** is true and accurate.
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