PRODUCT SPECIFICATION SHEET

Burkholderia Cepacia Agar Base (DM1042)

Intended Use

Burkholderia Cepacia Agar Base (DM1042) is a selective medium used for isolation of *Burkholderia cepacia* from the respiratory secretions of patients with cystic fibrosis from clinical specimen.

Product Summary and Explanation

Burkholderia cepacia was discovered by Walter Burkholder in 1949 as the cause of onion skin rot.[1] It is a catalase-producing, lactose-nonfermenting, Gram-negative bacteria. *Burkholderia cepacia* is an important human pathogen which most often causes pneumonia in immunocompromised individuals with underlying lung disease (such as cystic fibrosis or chronic granulomatous disease). The organism may lead to *Burkholderia cepacia* syndrome, a neutralizing pneumonia associated with fever that culminates in to a rapid and fatal clinical deterioration.[2] *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.[3] 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.

Principles of the Procedure

Burkholderia Cepacia Agar Base contains peptone and yeast extract in the medium which provides nitrogenous, vitamin B source and other essential nutrients for bacterial growth. 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 namely ticarcillin, polymixin B and gentamycin inhibit gram-negative bacteria. Phenol Red is a pH indicator which turns pink when it reacts with alkaline byproducts generated by the bacteria when it grows. *B. cepacia* metabolizes pyruvate forming alkaline end products. These end products elevate the pH of the medium.

Formula / Liter

Ingredients	Gms / Liter
Peptone	5.00
Yeast extract	4.00
Sodium pyruvate	7.00
Potassium dihydrogen phosphate	4.40
Disodium hydrogen phosphate	1.40
Bile salts	1.50
Ammonium sulphate	1.00
Magnesium sulphate	0.20
Ammonium ferrous sulphate	0.01
Phenol red	0.02
Crystal violet	0.001
Agar	12.00
Final pH: 6.2 ± 0.2 at 25°C	
Formula may be adjusted and/or supplemented as required to meet performance specifications	

Precautions

1. For Laboratory Use only.
2. IRRITANT. Irritating to eyes, respiratory system, and skin.

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Directions

1. Suspend 18.26 grams of the medium in 500 ml of distilled water.
2. Heat to boiling to dissolve the medium completely.
3. Autoclave at 121°C, 15 psi pressure, for 15 minutes / validated cycle.
4. Cool to 50°C and aseptically add the rehydrated contents of 1 vial of Burkholderia Cepacia Selective Supplement (MS144).
5. Mix well and pour into sterile Petri plates.

Quality Control Specifications

Dehydrated Appearance	Light yellow to pink homogeneous free flowing powder
Prepared Medium	Orange coloured clear to slightly opalescent gel forms in Petri plates
Reaction of 3.65% Solution	pH : 6.2 ± 0.2 at 25°C
Gel Strength	Firm, comparable with 1.2% Agar gel

Expected Cultural Response: Cultural characteristics observed, with addition of Burkholderia Cepacia Selective Supplement (MS144), after an incubation at 35-37°C for 48-72 hours.

Sr. No.	Organisms	Results to be achieved			
		Inoculum (CFU)	Growth	Recovery	Colour of colony
1.	*Burkholderia cepacia ATCC25608*	50-100	good-luxuriant	>=50%	sage green colonies with bright pink medium
2.	*Pseudomonas aeruginosa ATCC9027*	>=10^3	inhibited	0%	

The organisms listed are the minimum that should be used for quality control testing.

Test Procedure

Refer appropriate references for standard test procedures. 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.[4, 5]

Results

Refer appropriate references and procedures for interpretation of results.

Storage

Store the sealed bottle containing the dehydrated medium at 10- 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light.

Expiration

Refer to the expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if the appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitations of the Procedure

1. 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*.[6]
2. Consult appropriate texts for detailed information and recommended procedures.

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Packaging
Product Name : Burkholderia Cepacia Agar Base
Product Code : DM1042
Available Pack sizes : 500gm

References

1. Burkholder WH (1950). "Sour skin, a bacterial rot of onion bulbs". Phytopathology 40: 115–7.
2. Whitby P. W., 1998, J. Clin. Microbiol., 36:1642 1645
3. Gilligar, Gage, Bradshaw, schidlow and Deciscco, 1985, J. Clin. Microbiol., 22:5.
4. MacDonald Gilligan, Welch, Reller and Menegus, 1994, Vol. 5:1, Cystic Fibrosis Foundation, Washington, D.C.
5. Gilligan, 1996. Clin. Microbiol. Newsl. 18:83.
6. Christensen et al, 1980, J. Clin. Microbiol., 27:270.

Further Information

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