C.L.E.D. Agar w/Bromo Thymol Blue Plate

MP792

Intended use

Recommended for isolation, enumeration and identification of urinary pathogens on the basis of lactose fermentation.

Composition**

Ingredients	g/L
Peptone	4.000
Tryptone	4.000
HM Peptone B#	3.000
Lactose	10.000
L-Cystine	0.128
Bromothymol blue	0.020
Agar	15.000
Final pH (at 25°C)	7.3±0.2

**Formula adjusted, standardized to suit performance parameters
#- Equivalent to Beef extract

Directions

1. This product is available in a stack of 5 plates.
2. Plan the experiments so as to use all plates at a time.
3. Before taking the plates into the laboratory area where sample is to be processed, follow steps as below.
4. Product has sterile plate containing the medium with three outer protective layers.
5. Place the plates in inverted position while use. Ensure that the cover lids are intact.
6. The outer sterile packaging should be removed carefully by assistant. Personnel with sterile gloves should not touch the outer surface of sterile packaging. Outer packaging must not be placed on sterile surfaces.
7. The product now has two more inner sterile layers. This inner SBS must be handled by person wearing appropriate personal protective equipment (PPE) and sterile gloves. It can be placed on sterile surfaces and opened in sterile or aseptic conditions while working.
8. Remove the second layer and innermost layer and discard.
9. Sterile plate with medium is now ready for use.
10. Aseptically open the plate for inoculation or streaking under aseptic conditions, laminar air flow (LAF) only. Open the plate by holding the lid with three fingers of one hand and base onto the palm as per standard microbiological practice.
11. On completion of inoculation or streaking close the lid carefully and place the petriplate in inverted position.
12. Incubate as desired.
13. Follow good lab practices for procedures and disposal.

Principle And Interpretation

On a solid medium, Sandys reported that swarming of Proteus species can be controlled by restricting the electrolytes (1). Formerly swarming of Proteus was controlled by adding alcohol, surface-active agent, sodium azide, boric acid etc. to the medium (1). Later on Sandy's medium was modified by Mackey and Sandys (2), by replacing mannitol by lactose and sucrose and elevating concentration of agar and bromothymol blue. This formulation was further modified by the same authors, called C.L.E.D. (Cystine-Lactose-Electrolyte-Deficient) by deleting the sucrose and by including L-cystine for promoting the growth of cystine dependent dwarf colony coliforms (3). This medium is recommended for use in urinary bacteriology, promoting the growth of all urinary pathogens. C.L.E.D. Medium is also recommended for dip stick procedures and as dip inoculum transport medium for urine specimens (2,3,4).Peptone, Tryptone and HM Peptone B provides nitrogenous and carbonaceous compounds, long chain amino acids, vitamins and other essential growth nutrients. Lactose is the fermentable sugar. L-cystine supports the growth of dwarf coliform colony. Bromo thymol blue is the pH indicator which turns yellow at acidic pH.Bacteriuria may be quantitated by inoculating the surface of an agar medium by proper dilution. Inoculate the medium immediately after urine collection. It can also be inoculated by calibrated loop or duplicate dilution pour plate methods (5,6).

Type of specimen

Clinical samples - urine

Specimen Collection and Handling:

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (7,8).
After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

In Vitro diagnostic Use only. For professional use only. Read the label before opening the pack. 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. Initiation of antibiotic therapy, before collection of sample, low urine pH (less than 5) etc. may result in low bacterial count from infected patients.
2. *Shigella* species may not grow on this medium.
3. Individual organisms differ in their growth requirement and may show variable growth patterns on the medium.
4. Each lot of the medium has been tested for the organisms specified on the COA. It is recommended to users to validate the medium for any specific microorganism other than mentioned in the COA based on the user's unique requirement.
5. It is recommended to store the plates at 24-30°C to avoid condensation.

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.

Quality Control

Appearance

Sterile C.L.E.D. Agar w/ Bromo Thymol Blue in 90 mm disposable plates with smooth surface and absence of black particles/cracks/bubbles

Colour of medium

Green coloured coloured medium

Quantity of medium

25 ml of medium in 90 mm disposable plates.

pH

7.10-7.50

Sterility Check

Passes release criteria

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Colour of Colony
Enterococcus faecalis ATCC 29212 (00087*)	50-100	good-luxuriant	>=70%	slight yellowish or greenish
Escherichia coli ATCC 25922 (00013*)	50-100	good-luxuriant	>=70%	yellow, opaque, centre slightly deeper yellow
Klebsiella pneumoniae ATCC 13883 (00097*)	50-100	good-luxuriant	>=70%	yellow to whitish blue
## *Proteus hauseri* ATCC 13315	50-100	good-luxuriant	>=70%	blue
Salmonella Typhi ATCC 6539	50-100	good-luxuriant	>=70%	bluish
Staphylococcus aureus subsp. *aureus* ATCC 25923 (00034*)	50-100	good-luxuriant	>=70%	deep yellow

Key : (*) Corresponding WDCM numbers, (##) Formerly known as *Proteus vulgaris.*

Storage and Shelf Life

On receipt store between 20-30°C. Use before expiry date on the label. 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 (7,8).

Reference

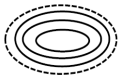
1.Sandys, 1960, J. Med. Lab. Technol., 17:224.
2.Mackey and Sandys, 1965, Br. Med. J., 2:1286.
3.Mackey and Sandys, 1966, Br. Med. J., 1:1173.
4.Dixson J. M. S. and Clark M. A., 1968, Conc. Med. Assoc. J., 99 (15).
5.Benner E. J., 1970, , Appl. Microbiol., 19(3), 409.
6.MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. I, Williams and Wilkins, Baltimore.
7.Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
8.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.

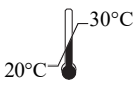
Revision :00/2024

Packaging

MP792-20PT-C.L.E.D. Agar w/ Bromothymol Blue Plate (90mmX 5Plates)
MP792-50PT-C.L.E.D. Agar w/ Bromothymol Blue Plate(90mmX 5Plates)


HiMedia Laboratories Pvt. Limited, Plot No.C-40, Road No.21Y, MIDC, Wagle Industrial Area, Thane (W) -400604, MS, India


Triple sterile barrier system with protective packaging outside



Storage temperature

EC REP CEpartner4U, Esdoornlaan 13, 3951DB Maarn, NL www.cepartner4u.eu

IVD ***In vitro* diagnostic medical device**


Do not use if package is damaged


Do not re-use


CE Marking

Contains biological material of animal origin