

C.L.E.D. Agar with Bromo Thymol Blue

Intended Use

For the enumeration, isolation and identification of urinary pathogens based on lactose fermentation.

Typical Composition (g/litre)

Peptone 4.0; Tryptone 4.0; Beef extract 3.0; Lactose 10.0; L-Cystine 0.128; Bromothymol blue 0.02; Agar 15.0

Mode of Action

This medium is recommended for use in urinary bacteriology, promoting the

growth of all urinary pathogens. Peptone, Tryptone and HM Peptone B provides nitrogenous and carbonaceous compounds, long chain amino acids, vitamins and other essential growth nutrients. It contains lactose as a reactive compound, which, when degraded to acid, causes bromothymol blue to change its colour to yellow.

Final pH (at 25°C) 7.3±0.2

Preparation

Suspend 36.15 grams in 1 liter 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. Mix well and pour into sterile Petri plates.

Final pH (at 25°C) 7.3±0.2

Storage

Store between 10-30°C in a tightly closed container and prepared medium at 20-30°C. Use before expiry date on the label.

Specimen

Clinical samples – urine

Clinical specimen collection, handling and processing, see general instructions of use

Experimental Procedure and Evaluation

Inoculate the sample on the surface of an agar medium by proper dilution. Inoculate immediately after urine collection. It can also be inoculated by calibrated loop or duplicate dilution pour plate method





Quality Control

Organism	Inoculum	Growth	Recovery	Colony colour
Enterococcus faecalis ATCC 29212	50 - 100	Good-luxuriant	>=70%	Slight yellowish or greenish
Escherichia coli ATCC 25922	50 - 100	Good-luxuriant	>=70%	Yellow, opaque, centre slightly deeper yellow
Klebsiella pneumoniae ATCC 13883	50 - 100	Good-luxuriant	>=70%	Yellow to whitish blue
Proteus vulgaris ATCC 13315	50 - 100	Good-luxuriant	>=70%	Blue
Salmonella Typhi ATCC 6539	50 - 100	Good-luxuriant	>=70%	Bluish
Staphylococcus aureus ATCC 25923	50 - 100	Good-luxuriant	>=70%	Deep yellow

Reference

- 1. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
- 2. 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
- 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)