Technical Information

CLED Agar w/Bromo Thymol Blue Plate

Product Code: PM 1792

Application: - For isolation and differentiation of urinary pathogens on the basis of lactose fermentation

Composition**

Ingredients	Gms / Litre
Peptic digest of animal tissue	4.000
Casein enzymic hydrolysate	4.000
Beef extract	3.000
Lactose	10.000
L-Cystine	0.128
Bromothymol blue	0.020
Agar	15.000
Final pH (at 25^{0}C)	7.3 ± 0.2

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

On a solid medium, Sandys reported that swarming of *Proteus* species can be checked by restricting the electrolytes [1] Which was earlier controlled by adding alcohol, surface-active agent, sodium azide, boric acid etc. to the medium [1]. Later this medium was modified by Mackey and Sandys[2], by replacing mannitol by lactose and sucrose and elevating concentration of agar and bromo thymol blue. This formulation was further modified by the same authors, and named as CLED (Cystine-Lactose-Electrolyte-Deficient) by replacing the sucrose and by including L-cystine for promoting the growth of cystine dependant dwarf colony coliforms[3]. This medium is recommended for use in urinary bacteriology, promoting the growth of all urinary pathogens. CLED Medium is also recommended for dip stick procedures and as dip inoculum transport medium for urine specimens [2-4].

Peptic digest of animal tissue, beef extract, casein enzymic hydrolysate provide 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 using proper dilution and or by calibrated loop [5-6]. Inoculate the medium immediately after urine collection. *Shigella* species may not grow on this medium. Initiation of antibiotic therapy, before collection of sample, low urine pH (less than 5) etc. may result in low bacterial count from patients with uninary tact infection.

Methodology

Ready to use sterile pour plates of CLED Agar w/Bromo Thymol Blue needs no preparation of media. The media can be used as per requirement. These plate are useful in detecting the presence of microorganism from different clinical specimens by conventional methods either streak, inoculate of surface speed of test inoculums (50-100 CFU) aseptically on the plate. Either streak, inoculate or surface spread the test inoculum (50-100 CFU) aseptically on the plate.

Quality Control

Physical Test

Appearance

Colour

Green coloured medium

Quantity of medium per Petri plate

25ml of medium

Chemical Test

pH 7.10- 7.50

Biological Test

Sterility Testing

Passes release criteria

Cultural Response

PM 1772: 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	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

Storage and Shelf Life

- Store between 15-25°C.
- Use before expiry date on the label.
- Don't freeze

Further Reading

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

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