

TM 405 - C.L.E.D. AGAR (W/ BROMO THYMOL BLUE) (BROLACIN AGAR)

INTENDED USE

For isolation and differentiation of urinary pathogens by lactose fermentation for isolation and differentiation of urinary pathogens by lactose fermentation.

PRODUCT SUMMARY AND EXPLANATION

On a solid medium, Sandys reported that swarming of *Proteus* species can be controlled by restricting the electrolytes. Formerly swarming of Proteus was controlled by adding alcohol, surface-active agent, sodium azide, boric acid etc. to the medium. Later on Sandy's medium was modified by Mackey and Sandys, 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. 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.

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.

COMPOSITION

Ingredients	Gms / Ltr
Peptone	4.000
Tryptone	4.000
Beef extract	3.000
Lactose	10.000
L-Cystine	0.128
Bromothymol blue	0.020
Agar	15.000

PRINCIPLE

Peptone, Tryptone and beef extract 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.

INSTRUCTION FOR USE

- Dissolve 36.15 grams in 1000 ml purified / distilled water.
- Heat to boiling to dissolve the medium completely.
- Sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes. Cool to 45-50°C.
- Mix well and pour into sterile Petri plates.

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder : Cream to yellow homogeneous free flowing powder.

: Green coloured, clear to slightly opalescent gel forms in Petri plates. Appearance of prepared medium

: 7.3±0.2 pH (at 25°C)

INTERPRETATION











Cultural characteristics observed after incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Colour of colony	Incubation Temperature	Incubation Period
Enterococcus faecalis	29212	50-100	Good- luxuriant	>=50%	Slight yellowish or greenish	35-37°C	18-24 Hours
Escherichia coli	25922	50-100	Good- luxuriant	>=50%	Yellow, opaque, centre slightly deeper yellow	35-37°C	18-24 Hours
Klebsiella pneumoniae	13883	50-100	Good- luxuriant	>=50%	Yellow to whitish blue	35-37°C	18-24 Hours
Proteus vulgaris	13315	50-100	Good- luxuriant	>=50%	Blue	35-37°C	18-24 Hours
Salmonella Typhi	6539	50-100	Good- luxuriant	>=50%	Bluish	35-37°C	18-24 Hours
Staphylococcus aureus subsp. aureus	25923	50-100	Good- luxuriant	>=50%	Deep yellow	35-37°C	18-24 Hours

PACKAGING:

In pack size of 100 gm and 500 gm bottles.

STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

REFERENCES

- 1. Benner E. J., 1970, , Appl. Microbiol., 19(3), 409.
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- 3. Mackey and Sandys, 1965, Br. Med. J., 2:1286.
- 4. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
- 5. Mackey and Sandys, 1966, Br. Med. J., 1:1173.
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NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices. *For Lab Use Only Revision: 08 Nov., 2019







