

CLED Agar | Ready-to-use Media



Rev: 0

Effective Date: 20/05/2024

REF FP90C1001

Intended Use:

For the isolation, enumeration, and presumptive identification of microorganisms from urine.

Principle Of The Procedure:

The nutrients in CLED Agar are supplied by peptones, pancreatic digests of gelatin and casein, and beef extract. Lactose is included to provide an energy source for organisms capable of utilizing it by a fermentative mechanism. The cystine permits the growth of dwarf colony coliforms. Bromothymol blue is used as a pH indicator to differentiate lactose fermenters from lactose-no fermenters. Organisms that ferment lactose will lower the pH and change the color of the medium from green to yellow. Electrolyte sources are reduced to restrict the swarming of *Proteus* species.

Product Summary:

In 1960, Sandys reported on the development of a new method of preventing the swarming of *Proteus* on solid media by restricting the electrolytes in the culture medium. Previous chemical methods used to inhibit swarming by *Proteus* included the addition of chloral hydrate, alcohol, sodium azide, surface-active agents, boric acid, and sulfonamides to the culture medium. This electrolyte-deficient medium of Sandys was modified by Mackey and Sandys for use in urine culture by substituting lactose and sucrose for the mannitol and increasing the concentrations of the bromothymol blue indicator and of the agar. These two investigators further modified the medium by the incorporation of cystine to enhance the growth of cystine-dependent “dwarf colony” coliforms and by deletion of sucrose. They designated the new medium as Cystine-Lactose-Electrolyte-Deficient (CLED) medium and reported it to be ideal for dip-inoculum techniques and for urinary bacteriology in general¹⁻³

Formulation* (PER LITER):

Pancreatic Digest of Gelatin	4.0g	L-Cystine	128.0mg
Pancreatic Digest of Casein	4.0g	Bromothymol Blue	0.02g
Beef Extract	3.0g	Agar	15.0g
Lactose	10.0g		

pH 7.3 +/- 0.2

*Adjust and/or supplemental as required to meet performance criteria

Procedure

Materials Provided

90mm CLED Agar.

Materials Required But Not Provided

Ancillary culture media, reagents, and laboratory equipment as required.

Test Procedure

1. Inoculate and streak the specimen as soon as possible after it is received in the laboratory with an aseptic technique.
2. It is recommended that quantitative methods be used for culturing urine specimens.
3. Incubate at $35 \pm 2^{\circ}\text{C}$ for 18-24 hours.

Results

Count the number of colonies on the plate. Multiply by an appropriate number to convert the count to CFU per mL of sample. Contaminant bacteria usually appear in low numbers which vary in colonial morphology. Urinary pathogens will usually yield high counts having uniform colonial morphology and should be sub-cultured directly to routine media for identification and susceptibility testing.

Quality Control

Inoculate representative samples with the following strains. Incubate the inoculated plates at $35 \pm 2^{\circ}\text{C}$ for 18 to 24 hrs. to allow colonies to develop on the medium.⁴

Strains	ATCC®	Growth Results
<i>Proteus mirabilis</i>	12453	Growth; colonies blue, medium blue-green to blue
<i>Escherichia coli</i>	25922	Growth; colonies yellow, medium yellow
<i>Staphylococcus aureus</i>	25923	Growth; colonies small, yellow; medium yellow
<i>Proteus vulgaris</i>	8427	Growth; colonies colorless to blue; swarming inhibited; slight spreading acceptable
Uninoculated plate	-	No growth

Storage And Shelf Life:

CLED Agar should be stored at 2 to 8°C in their original pack wrapping until before use. Avoid freezing and overheating. The plates may be inoculated up to the expiration date (see package label) and incubated for the recommended incubation times.

Warning And Precautions:

For in vitro diagnostic use. For Professional Use Only. Do Not Reuse.

Do not use plates if they show evidence of microbial contamination, discoloration, drying, cracking, or other signs of deterioration.













Limitation Of The Procedure

This medium is for laboratory use only and is not intended for the diagnosis of disease or other conditions. Identifications are presumptive and colonies should be identified using appropriate methods

Reference

1. Mackey, J.P., and G.H. Sandys. 1965. Laboratory diagnosis of infection of the urinary tract in general practice by means of a dip-inoculum transport medium. Br. Med. J. 2:1286–1288.
2. Sandys, G.H. 1960. A new method of preventing swarming of *Proteus* sp. with a description of a new medium suitable for use in routine laboratory practice. J. Med. Lab. Technol. 17:224–233.
3. Mackey, J.P., and G.H. Sandys. 1966. Diagnosis of urinary infections. Br. Med. J. 1:1173.
4. Mary Jo Zimbro, 2009, Difco™ and BBL™ Manual of Microbiological Culture Media, Second Edition, Becton, Dickinson and Company, Sparks, MD.

Packaging Symbol

Symbol	Definition
	Catalogue number
	In Vitro Diagnostic Medical Device
	Batch code
	Date of manufacture
	Temperature limit
	Use-by date
	Keep away from sunlight
	Do not re-use
	Fragile, handle with care
	Consult instructions for use or consult electronic instructions for use
	Do not use if packaging damaged and consult instructions for use
	Manufacturer

Further Information:

For further information please contact your Biomed MDx representative.



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