Cystine Lactose Electrolyte Deficient (CLED) Agar (NCM0034)

Revision: 0 Effective Date: 5/11/2023

Intended Use
Cystine Lactose Electrolyte Deficient (CLED) Agar is used for the differentiation and enumeration of microorganisms in urine.

Description
A medium for urine culture where the absence of electrolytes inhibits the swarming of *Proteus* spp. Cystine is added for the benefit of those organisms which have a specific cystine requirement. Differentiation of lactose and non-lactose fermenters is achieved using bromothymol blue as pH indicator. This medium supports the growth of *Streptococcus pyogenes* and most other fastidious organisms that do not require blood.

Typical Formulation

Balanced Peptone	4.0 g/L
Beef Extract	3.0 g/L
Tryptone	4.0 g/L
Lactose	10.0 g/L
L-Cystine	0.128 g/L
Bromothymol Blue	0.02 g/L
Agar	15.0 g/L

pH: 7.3 ± 0.2 at 25°C
Formula may be adjusted and/or supplemented as required to meet performance specifications.

Precaution
Refer to SDS

Preparation
1. Suspend 36 grams of the medium in one liter of purified water.
2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
3. Autoclave at 121°C for 15 minutes.
4. Cool to 45-50°C.

Test Procedure
For a complete discussion on collection and processing of urine cultures refer to appropriate references.

Quality Control Specifications
Dehydrated Appearance: Powder is homogeneous, free flowing, and light beige.

Prepared Appearance: Prepared medium is clear and blue-green.

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Expected Cultural Response: Cultural response on CLED Agar at the appropriate atmosphere and temperature and examined for growth at 18 – 24 hours incubation.

MICROORGANISM	ATCC	APPROX. INOCULUM (CFU)	EXPECTED RESULTS		ACTUAL RESULTS	
			Growth	Reaction	Growth	Reaction
Escherichia coli	25922	50-200	≥70%	Yellow colonies	Meets Expected Result	
Enterococcus faecalis	29212	50-200	≥70%	Yellow colonies	Meets Expected Result	
Proteus mirabilis	12453	4 Quad Streak	Growth	Blue-green colonies; partial to inhibited swarming	Meets Expected Result	
Proteus mirabilis	29906	4 Quad Streak	Growth	Blue-green colonies; Swarming inhibited	Meets Expected Result	
Salmonella typhimurium	14028	4 Quad Streak	Growth	Blue-green colonies	Meets Expected Result	
Staphylococcus aureus	25923	50-200	>70%	Yellow colonies	Meets Expected Result	

The organisms listed are the minimum that should be used for quality control testing.

Results
Refer to appropriate references for results.

Expiration
Refer to expiration date stamped on container. The dehydrated medium should be discarded if not free flowing, or if appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitation of the Procedure
Due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.

Storage
Store dehydrated culture media at 2-30°C away from direct sunlight. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping container tightly closed.

References
1. Mackey, J.P. and Sandys, G.H. (1966). Diagnosis of urinary infections. Brit.Med.J. 1: 1173.
2. Guttman, D and Naylor, G.R.E. (1967). Dip-slide: an aid to quantitative urine culture in general practice. Brit.Med. J. 3: 343-345.

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