CLED AGAR

INTENDED USE
Remel CLED Agar is a solid medium recommended for use in qualitative procedures for the cultivation of microorganisms from urine specimens.

SUMMARY AND EXPLANATION
In 1960, Sandys developed a solid culture medium that prevented the swarming of *Proteus* by restricting electrolytes.[1] Mackey and Sandys modified the medium for use with urine cultures by substituting lactose and sucrose for mannitol, and increasing the concentration of brom thymol blue and agar.[2] Further modification by addition of cystine and deletion of sucrose resulted in Cystine Lactose Electrolyte Deficient (CLED) agar which is recommended for detection of bacteriuria by quantitative culture of urine.[3]

PRINCIPLE
Casein peptone supplies amino acids, nitrogenous compounds, and peptides essential for the growth of bacteria. Beef extract supplies vitamins and carbohydrates. Lactose provides a carbohydrate source of energy. Cystine enhances the growth of cystine-dependent coliforms. Brom thymol blue is a pH indicator which differentiates lactose fermenters (yellow) from nonfermenters. Electrolytes are reduced in order to restrict the swarming of *Proteus.* Bacteria may be quantitated by inoculating the surface of the medium with appropriate dilutions of the urine sample.

REAGENTS (CLASSICAL FORMULA)*

Lactose	10.0 g	L-Cystine	0.128 g
Casein Peptone	4.0 g	Brom Thymol Blue	0.02 g
Gelatin	4.0 g	Agar	15.0 g
Beef Extract	3.0 g	Demineralized Water	1000.0 ml

pH 7.3 ± 0.2 @ 25°C

*Adjusted as required to meet performance standards.

PROCEDURE
1. Inoculate and streak the specimen as soon as possible after it is received in the laboratory.
2. The use of a calibrated loop is recommended to facilitate counting colonies and estimate the colony forming units (CFU)/ml.
3. Incubate plates in ambient air at 33-37°C for 18-24 hours.
4. Determine colony count and observe colonial characteristics and morphology.
5. Proceed with identification of the organisms isolated following established laboratory procedures.

QUALITY CONTROL
All lot numbers of CLED Agar have been tested using the following quality control organisms and have been found to be acceptable. Testing of control organisms should be performed in accordance with established laboratory quality control procedures. If aberrant quality control results are noted, patient results should not be reported.

CONTROL	INCUBATION	RESULTS
Escherichia coli ATCC® 25922	Ambient, 18-24 h @ 33-37°C	Yellow opaque colonies
Proteus vulgaris ATCC® 8427	Ambient, 18-24 h @ 33-37°C	Blue colonies with inhibited swarming
Staphylococcus aureus ATCC® 25923	Ambient, 18-24 h @ 33-37°C	Deep yellow colonies

LIMITATIONS
1. CLED Agar is nonselective, but due to the lack of electrolytes *Shigella* spp. usually do not grow.[4]

BIBLIOGRAPHY

1. Sandys, G.H. 1960. J. Med. Lab. Technol. 17:224-233.
2. Mackey, J.P. and G.H. Sandys. 1965. Br. Med. J. 2:1286-1288.
3. Mackey, J.P. and G.H. Sandys. 1966. Br. Med. J. 1:1173.
4. MacFaddin, J.F. 1985. Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria. Vol. 1. Williams & Wilkins, Baltimore, MD.

Refer to the front of Remel *Technical Manual of Microbiological Media* for **General Information** regarding precautions, product storage and deterioration, specimen collection, storage and transportation, materials required, quality control, and limitations.

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