

## Cled agar with Andrade Indicator

REF.	Pack size
609 01 100	100 gm
609 01 500	500 gm

### Intended Use

CLED Agar with Andrade indicator is a differential culture medium intended for use in isolation, enumeration and identification of bacteria such as *E. coli* in sterile urine specimens.

### Background

CLED Agar is an abbreviation for Cystine Lactose-Electrolyte-Deficient Agar. Sandys developed an electrolyte-deficient medium that prevented *Proteus* sp. from swarming. This medium was modified by substituting lactose and sucrose for mannitol, and increasing the amount of indicator and agar. CLED agar formula was further modified by the incorporation of cystine and omitting of sucrose; which made it ideal for dip inoculation techniques. This medium supports the growth of urinary pathogens and provides distinct colony morphology.

### Principle

C.L.E.D. Agar medium Andrade indicator (Acid Fuchsin in 1N Sodium Hydroxide) is incorporated. The essential growth nutrients are supplied by peptone, tryptone and beef extract. Lactose is the carbohydrate source. L-cystine permits the growth of "dwarf colony" coliforms. Addition of Andrade indicator enhances the appearance of colony and aids in the identification of microorganisms. At different pH values, the colour of the medium varies from the standard medium.

Components	gm/Liter
Beef Extract	3.0
Peptone	4.0
Tryptone	4.0
Bromothymol Blue	0.02
L-Cysteine	0.128
Agar	15.0
Lactose	10.0
Andrade Indicator	0.10

Final pH (at 25°C) 7.5 ± 0.2

### Preparation, Storage and Stability

Store the dehydrated medium at 10-30°C and use before the expiry date on the label. Store the prepared medium at 2-8°C. After the desired amount of medium is taken out, replace the cap tightly to protect from hydration.

### Procedure

1. Suspend 36.25 g of the powder in 1 L distilled water and mix well. 2. Boil with frequent agitation to dissolve the powder completely. 3. Sterilize by autoclaving at 121°C for 15 minutes.

### Quality Control Appearance

1-Dehydrated Appearance : Yellow coloured, homogeneous, free flowing powder.

2- Prepared Appearance : Green coloured, slightly opalescent gel.

3- Cultural Response : Cultural characteristics after 18-24 hours at 30-35°C (As per pharmacopeia or 35± 2°C for clinical specimens)

Organisms (ATCC)	Growth	Colour of the Colony
<i>Escherichiacoli</i> halo	Good	Bright pink with pink
<i>Staphylococcus aureus</i>	Good	Golden Yellow
<i>Enterococcus faecalis</i> orange	Good	Yellow, green or
<i>Proteus mirabilis</i>	Good	Blue-green
<i>Streptococcus pyogenes</i>	Good	Greyish green
<i>Klebsiella aerogenes</i>	Good	Greyish green

### Interpretation of the results

- Count the number of colonies on the dipstick. Multiply by dilution factor to convert the count to CFU per ml of the sample.
- Contaminant bacteria usually appear in low numbers and vary in colony morphology

### Precautions

- Factors that may cause urine counts from infected patients to be low include: rapid rate of urine flow, prior initiation of antimicrobial therapy, a urine pH of less than 5 and a specific gravity of less than 1.003.
- The medium should not be incubated for more than 24 hours since, if lactose fermenters predominate, the whole medium may turn pink, masking the presence of non-lactose fermenters
- Shigella* species may not grow on this medium.

### Bibliography

- Sandys, 1960, J. Med. Lab. Technol., 17:224.
- Mackey and Sandys, 1965, Br. Med. J., 2:1286.
- MacKey and Sandys, 1966, Br. Med. J., 1:1173.
- Dixon J. M. S. and Clark M. A., 1968, Conc. Med. Assoc. J., 99 (15).