C.L.E.D. Agar with Andrade Indicator

Intended Use

C.L.E.D. Agar with Andrade Indicator is used for isolation, enumeration and presumptive identification of microorganisms from urine, giving good colonial differentiation.

Summary

Sandy's observed that restricting the electrolytes on a solid medium might prevent the swarming of *Proteus*. Previous chemical methods used to inhibit swarming of *Proteus* included the addition of chloral hydrate, alcohol, sodium azide, surface-active agents, boric acid and sulphonamides to the culture medium. This electrolyte-deficient medium was modified for use in urine culture by substituting lactose and sucrose instead of mannitol and increasing the concentrations of bromothymol blue indicator and agar. The medium was further modified by the incorporation of cystine in order to enhance the growth of cystine dependent "dwarf colony" coliforms and by the deletion of sucrose. This new medium, Cystine-Lactose-Electrolyte-Deficient (C.L.E.D.) Agar is ideal for dip inoculation techniques and for urinary bacteriology in general.

Principle

C.L.E.D. Agar with Andrade indicator is similar to C.L.E.D. Agar with Bromothymol blue except in this medium Andrade indicator (Acid Fuchsin in 1N Sodium Hydroxide) is incorporated. The essential 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 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. For better results, the medium should not be incubated for more than 24 hours because if lactose fermenters predominate, the entire medium may turn pink masking the presence of non-lactose fermenters. Inoculate the medium immediately after urine collection. *Shigella* species may not grow on this medium. Prior initiation of antibiotic therapy, low urine pH (less than 5) etc. may result in low urine count from infected patients.

Formula*

Ingredients	**g/L**
Lactose	10.0
Tryptone	4.0
Peptone	4.0
Beef Extract	3.0
L-Cystine	0.128
Andrade Indicator	0.10
Bromothymol Blue	0.02
Agar	15.0
Final pH (at 25°C)	7.5 ± 0.2

*Adjusted to suit performance parameters.

Directions

1. Loosen the cap.
2. Melt the medium completely in a water bath at 100°C. Do not remove the cap of the bottle while melting.
3. Cool to 45°C-50°C, mix well and pour into presterile petriplates.

Quality Control

Appearance: Greenish blue coloured, clear gel without any bubbles.

Appearance and Gelling of Poured Plate: Greenish blue coloured, slightly opalescent with firm gel forms in petriplates.
Cultural Response: Cultural characteristics observed after an incubation of 18-24 hours at 30°C-35°C.

Organism (ATCC)	Growth	Colour of Colony
Proteus mirabilis (25933)	Good	Blue-green transluscent colonies
Escherichia coli (8739)	Good	Pink colonies with pink medium
Enterococcus faecalis (29212)	Good	Green colonies
Pseudomonas aeruginosa (27853)	Good	Colourless colonies
Staphylococcus aureus (25923)	Good	Golden yellow colonies with pink medium
Klebsiella aerogenes (13048)	Good	Greyish green colonies
Streptococcus pyogenes (19615)	Good	Greyish green colonies

Note: Inoculum cfu for good growth is 10-100.

Remarks

1. Do not use media bottles that exhibit any damage, cracks, microbial contamination, discoloration, drying or other sign of deterioration.
2. Ensure that the temperature of water bath is at 100°C so that the medium melts completely. Cooler water baths give rise to lumpy, uneven medium.
3. Before pouring into sterile petriplates, gently swirl the bottle to check whether the entire contents are properly mixed and melted.
4. Good laboratory practices and hazard precautions must be observed at all times.
5. After use media containers, prepared plates, sample, sample containers and other contaminated materials must be sterilized or incinerated before discarding.

Storage and Stability

1. Store the ready to use C.L.E.D. Agar with Andrade Indicator at 15°C-25°C in a cool, dry place away from light.
2. Stability of the kit is as per expiry date mentioned on the label.

Limitations

1. 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.
2. 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.
3. *Shigella* species may not grow on this medium.

Warranty

This product is designed to perform as described on the label and package insert. The manufacturer disclaims any implied warranty of use and sale for any other purpose.

References

1. Bevis, T.D. (1968). A modified electrolyte- deficient culture medium. J.Med. Lab. Tech., 25: 38-41.
2. Mackey, J.P. and Sandys, G.H. (1966). Diagnosis of urinary infections, Brit. Med. J., 1: 1173.
3. Sandys, G.H. (1960). A new medium for preventing swarming of *Proteus* spp. with a description of a new medium suitable for use in routine laboratory practice. J. Med. Lab. Tech., 17: 224-233.
4. Data on file: Microxpress®, A Division of Tulip Diagnostics (P) Ltd.

Product Presentation:

Cat. No.	Product Description	Pack Size
203030270100	Bottle Media	100 mL

Disclaimer

Information provided is based on our inhouse technical data on file, it is recommended that user should validate at his end for suitable use of the product.