



CAL Agar (Cellobiose Arginine Lysine Agar)

M893

CAL (Cellobiose Arginine Lysine) Agar is used for selective isolation and biochemical characterization of *Yersinia enterocolitica*.

Composition**

| Ingredients | Gms / Litre |
|------------------------|-------------|
| Yeast extract | 3.000 |
| Sodium chloride | 5.000 |
| Cellobiose | 3.500 |
| L-Arginine | 6.500 |
| L-Lysine hydrochloride | 6.500 |
| Sodium deoxycholate | 1.500 |
| Neutral red | 0.030 |
| Agar | 20.000 |
| Final pH (at 25°C) | 7.1±0.2 |

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 46.03 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. DO NOT OVERHEAT OR AUTOCLAVE. Mix well and pour into sterile Petri plates.

Principle And Interpretation

Yersinia enterocolitica is a significant invasive enteric pathogen belonging to the family *Enterobacteriaceae*, which causes several well-recognized diseases especially in younger persons and several uncommon post-infection syndromes (1). Enterocolitis caused by *Y. enterocolitica* is characterized by diarrhoea, low fever and abdominal pain. CAL Agar used for selective isolation of *Y. enterocolitica* was originally formulated by Dudley and Shotts (2). CAL Agar is a differential medium as it differentiates *Yersinia* on the basis of cellobiose fermentation and lysine or arginine decarboxylation. CAL Agar is generally used for the isolation and characterization of *Y. enterocolitica* from faecal specimens (2) as the organism is biochemically similar to other *Enterobacteriaceae* (3). CAL Broth is used for the enumeration of *Y. enterocolitica* from water and other liquid specimens (3).

Yeast extract provides essential nutrients to the organisms. Cellobiose is the fermentable carbohydrate. Sodium chloride maintains the osmotic equilibrium. Sodium deoxycholate makes the medium selective by inhibiting the accompanying gram-positive bacteria, which may cause contamination during cultivation. L-arginine and L-lysine are the amino acids, decarboxylation of which makes the medium differential. Neutral red is the indicator, which turns red under acidic conditions.

Quality Control

Appearance

Light yellow to pink homogeneous free flowing powder

Gelling

Firm, comparable with 2.0% Agar gel.

Colour and Clarity of prepared medium

Red coloured, clear to slightly opalescent gel forms in Petri plates.

Reaction

Reaction of 4.6% w/v aqueous solution at 25°C. pH : 7.1±0.2

pH

6.90-7.30

Cultural Response

M893: Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours.

| Organism | Inoculum (CFU) | Growth | Cellobiose | Arginine Decarboxylation | Lysine Decarboxylation |
|-------------------------------------------|----------------|----------------|-------------------|--------------------------|------------------------|
| <i>Escherichia coli</i> ATCC 25922 | 50-100 | good | negative reaction | variable reaction | variable reaction |
| <i>Proteus mirabilis</i> ATCC 25933 | 50-100 | good | negative reaction | negative reaction | negative reaction |
| <i>Pseudomonas aeruginosa</i> ATCC 27853 | 50-100 | good | negative reaction | negative reaction | positive reaction |
| <i>Yersinia enterocolitica</i> ATCC 27729 | 50-100 | good-luxuriant | positive reaction | negative reaction | negative reaction |

Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label.

Reference

1. Cover T.L., and Aber R.C., 1989 *Yersinia Enterocolitica*, N. Engl. J. Med., 32:16-24
2. Dudley M.V. and Shotts E.B., 1979, J. Clin. Microbiol., 10(2):180.
2. MacFaddin J.F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore.

Revision : 2 / 2015

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