Simmons Citrate Agar

M099S

Simmons Citrate Agar is used for differentiation between faecal coli and members of the aerogenes group on the basis of citrate utilization.

Composition**

Ingredients	Gms / Litre
Ammonium dihydrogen phosphate	1.000
Magnesium sulphate	0.200
Dipotassium phosphate	1.000
Sodium citrate	2.000
Sodium chloride	5.000
Bromo thymol blue	0.080
Agar	15.000
Final pH (at 25°C)	6.8±0.1

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 24.28 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Mix well and distribute in tubes. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool the tubes as slants.

Principle And Interpretation

These media are used for the differentiation between *Enterobacteriaceae* and the members of aerogenes group on the basis of citrate utilization as sole carbon source. Initially the citrate medium was developed by Koser (1) containing ammonium salt as the only nitrogen source and citrate as the only carbon source for differentiating *Escherichia coli* and *Enterobacter aerogenes* by IMViC tests. Later on Simmons (2) modified Kosers formulation by adding agar and bromothymol blue (3). It is recommended by APHA (4).It is also recommended by BIS (5) for isolation of *Escherichia coli*

Ammonium dihydrogen phosphate and sodium citrate serves as the sole nitrogen and carbon source respectively. Bromo thymol blue is the pH indicator. The organisms produce an alkaline reaction which is indicated by the change in colour from green to blue.

Quality Control

Appearance
Yellow coloured homogeneous free flowing powder

Gelling
Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium
Forest green coloured slightly opalescent gel forms as slants.

Reaction
Reaction of 2.43% w/v aqueous solution at 25°C. pH : 6.8±0.1

pH
6.70-6.90

Cultural Response
Cultural characteristics observed after an incubation at 35-37°C for 18 - 24 hours.

Cultural Response

Organism	Inoculum (CFU)	Growth	Citrate Utilisation

Cultural Response

Enterobacter aerogenes ATCC 13048	50-100	good-luxuriant	Positive reaction, blue colour
Escherichia coli ATCC 25922	50-100	inhibited	
Salmonella Enteritidis ATCC 13076	50-100	good-luxuriant	Positive reaction, blue colour
Salmonella Typhimurium ATCC 14028	50-100	good-luxuriant	Positive reaction, blue colour
Salmonella Typhi ATCC 6539	50-100	fair to good	Negative reaction, green colour
Shigella dysenteriae ATCC 13313	50-100	inhibited	Negative reaction, green colour

Storage and Shelf Life

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

Reference

1. Koser, 1923, J. Bact., 8:493.
2. Simmons, 1926, J. Infect. Dis., 39:209.
3. MacFaddin J., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore.
4. American Public Health Association, 1981, Standard Methods for the Examination of Water and Wastewater, 15th ed., APHA Inc., Washington,D.C.
5. Bureau of Indian Standards, IS:5887 (Part II) 1976, reaffirmed 1986

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