

CM 22156 - SIMMONS CITRATE AGAR (IS : 5887 (Part I) 1976, reaffirmed 2005)

INTENDED USE

For differentiation between faecal coliforms and members of the aerogenes group on the basis of citrate utilization.

PRODUCT SUMMARY AND EXPLANATION

Simmons Citrate Agar is a synthetic test agar proposed by Simmons (1926) for the identification of microorganisms (particularly of enteric Gram-negative bacilli from clinical specimens) on the basis of their metabolism of citrate, being the sole carbohydrate source. The media is 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 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 modified Koser's formulation by adding agar and bromothymol blue. It is recommended by APHA. It is also recommended by BIS for isolation of Escherichia coli.

COMPOSITION

Ingredients	Gms / Ltr
Agar	15.000
Sodium chloride	5.000
Sodium citrate	2.000
Ammonium dihydrogen phosphate	1.000
Dipotassium phosphate	1.000
Magnesium sulphate	0.200
Bromo thymol blue	0.080

PRINCIPLE

The medium contains Ammonium dihydrogen phosphate and sodium citrate which serves as the sole nitrogen and carbon source respectively. Bromothymol blue is the pH indicator. Dipotassium hydrogen phosphate acts as a buffer. Sodium chloride maintains the osmotic balance. Metabolism of citrate leads to alkalization of the medium, which is indicated by a change in the color of the pH indicator bromothymol blue from green to deep blue. Magnesium sulfate is a cofactor for a variety of metabolic reactions.

INSTRUCTION FOR USE

- Dissolve 24.28 grams in 1000 ml distilled water.
- Gently heat to boiling with gentle swirling and dissolve the medium completely.
- Mix well and distribute in tubes.
- Sterilize by autoclaving at 15 psi (121°C) for 15 minutes.
- Cool the tube as slants.

QUALITY CONTROL SPECIFICATIONS

Appearance of Dehydrated powder	:	Yellow colour, homogeneous free flowing powder.
Appearance of Prepared medium- Basal medium	:	Forrest green colored, slightly opalescent gel .
pH (at 25°C)	:	6.8± 0.2



INTERPRETATION

Cultural characteristics observed after an incubation at 35 ± 2°C for 18-24 hours for bacteria. Recovery rate is considered 100% for bacteria growth on Soya Agar.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Colour of colony	Incubation Temperature	Incubation Period
Klebsiella aerogenes	13048	50-100	Luxuriant	>=50%	Positive, blue colour	35 ± 2°C	18-24 hours
Salmonella enteritidis	13076	50-100	Luxuriant	>=50%	Positive, blue colour	35 ± 2°C	18-24 hours
Salmonella typhimurium	14028	50-100	Luxuriant	>=50%	Positive, blue colour	35 ± 2°C	18-24 hours
Salmonella Typhi			Fair to Good		Negative, green colour		
Escherichia coli	6539	50-100	Fair to Good	30-40%	-	35 ± 2°C	18-24 hours
Shigella dysenteriae	25922	≥1000	Inhibited	0%	-	35 ± 2°C	18-24 hours
	13313	≥1000	Inhibited	0%		35 ± 2°C	18-24 hours

PACKAGING:

In 100gm & 500gm packaging size.

STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers below 25°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out, replace the cap tightly to protect from hydration.

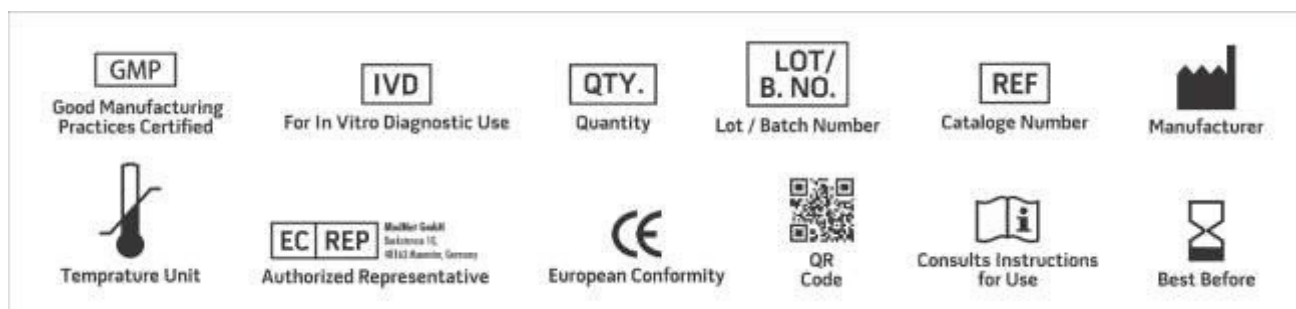
Product Deterioration: Do not use powder if they show evidence of microbial contamination, discoloration, drying, or other signs of deterioration.

DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

REFERENCES

1. Koser, 1923, J. Bact., 8:493.
2. Simmons, J.S. (1926). A culture medium for differentiating organisms of typhoid-colon aerogenes groups and for isolating of certain fungi. J. Infect. Dis. 39: 209-241.
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.



NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.

