



## Lactose Broth

M1003S

Recommended for the detection of coliform bacteria in water, food, dairy products. Recommended by BIS Committee under the specifications IS:5401(1)-2012 & ISO 4832:2006.

### Composition\*\*

Ingredients	g / L
Peptone	5.000
HM Peptone B#	3.000
Lactose	5.000
Final pH ( at 25°C)	6.9±0.1

\*\*Formula adjusted, standardized to suit performance parameters

# Equivalent to Beef extract

### Directions

Suspend 13 grams in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely. For larger inocula (10 ml or more, concentrated medium may be prepared to account for medium dilution by the inoculum. Dispense in 5 ml amounts in tubes containing inverted fermentation vial (Durhams tube as desired. Sterilize by autoclaving at 15 lbs pressure (121°C for 15 minutes.

Note : If desired, for detecting acid production, 3 ml of an aqueous solution of bromocresol purple (prepared by grinding 0.5 g of bromocresol purple in 100 ml of 0.01 N sodium hydroxide until dissolved may be added to one litre of the medium.

### Principle And Interpretation

Lactose Broth is recommended by APHA in the performance and confirmation of the presumptive test for coliform bacteria in water (1), food (2) and milk (3,4). Present formulation is recommended by BIS for detection and estimation of coliform bacteria in food stuff as a confirmatory medium (5). As per BIS (5) the discrete colonies obtained from EMB Agar Plates (M022S are inoculated in Lactose Broth (M1003S. Formation of gas in the lactose tubes indicates presence of coliforms. This medium can be used as an alternate to Lauryl Sulphate Broth in the presumptive test of the MPN of standard coliforms.

Peptone and HM peptone B supply carbon, nitrogen compounds, long chain amino acids, vitamins and other essential nutrients to the organisms. Lactose is a fermentable carbohydrate for the coliforms. Tubes of Lactose Broth are inoculated with dilutions of water or milk, etc. under test, and incubated at 35°C and examined for gas formation after 24 and 48 hours. Members of the coliform group are defined as aerobic and facultative anaerobic gram-negative and non-sporing bacilli which ferment lactose with gas formation within 48 hours at 35°C. In testing dairy products, Lactose Broth is used only in the completed test (6). Large water samples may require double strength Lactose Broth to minimize the final volume.

### Type of specimen

Food and dairy samples ; Water samples

### Specimen Collection and Handling:

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (2,3,4).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards(1). After use, contaminated materials must be sterilized by autoclaving before discarding.

### Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

### Limitations

1. Further biochemical and serological testing is required for complete identification.

## Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

## Quality Control

### Appearance

Yellow coloured homogeneous free flowing powder

### Colour and Clarity of prepared medium

Light to medium amber coloured clear solution without any precipitate.

### Reaction

Reaction of 1.3% w/v aqueous solution at 25°C. pH : 6.9±0.1

### pH

6.80-7.00

### Cultural Response

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

Organism	Inoculum (CFU)	Growth	Gas
# <i>Klebsiella aerogenes</i> ATCC 13048 (00175*)	50-100	luxuriant	Positive reaction
<i>Enterococcus faecalis</i> ATCC 29212 (00087*)	50-100	luxuriant	Negative reaction
<i>Escherichia coli</i> ATCC 25922 (00013*)	50-100	luxuriant	Positive reaction
<i>Pseudomonas aeruginosa</i> ATCC 10145 (00024*)	50-100	luxuriant	Negative reaction

Key : (\*) Corresponding WDCM numbers

(#) Formerly known as *Enterobacter aerogenes*.

## Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

## Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (6,7).

## Reference

- Lipps WC, Braun-Howland EB, Baxter TE, eds. Standard methods for the Examination of Water and Wastewater, 24th ed. Washington DC:APHA Press; 2023
- Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
- American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D. C.
- Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
- Bureau of Indian Standards, IS : 5401 - 1969, 2012 (Second reprint - June 1990).
- Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
- Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

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