



Lactose Broth

M1003

Intended use

Lactose Broth is used for the detection of coliform bacteria in water, foods, dairy products and clinical samples.

Composition**

Ingredients	Gms / Litre
Peptone	5.000
HM Peptone B [#]	3.000
Lactose	5.000
Final pH (at 25°C)	6.9±0.2

**Formula adjusted, standardized to suit performance parameters

[#] - Equivalent to Beef extract

Directions

Suspend 13.0 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 tubes containing inverted fermentation vial (Durhams tube) as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Principle And Interpretation

Examination of water, foods, ingredients and raw materials, for the presence of marker groups such as coliforms is one of the most common tests in a microbiology laboratory, partly because of the relative ease and speed with which these tests can be accomplished. Where it is claimed that drinking water has been processed for safety, the finding of such organism demonstrates a failure of the process. It is a valuable bacterial indicator for determining the extent of fecal contamination of recreational surface waters or drinking water (1).

Lactose Broth is recommended by APHA in the performance and confirmation of the presumptive test for coliform bacteria in water (2), food (3) and milk (4). This medium was initially listed as an alternative to Lauryl Sulfate Broth in the presumptive Standard Total Coliform Multiple-Tube (MPN) Test for water analysis. Although it is not the original formulation, Lactose Broth provides excellent results in Eijkman Assays of gas production at 45°C, which is a characteristic of *Escherichia coli*. While preparing this medium it is important to avoid overheating and to distribute it into tubes before sterilization.

Peptone and HM Peptone B in the medium supply nitrogenous and carbonaceous compounds, long chain amino acids 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-spore forming 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 (3). Large water samples may require double strength Lactose Broth to minimize the final volume.

Type of specimen

Clinical samples, 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 (3,4).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards.(2)

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 :

Further biochemical tests must be carried out for confirmation.

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

Cream to yellow 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±0.2

pH

6.70-7.10

Cultural Response

M1003: Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours.

Organism	Inoculum (CFU)	Growth	Gas
Cultural Response			
# <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 27853 (00025*)	50-100	luxuriant	negative reaction
<i>Pseudomonas aeruginosa</i> ATCC 9027 (00026*)	50-100	luxuriant	
<i>Escherichia coli</i> ATCC 8739 (00012*)	50-100	luxuriant	positive reaction
<i>Escherichia coli</i> NCTC 9002	50-100	luxuriant	positive reaction

Key : * Corresponding WDCM numbers

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 (5,6).

Reference

1. Corry J. E. L., Curtis G. D. W., and Baird R. M., Culture Media for Food Microbiology, Vol. 34, Progress in Industrial Microbiology, 1995, Elsevier, Amsterdam
2. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.
3. 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.
4. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
6. 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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