



MacConkey Broth w/ Neutral Red

M007

Intended use

Recommended for the selective enrichment and enumeration of coliforms.

Composition**

Ingredients	Gms / Litre
Peptone	20.000
Lactose	10.000
Bile salts	5.000
Sodium chloride	5.000
Neutral red	0.075
Final pH (at 25°C)	7.4±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 40.07 grams in 1000 ml purified/ distilled water. Heat if necessary to dissolve the medium completely. Distribute into tubes with inverted Durhams tubes. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool the tubes before inoculation.

Principle And Interpretation

MacConkey Broth is widely used as a differential medium for detection and enumeration of coliforms from a wide variety of clinical, food and water samples. Identification is based on colour change of the medium due to the indicator neutral red used (5, 6).

Peptone provides necessary nitrogen source. Lactose serves as the fermentable carbohydrate source. Sodium chloride maintains the osmotic balance of the cells. The selective action of these media is attributed to the presence of bile salts, which are inhibitory to most species of gram-positive bacteria. Gram-negative bacteria usually grow well on these media and are differentiated by their ability to ferment lactose. The colour change of the medium shown by lactose-fermenters is due to production of acid from lactose and a subsequent colour change of the indicator dye when the pH of the media falls below 6.8. Lactose non-fermenting strains, such as *Shigella* and *Salmonella* do not alter the appearance of the medium. The medium turns pink in case of lactose fermenters and yellow in case of non-lactose-fermenters, due to neutral red. MacConkey Broth, which contains neutral red as an indicator is considered as a standard medium for the primary isolation as well as presumptive identification of coliform-aerogenes group of organisms in food and water.

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 (1,7,8). 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 :

1. Some strains may show poor growth due to strain variation.

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

Pale yellow to pink homogeneous free flowing powder

Colour and Clarity of prepared medium

Red coloured clear solution without any precipitate

Reaction

Reaction of 4.01% w/v aqueous solution at 25°C. pH : 7.4±0.2

pH

7.20-7.60

Cultural Response

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

Organism	Inoculum (CFU)	Growth	Acid	Gas
# <i>Klebsiella aerogenes</i> ATCC 13048 (00175*)	50-100	luxuriant	positive reaction	positive reaction
<i>Escherichia coli</i> ATCC 25922 (00013*)	50-100	luxuriant	positive reaction	positive reaction
<i>Klebsiella pneumoniae</i> ATCC 13883 (00097*)	50-100	luxuriant	positive reaction	positive reaction
<i>Proteus mirabilis</i> ATCC 25933	50-100	luxuriant	negative reaction	negative reaction
<i>Salmonella Choleraesuis</i> ATCC 12011	50-100	fair to good	negative reaction	negative reaction
<i>Staphylococcus aureus</i> subsp. <i>aureus</i> ATCC 25923 (00034*)	>=10 ⁴	inhibited		
<i>Enterococcus faecalis</i> ATCC 29212	50-100	none-poor	positive reaction	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 (3,4).

Reference

1. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D.C.
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. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
4. 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.
5. MacConkey A. T., 1900, The Lancet, ii: 20.
6. MacConkey A. T., 1905, J. Hyg. 5: 333.
7. 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.
8. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.

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