



M-Lauryl Sulphate Broth

Intended Use:

Recommended for enumeration of Escherichia coli in water using membrane filtration technique.

Composition**

Ingredients	Gms / Litre
Peptone	39.000
Yeast extract	6.000
Lactose	30.000
Phenol red	0.200
Sodium lauryl sulphate (SLS)	1.000
Final pH (at 25°C)	7.4±0.2
**Formula adjusted, standardized to suit performance parameters	

Directions

Suspend 76.2 grams in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense as desired and sterilize by steaming for 30 minutes on three consecutive days or by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C.

Principle And Interpretation

The membrane filter technique is used to test relatively large volumes of samples. It is extremely useful in monitoring drinking water and a variety of natural waters (5). The earlier medium used to detect coliforms in water employed bile salts as the selective agent. This was replaced with Teepol by Burman (2). The effectiveness of teepol was demonstrated earlier (8,9). M-Lauryl Sulphate Broth is similar to this medium, the only difference being the use of sodium lauryl sulphate as the inhibitory agent instead of teepol. M-Lauryl Sulphate Broth is recommended for enumeration of *Escherichia coli* and coliforms using membrane filtration technique (4,10).

An absorbent pad is saturated with M-Lauryl Sulphate Broth. The filter, through which the water sample is passed, is aseptically

placed on this saturated absorbent pad, with face upwards.

Burman (3) recommended the following incubation temperatures and durations.

Unchlorinated waters:

Coliform organisms : 4 hours at 30°C followed by 14 hours at 35°C

Escherichia coli : 4 hours at 30°C followed by 14 hours at 44°C

Non-chlorinated organisms benefit from 4 hours incubation at 30°C but chlorinated organisms require 6 hours incubation at 25°C. After incubation, yellow colonies are formed which should be confirmed further.

Peptone and yeast extract act as a source of nitrogen, carbon and amino acids. Lactose is the source of fermentable carbohydrate. Phenol red serves as an indicator. Sodium lauryl sulphate inhibits gram-positive bacteria.

Type of specimen

Water samples

Specimen Collection and Handling:

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 protectionFollow 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

Please refer disclaimer Overleaf.

1. Due to varying nutritional requirements, some strains may be encountered that grow poorly.

2. If the inoculum is too heavy, the sheen may be suppressed.

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

Light yellow to pink homogeneous free flowing powder

Colour and Clarity of prepared medium

Red coloured clear solution without any precipitate

Reaction

Reaction of 7.62% w/v aqueous solution at 25°C. pH : 7.4±0.2

pН

7.20-7.60

Cultural Response

Cultural characteristics on membrane filter after an incubation at different temperatures for 24 hours

Organism	Inoculum (CFU)	Growth at 35-37°C	Growth at 44°C	Colour of Colony on Membrane
# Klebsiella aerogenes ATCC 13048 (00175*)	50-100	luxuriant	inhibited	yellow
Bacillus subtilis ATCC subsp. spizizenii 6633 (00003*)	>=10 ⁴	inhibited	inhibited	
Staphylococcus aureus subsp. aureus ATCC 25923 (00034*)	>=10 ⁴	inhibited	inhibited	
Enterococcus faecalis ATCO 29212 (00087*)	$C >= 10^4$	inhibited	inhibited	
Escherichia coli ATCC 25922 (00013*)	50-100	luxuriant	luxuriant	yellow

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 15-25C. 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.

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 (7,11).

Reference

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- 5. Eaton A. D., Clesceri L. S. and Greenberg A. E., (Eds.), 1995, Standard Methods for the Examination of Water and Wastewater, 19th Ed., American Public Health Association, Washington, D.C.
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11. 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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