

For identification and differentiation of *E. coli* and Total coliforms

M-E. coli Broth

Recommended for the detection, differentiation and enumeration of *Escherichia coli* and coliforms in water samples by membrane filtration technique.

M1426

Composition **

Ingredients	Grams/Litre
Tryptone	20.00
Bile salts mixture	1.50
Chromogenic mixture	0.175

Final pH (at 25°C) 7.2 ± 0.2

** Formula adjusted, standardized to suit performance parameters

Directions

Suspend 21.67 grams in 1000 ml distilled water. Heat if necessary to dissolve the medium completely. DO NOT AUTOCLAVE. Cool to 45-50°C. Aseptically add desired quantity (2 to 5 ml) of broth on sterile absorbent cotton pad or sterile filter paper for saturation. The medium should be used within 24 hours of rehydration.

Principle and Interpretation

M-E.coli Broth is used for detection and differentiation of *Escherichia coli* and coliforms in water samples using membrane filter technique. It is based on Tryptone Bile Agar used for detection of *Escherichia coli* in foods (1) where recovery of *Escherichia coli* is faster, more reliable and accurate.

The water sample is filtered through membranes and then placed on pad saturated with M-E.coli Broth and incubated at 37°C in sealed Petri plates. Glucuronidase test is used increasingly for detection of *E. coli* in water and food microbiology as *E. coli* is an important indicator of fecal contamination in samples from the food processing and water purification plants. Other *Escherichia* spp. do not produce this enzyme (3). The medium contains chromogenic mixture which helps to detect glucuronidase activity of *Escherichia coli* (2). This specific enzyme differentiates *Escherichia coli* from other coliforms. *Escherichia coli* cells split the chromogenic mixture with the help of the enzyme glucuronidase to give blue to green colouration to the colonies. Coliforms other than *Escherichia coli* turn red as they reduce TTC (2,3,5-triphenyl tetrazolium chloride). Thus, the resulting colour distinction allows simple interpretation of test without further confirmation.

Tryptone provides nitrogen and carbon source, long chain amino acids, vitamins and other essential growth nutrients to the organisms. Bile salt mixture inhibit gram-positive organisms.

Type of specimen

Water samples

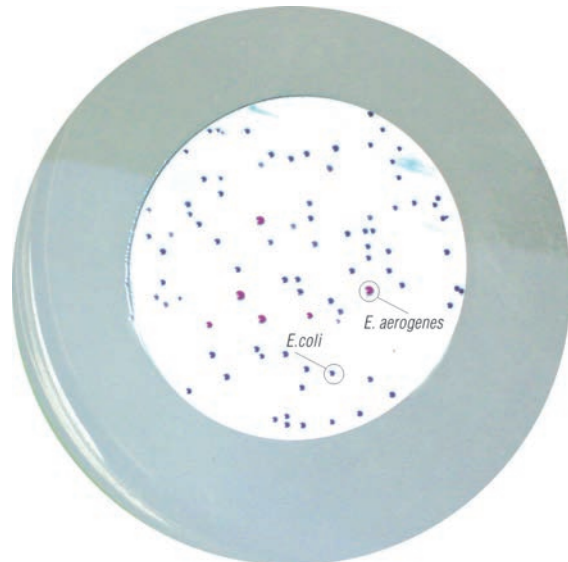
Specimen Collection and Handling

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (4).

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



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Limitations

1. β -glucuronidase is present in 97% of *E. coli* strains, however few *E. coli* may be negative.
2. Since the medium is highly selective, some strains may show poor growth due to nutritional variations.

Performance and Evaluation

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

Quality Control

- Appearance of Powder** : Light yellow to beige coloured, homogeneous, free flowing powder.
- Colour and Clarity of prepared medium** : Light yellow coloured, clear solution without any precipitate
- Reaction** : Reaction of 2.17% w/v aqueous solution at 25°C. pH : 7.2 ± 0.2.
- Cultural Response** : Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organisms (ATCC)	Inoculum (CFU)	Growth	Colour of colony on membrane filter
<i>Escherichia coli</i> (25922) (00013*)	50-100	luxuriant	blue
# <i>Klebsiella aerogenes</i> (13048) (00175*)	50-100	luxuriant	red
<i>Staphylococcus aureus</i> subsp <i>aureus</i> (25923) (00034*)	≥10 ³	inhibited	—

Key : * : corresponding WDCM Numbers

: Formerly known as *Enterobacter aerogenes*

Storage and Shelf-life

Store between 2-8°C in a tightly closed container and the prepared medium at 2-8°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).

References

1. Anderson J. M. and Baird Parker A.C., (1975), J. Appl. Bact., 39:111.
2. Hansen W. and Yourassawsky E., (1984), J. Clin. Microbiol. 20:1177.
3. Rice, E.W., Allen, M.J., Brenner, D.J., Edberg, S.C., 1991. Appl. Environ. Microbiol. 57, 592–593.
4. 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
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.

Ready Prepared Media			
Code	Product Name	Usage	Packing
Category : DriFilter Membrane Nutrient Pad			
MF027	M-E.coli Medium (without Membrane Filter)	for detection and enumeration of total coliforms and <i>E. coli</i> based on chromogenic differentiation	50 plts
MF027E	M-E.coli Medium (Economy Pack) (without Membrane Filter)	for detection and enumeration of total coliforms and <i>E. coli</i> based on chromogenic differentiation	50 pcs
MF027F	M-E. coli Medium w/ Sterile Membrane Filter	for detection and enumeration of total coliforms and <i>E. coli</i> based on chromogenic differentiation	50 plts