

Chromogenic Coliform Medium w/sterile membrane filter

MF034F

Intended use

for detection of *E.coli* and coliforms in water samples.

Composition**

Proprietary

Directions

The test sample should be filtered through a sterile membrane filter having pore size of 0.22 μ / 0.45 μ . Rehydrate the nutrient pad with 2.0-2.5 ml sterile distilled / purified water. After filtration, remove the membrane filter aseptically using sterile forceps. Place the membrane filter on rehydrated nutrient pad. Incubate the inoculated nutrient. Interpret the results qualitatively by observing the presence or absence of growth and quantitatively by counting the number of colonies on the surface of the membrane filter and calculating CFU/ml.

Principle And Interpretation

This is a selective medium for detection of *Escherichia coli* and total coliforms in water samples (1). The medium contains three chromogenic substrates. The enzyme β -D-galactosidase produced by coliforms cleaves 6-chloro-3-indoxyl- β -D-galactopyranoside to form pink to red coloured colonies (3). The enzyme β -D-glucuronidase produced by *E.coli*, cleaves 5-bromo-4-chloro-3-indoxyl- β -D-glucuronic acid (2). Colonies of *E.coli* give dark blue to violet coloured colonies due to cleavage of both the chromogens. The presence of the third chromogen IPTG enhances the colour reaction. Addition of L-Tryptophan improves the indole reaction thereby increasing the detection reliability.

To confirm *E.coli*, add a drop of Kovacs reagent on the dark blue to violet colony. Formation of cherry red colour indicates a positive reaction.

Type of specimen

Water samples - Water and wastewater

Specimen Collection and Handling

DriFilter Membrane Nutrient Pad Medium is ready to use sterile culture media in the form of a 50 mm biological inert absorbent pads impregnated with MacConkey medium, then dried and sterilized in 55 mm petri plate. They eliminate the need of laborious media preparation and autoclaving procedures. The nutrient pads are to be just rewetted with sterile distilled water and are ready to use. Use of nutrient pads allows larger sample volumes to be tested at a time. Interpretation of results is directly by counting the CFUs and also quantifies the microbial load present in the sample. 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 testing is required for identification of microorganism.
2. Certain variations in colour may be observed .

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

Dry filter membrane pad of 50mm diameter

Colour

Light pink coloured nutrient pad

Sterility test

Passes release criteria

Cultural Response

Cultural characteristics observed after an incubation at 34-38°C for 24 hours.

Organism	Inoculum (CFU)	Growth	Colour of Colony on membrane
<i>Citrobacter freundii</i> ATCC 43864 (00006)*	50-100	luxuriant	pale pink to pink
<i>Enterobacter aerogenes</i> ATCC 13048 (00175)*	50-100	luxuriant	pale pink to pink
<i>Escherichia coli</i> ATCC 25922 (00013)*	50-100	luxuriant	dark blue to violet
<i>Escherichia coli</i> ATCC 8739 (00012)*	50-100	luxuriant	dark blue to violet
<i>Enterococcus faecalis</i> ATCC 19433 (00009)*	$\geq 10^3$	inhibited	
<i>Pseudomonas aeruginosa</i> ATCC 10145(00024)*	50-100	luxuriant	colourless

Key * : Corresponding WDCM numbers

Storage and Shelf Life

On receipt, Store between 2-8°C. Use before expiry date on the label.

Reference

- 1.International Organization for Standardization. Water quality: Enumeration of *E.coli* and coliform bacteria. Part I- Membrane filtration methods for bacteria with low bacterial background flora. ISO 9308-1:2014.
- 2.Kilian M. and Bülow P., 1976, Acta. Pathol. Microbiol. Scand Sect. B, 84:245.
- 3.Manafi M. and Kneifel W., 1989, Zentralbl. Hyg., 189:225.

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