HiCrome™ M-Coliconfirm Agar Base

M2058

Intended use

Recommended for the selective isolation and identification of *E.coli* and coliforms from water samples using membrane filtration technique.

Composition**

Ingredients	Gms / Litre
Tryptone	8.000
Growth factors	2.200
Buffers	3.300
Chromogenic mixture	3.950
Selective mix	1.520
Agar	15.000
Final pH (at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 33.97 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C. Aseptically add the rehydrated contents of one vial of TTC Solution, 1% (FD057). Mix well and pour into sterile Petri plates.

Principle And Interpretation

It is possible to remove bacteria from fluids by passing them through filters with such small pore size that bacteria are arrested. This filtration technique enables fairly large volumes of water to pass rapidly under pressure, but prevents the passage of any bacteria present. These nutrients are retained on the surface of the membrane which is then brought into contact with suitable liquid nutrients. These diffuse upwards through the pores thereby inducing the organisms to grow as surface colonies which can be counted (1).

Escherichia coli, a member of the family *Enterobacteriaceae* is a normal flora of the intestinal tract of humans and a variety of animals. Although most of the *E.coli* does not cause gastrointestinal illness, certain groups of *E.coli* can cause life threatenng diarrhoea and severe sabilities. For water testing, detection of *E.coli* and total coliforms is very important. There are various media available for the detection of *E.coli* and total coliforms.

Tryptone, growth factors provide nitrogeneous and carbonaceous compound, long chain amino acids, vitamins and other essential nutrients. Phosphates buffers the medium well. Selective agents inhibit gram positive bacteria and other accompanying bacteria. The chromogenic mix incorporated in the medium aids in the detection of β-glucuronidase positive organisms which gives blue coloured organisms. Coliforms produce red coloured colonies other than *Escherichia coli* as they reduce TTC (2,3,5-triphenyl tetrazolium chloride). Thus, the resulting colour distinction allows simple interpretation of test without further confirmation.

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 (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 guidleines should be followed while handling specimens. Saftey guidelines may be referred in individual safety data sheets

Limitations :

1.This medium is based on the principle of detection of β-glucuronidase enzyme. 97% of the *E.coli* are β-glucuronidase positive and will result in blue colour.
2. Certain other *Salmonella* and *Shigella* species may also exhibit β glucuronidase activity.

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

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Light blue coloured, clear to slightly opalescent gel forms in Petri plates

Reaction

Reaction of 3.4% w/v aqueous solution at 25°C. pH : 7.2±0.2

pH

7.00-7.40

Cultural Response

M2058: Cultural characteristics observed on membrane filter after an incubation at 35-37°C for 18-24 hours .

Organism	Inoculum (CFU)	Growth	Colour of Colony
Escherichia coli ATCC 25922 (00013*)	50-100	luxuriant	blue
Klebsiella pneumoniae ATCC 13883 (00097*)	50-100	luxuriant	maroon- red
# Klebsiella aerogenes ATCC 13048 (00175*)	50-100	luxuriant	maroon- red
Salmonella Enteritidis ATCC 13076 (00030*)	50-100	luxuriant	maroon- red
Staphylococcus aureus subsp. aureus ATCC 25923 (00034)*	>=10^3	inhibited	-
Enterococccus faecalis ATCC 29212 (00087)*	>=10^3	inhibited	-
Aeromonas hydrophila ATCC 7966	50-100	none-poor	-

Key: (#) Formerly known as *Enterobacter aerogenes, (*)* corresponding *WDCM numbers*

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 inorder 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. Cruickshank R., Duguid J. P., Marmion B. P., Swain R. H. A., (Eds.), Medical Microbiology, 1975, 12th Ed. Vol. II, Churchill Livingstone

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 Handb0ook. 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.

Revision : 00 / 2017