

HiCrome™ M-Coliform Differential Agar Base

Recommended as a selective and differential agar for the detection of coliform bacteria using membrane filtration technique.

M1951

Composition **

Ingredients	Grams/Litre
Peptone	5.00
Tryptone	10.00
Yeast extract	3.00
Lactose	12.50
Sodium deoxycholate	0.15
Aniline Blue	0.10
Chromogenic substrate	0.50
Agar	15.00

Final pH (at 25°C) 7.2 ± 0.2

** Formula adjusted, standardized to suit performance parameters

Directions

Suspend 46.25 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. DO NOT AUTOCLAVE. Cool to 45- 50°C. Aseptically add the rehydrated contents of one vial of Monensin Selective supplement (FD309). Mix well and pour into sterile Petri plates.

Principle and Interpretation

HiCrome™ M-Coliform Differential Agar Base is based on coliform enumeration medium, M-FC Agar (1). This medium was modified for detection and enumeration of total coliforms by addition of Monensin supplement to improve the recovery of injured coliforms (2).

Peptone, Tryptone and Yeast extract provides nitrogenous compounds, carbonaceous compounds, long chain amino acids and other growth nutrients and vitamins. Lactose is the fermentable carbohydrate. Monensin and sodium deoxycholate acts as selective agents, inhibiting Gram-positive bacteria. Aniline blue forms the indicator system of the medium. The chromogenic substrate is utilized by *E.coli* which detects the presence of β -glucuronidase. The medium helps injured coliforms to grow in the presence of selective agents.

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 (3).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

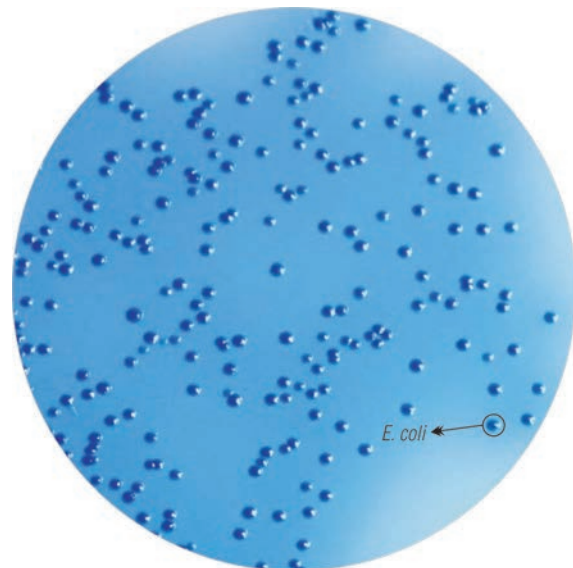
Read the label before opening the container. Wear protective gloves/ protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets

Limitations

1. 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.



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Quality Control

- Appearance of Powder** : Light yellow to greyish yellow homogeneous free flowing powder
- Gelling** : Firm, comparable with 1.5% Agar gel.
- Colour and Clarity of prepared medium** : Light yellow, clear to slightly opalescent gel forms in Petri plates
- Reaction** : Reaction of 4.63% 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	Recovery	Colour of colony (On membrane filter)
<i>Escherichia coli</i> (25922) (00013*)	50-100	good-luxuriant	≥50%	blue
<i>Proteus vulgaris</i> (13315)	50-100	good-luxuriant	≥50%	tan
<i>Bacillus spizizenii sub spizizenii</i> (6633) (00003*)	≥10 ³	inhibited	0%	

Key : * : 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 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 (4, 5).

References

1. Brodsky, M. H., P. Entis, A. N. Sharpe, and G. A. Jarvis. 1982. Enumeration of indicator organisms in foods using the automated hydrophobic membrane filter technique. *J. Food Prod.* 45:292-296.
2. Entis, P., and P. Boleszczuk. 1990. Direct enumeration of coliforms and *Escherichia coli* by hydrophobic grid membrane filter in 24 hours using MUG. *J. Food Prot.* 53:948-952.
3. 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
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
5. 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