



Instructions for Use

M-FC AGAR WITH 1% ROSOLIC ACID

Cat. no. G126	m-FC Agar with 1% Rosolic Acid, 15x60mm Plate, 12ml	10 plates/bag
Cat. no. G127	m-FC Agar with 1% Rosolic Acid, 15x100mm Plate, 18ml	10 plates/bag
Cat. no. G272	m-FC Agar with 1% Rosolic Acid, 11x50mm Plate, 6ml	10 plates/bag
Cat. no. G391	m-FC Agar with 1% Rosolic Acid, 15x60mm Plate, 12ml	10 plates/bag

INTENDED USE

Hardy Diagnostics m-FC Agar with 1% Rosolic Acid is recommended for the detection of fecal coliforms in water samples by membrane filtration and elevated temperature.

This product is not intended to be used for the diagnosis of human disease.

SUMMARY

Geldreich et al. formulated a medium to enumerate fecal coliforms (m-FC) using the membrane filter (MF) technique without prior enrichment. ⁽¹⁾ Fecal coliforms, which are found in the gastrointestinal tracts and feces of warm-blooded animals, are differentiated from coliforms from environmental sources by their ability to grow at elevated temperatures: 44.5 +/- 0.5°C. ⁽²⁾

The m-FC method for detection of fecal coliforms can be used for monitoring all types of water. However, because many coliforms may also come naturally from environmental sources, it is recommended that *Escherichia coli* be the required indicator for freshwater testing and *E. coli* or enterococci be the required indicators for marine water testing, since these microorganisms are known fecal contaminants. ⁽⁵⁾

Hardy Diagnostics m-FC Agar with 1% Rosolic Acid contains peptones as a source of carbon, nitrogen, vitamins and minerals. Yeast extract supplies B-complex vitamins that help stimulate bacterial growth. Lactose is a carbohydrate that can be fermented by fecal coliforms at elevated temperatures. Bile salts no. 3 inhibit the growth of undesirable microorganisms. Agar is added as the solidifying agent. Finally, aniline blue and rosolic acid are utilized to differentiate fecal from non-fecal coliforms.

FORMULA

Ingredients per liter of deionized water:*

Lactose	12.5gm
Tryptose	10.0gm
Proteose Peptone No. 3	5.0gm
Sodium Chloride	5.0gm
Yeast Extract	3.0gm
Bile Salts No. 3	1.5gm
Aniline Blue	0.1gm
Rosolic Acid	0.1gm
Agar	15.0gm

Final pH 7.4 +/- 0.2 at 25°C.

* Adjusted and/or supplemented as required to meet performance criteria.

STORAGE AND SHELF LIFE

Storage: Upon receipt store at 2-8°C. away from direct light. Media should not be used if there are any signs of deterioration (shrinking, cracking, or discoloration), contamination, or if the expiration date has passed. Product is light and temperature sensitive; protect from light, excessive heat, moisture, and freezing.

The expiration dating on the product label applies to the product in its intact packaging when stored as directed. The product may be used and tested up to the expiration date on the product label and incubated for the recommended quality control incubation times.

Refer to the document "[Storage](#)" for more information.

PRECAUTIONS

This product may contain components of animal origin. Certified knowledge of the origin and/or sanitary state of the animals does not guarantee the absence of transmissible pathogenic agents. Therefore, it is recommended that these products be treated as potentially infectious, and handle observing the usual universal blood precautions. Do not ingest, inhale, or allow to come into contact with skin.

This product is for laboratory use only. It is to be used only by adequately trained and qualified laboratory personnel. Observe approved biohazard precautions and aseptic techniques. All laboratory specimens should be considered infectious and handled according to "standard precautions." The "Guidelines for Isolation Precautions" is available from the Centers for Disease Control and Prevention at www.cdc.gov/ncidod/dhqp/gl_isolation.html.

For additional information regarding specific precautions for the prevention of the transmission of all infectious agents from laboratory instruments and materials, and for recommendations for the management of exposure to infectious disease, refer to CLSI document M-29: *Protection of Laboratory Workers from Occupationally Acquired Infections: Approved Guideline*.

Sterilize all biohazard waste before disposal.

Refer to the document "[Precautions When Using Media](#)" for more information.

Refer to the document [SDS Search](#) instructions on the Hardy Diagnostics' website for more information.

PROCEDURE

Sample Collection: Consult listed references for information on sample collection. ⁽⁶⁻¹⁰⁾

Method of Use:

1. Filter duplicate specimens through separate membrane filters.
2. Roll membrane filter used to collect the water sample onto the agar surface. Avoid the formation of air bubbles between the filter and agar surface.
3. Place plates into separate waterproof plastic bags and seal bags so as to prevent leakage.
4. Incubate plates, by immersion, in two separate water baths: one set at 35 +/- 2°C. and one set at 44.5 +/- 0.5°C.
5. Anchor plates below water surface to maintain critical temperature requirements and incubate for 24 +/- 2 hours. Place all inoculated plates in water bath within 30 minutes after filtration for best results. As an alternative, an appropriate and accurate solid heat sink or equivalent incubator may be used.

INTERPRETATION OF RESULTS

Colonies produced by fecal coliforms will appear as varying shades of blue-colored colonies on the membrane filter. Coliforms from other sources should be inhibited; however, if present they may appear as gray to cream-colored colonies.

Calculate fecal coliform densities as directed by listed references. ^(2,4-6,9)

LIMITATIONS

It is recommended that biochemical, immunological, molecular, or mass spectrometry testing be performed on colonies from pure culture for complete identification.

Even with the selective action of the elevated incubation temperature and addition of rosolic acid, a few non-fecal coliforms may be observed. To eliminate the potential for *Klebsiella* contamination, it may be helpful to elevate the incubation temperature to 45 +/- 0.2°C. ⁽²⁾ In addition, it is recommended that biochemical and/or serological tests be performed on colonies from pure culture for complete and accurate identification.

Refer to the document "[Limitations of Procedures and Warranty](#)" for more information.

MATERIALS REQUIRED BUT NOT PROVIDED

Standard microbiological supplies and equipment such as loops, filter membranes, petri dishes, swabs, applicator sticks, other culture media, incinerators, and incubators, etc., as well as serological and biochemical reagents, are not provided.

QUALITY CONTROL

Hardy Diagnostics tests each lot of commercially manufactured media using appropriate quality control microorganisms and quality specifications as outlined on the Certificates of Analysis (CofA). The following organisms are routinely used for testing at Hardy Diagnostics:

Test Organisms	Inoculation Method*	Incubation			Results
		Time	Temperature	Atmosphere	
<i>Escherichia coli</i> ATCC® 25922	MF	24hr	44.5°C	Aerobic	Growth; blue colonies
<i>Enterobacter aerogenes</i> ATCC® 13048	MF	24hr	44.5°C	Aerobic	Slight to no growth

* Refer to the document "[Inoculation Procedures for Media QC](#)" for more information.

USER QUALITY CONTROL

End users of commercially prepared culture media should perform QC testing in accordance with applicable government regulatory agencies, and in compliance with accreditation requirements. Hardy Diagnostics recommends end users check for signs of contamination and deterioration and, if dictated by laboratory quality control procedures or regulation, perform quality control testing to demonstrate growth or a positive reaction and to demonstrate inhibition or a negative reaction, if applicable. Hardy Diagnostics quality control testing is documented on the certificates of analysis (CofA) available from Hardy Diagnostics [Certificates of Analysis](#) website. In addition, refer to the following document "[Finished Product Quality Control Procedures](#)," for more information on QC or see reference(s) for more specific information.

PHYSICAL APPEARANCE

Hardy Diagnostics m-FC Agar with 1% Rosolic Acid should appear clear to slightly hazy and purple-red to purple-blue in color.

REFERENCES

1. Geldreich E.E., H.F. Clark, C.B. Huff, and L.C. Best. 1965. Fecal coliform organism medium for the membrane filter technique. *J. Am. Water Works Assoc.* ; 57:208-214.
2. Clesceri, L.S., A.E. Greenberg and A.D. Eaton (ed.). 1998. *Standard Methods for the Examination of Water and Wastewater* , 20th ed. American Public Health Association, Washington, D.C.
3. Horwitz, W. 2000. *Official Methods of Analysis of AOAC International* , 17th ed. AOAC International. Gaithersburg, MD.
4. U.S. Environmental Protection Agency. 1992. *Manual for the Certification of Laboratories Analyzing Drinking Water* . EPA-814B-92-002. Office of Ground Water and Technical Support Division, USEPA, Cincinnati, OH.
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6. Anderson, N.L., et al. *Cumitech 3B; Quality Systems in the Clinical Microbiology Laboratory*, Coordinating ed., A.S. Weissfeld. American Society for Microbiology, Washington, D.C.
7. Jorgensen., et al. *Manual of Clinical Microbiology*, American Society for Microbiology, Washington, D.C.
8. Tille, P., et al. *Bailey and Scott's Diagnostic Microbiology*, C.V. Mosby Company, St. Louis, MO.
9. American Public Health Association. *Standard Methods for the Examination of Water and Wastewater*, APHA, Washington, D.C.
10. Association of Official Analytical Chemists. *Official Methods of Analysissm* , AOAC, Washington, D.C.

ATCC is a registered trademark of the American Type Culture Collection.

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