m Endo Broth MF™

Intended Use

m Endo Broth MF™* is used for enumerating coliform organisms in water by membrane filtration.

**MF is a trademark of Millipore Corporation.*

Summary and Explanation

m Endo Broth MF is prepared according to the formulation of the Millipore Corporation[1] for selectively isolating coliform bacteria from water and other specimens using the membrane filtration technique. The medium is a combination of the former m HD Endo Medium and Lauryl Tryptose Broth.

The American Public Health Association (APHA) specifies using m Endo Broth MF in the standard total coliform membrane filtration procedure for testing water[2] and bottled water.[3] APHA also specifies using m Endo Broth MF in the delayed-incubation total coliform procedure by adding sodium benzoate to make m Endo preservative medium.[2] The coliform bacteria are defined as bacteria that produce a red colony with a metallic sheen within 24 hours incubation at 35°C on an Endo-type medium.

Principles of the Procedure

m Endo Broth MF contains peptones as sources of carbon, nitrogen, vitamins and minerals. Yeast extract supplies B-complex vitamins, which stimulate bacterial growth. Lactose is the carbohydrate. Phosphates are buffering agents. Sodium chloride maintains the osmotic balance of the medium. Sodium desoxycholate and sodium lauryl sulfate are added as inhibitors of gram-positive bacteria. Basic fuchsin is a pH indicator. Sodium sulfite is added to decolorize the basic fuchsin solution. The ethanol additive increases the antibacterial nature of the formulation.

Lactose-fermenting bacteria produce acetaldehyde that reacts with the sodium sulfite and fuchsin to form red colonies. The development of a metallic sheen occurs when the organism produces aldehydes with the rapid fermentation of lactose. If the inoculum is too heavy, the sheen will be suppressed. Lactose-nonfermenting bacteria form clear, colorless colonies.

Formula

Difco™ m Endo Broth MF™

Approximate Formula* Per Liter

Yeast Extract	1.5	g
Casitone	5.0	g
Thiopeptone	5.0	g
Tryptose	10.0	g
Lactose	12.5	g
Sodium Desoxycholate	0.1	g
Dipotassium Phosphate	4.375	g
Monopotassium Phosphate	1.375	g
Sodium Chloride	5.0	g
Sodium Lauryl Sulfate	0.05	g
Sodium Sulfite	2.1	g
Basic Fuchsin	1.05	g

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

Directions for Preparation from Dehydrated Product

1. Dissolve 48 g of the powder in 1 L of purified water containing 20 mL nondenatured ethanol.
2. Heat to boiling but avoid over-heating. DO NOT AUTOCLAVE.
3. Test samples of the finished product for performance using stable, typical control cultures.

CAUTION: Protect from light. Use immediately.

Procedure

1. Place a membrane filter absorbent pad inside a sterile 60 mm Petri dish.
2. Add 1.8-2.0 mL m Endo Broth MF to each pad.
3. Filter the water sample through a membrane filter.
4. Place filter top side up on the pad using a rolling motion to avoid entrapping air bubbles.
5. Invert the dish and incubate for 22-24 hours at 35 ± 0.5°C.
6. Observe and count all colonies that are red and have a metallic sheen.

Expected Results

All colonies that are red and have the characteristic metallic sheen are considered coliforms. The sheen may cover the entire colony, may only be in the center or may appear only around the edges.

Limitations of the Procedure

Occasionally, noncoliform organisms may produce typical sheen colonies. Coliform organisms may also occasionally produce atypical colonies (dark red or nucleated colonies without sheen). It is advisable to verify both colony types.[2]

User Quality Control

Identity Specifications

Difco™ m Endo Broth MF™

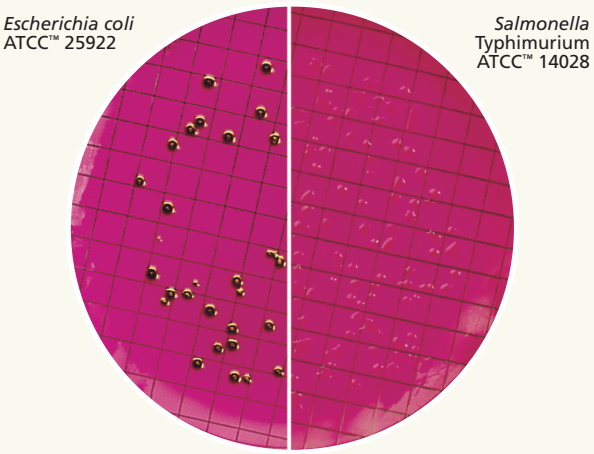
Dehydrated Appearance:	Pinkish purple, free-flowing, homogeneous.
Solution:	4.8% solution, partially soluble in purified water containing 2% ethanol upon boiling. Solution is pinkish-red, opalescent with precipitate.
Prepared Appearance:	Pinkish-red, opalescent with precipitate.
Reaction of 4.8% Solution at 25°C:	pH 7.2 ± 0.1

Cultural Response

Difco™ m Endo Broth MF™

Prepare the medium per label directions. Use the membrane filter technique to inoculate filters. Incubate on pads saturated with m Endo Broth MF at 35 ± 2°C for 24 ± 2 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY	COLONY COLOR
Escherichia coli	25922	20-80	Good	Red with green metallic sheen
Salmonella enterica subsp. *enterica* serotype Typhimurium	14028	20-80	Good	Colorless to pink
Staphylococcus aureus	25923	10^3-2×10^3	Marked inhibition	–



References

1. Fifield and Schaufus. 1958. J. Am. Water Works Assoc. *50*:193.
2. Eaton, Rice and Baird (ed.). 2005. Standard methods for the examination of water and wastewater, 21st ed., online. American Public Health Association, Washington, D.C.
3. Kim and Feng. 2001. *In* Downes and Ito (ed.), Compendium of methods for the microbiological examination of foods, 4th ed. American Public Health Association, Washington, D.C.

Availability

Difco™ m Endo Broth MF™

COMPF SMD SMWW

Cat. No.	274920	Dehydrated – 100 g
	274930	Dehydrated – 500 g