

M – ENDO AGAR LES**TM 174**

For enumeration of coliforms in water using a two step membrane filter technique

Composition

Ingredients	Gms/Ltr.
Agar	15.00
Casein enzymatic hydrolysate	11.02
Lactose	9.40
Peptic digest of animal tissue	3.70
Sodium chloride	3.70
Dipotassium phosphate	3.30
Sodium sulphite	1.60
Yeast extract	1.20
Monopotassium phosphate	1.00
Basic fuschin	0.80
Sodium deoxycholate	0.10
Sodium lauryl sulphate	0.05

* Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers below 25°C and protect from direct Sunlight.

Instructions for Use

Dissolve 51gms in 980ml distilled water. Gently heat to boiling with gentle swirling and dissolve the medium completely. **DO NOT AUTOCLAVE**. Mix well before use the medium. Cool to 45-50°C and aseptically add 20 ml of 95% Ethanol. Mix well and dispense into sterile Petri plates.

Appearance: Rose color, slightly opalescent
pH (at 25°C): 7.2 ± 0.2

Principle

M – ENDO AGAR LES is used for enumeration of coliforms in water using a two step membrane filter technique. Medium contains Casein enzymatic hydrolysates and Peptic digest of animal tissue as the sole sources of carbon, nitrogen, vitamins and minerals. Yeast extract supplies B-complex vitamins, which stimulate bacterial growth in the medium. Lactose serves as the carbohydrate source. Monopotassium and Dipotassium phosphate are buffering agents. Sodium chloride maintains the osmotic balance of the medium. Sodium deoxycholate and Sodium lauryl sulphate are added as inhibitors. Basic fuchsin is a pH indicator. Sodium sulphite is added to decolorize the basic fuchsin solution. Agar is the solidifying agent. Ensure that ethanol used in rehydration procedure is not denatured. Prepare medium in a sterile flask and use a boiling water bath or, if constantly attended, a hot plate with a stir bar to bring medium to the boiling point. Do not boil medium. This media is used as **Multiple Tube Fermentation (MTF) Media**. Use double strength Lauryl Sulphate Broth or Lactose broth in the presumptive test and single strength Brilliant Green Lactose Bile Broth (BGLB) in the confirmed test. Sterilize the media at 121°C for 12 minutes. Final pH shall be 6.8 ± 0.2 or 7.2 ± 0.2 for BGLB broth. If MTF media are refrigerated after sterilization, incubate overnight at 35°C ± 0.5°C before use. Discard tubes showing growth or bubbles. Use MTF media prepared in tubes with loose fitting closures within one week. Store broth media in screw cap tubes or bottles no longer than three months, provided media are stored in the dark. Discard media if

PRODUCT DATA SHEET

evaporation exceeds 10 percent of original volume. M- Endo Agar LES shall be used for the completed test. Refrigerated medium be used within two weeks.

Interpretation

Cultural characteristics observed after inoculating (10^3 CFU/ml), on incubation at $35 \pm 0.5^\circ\text{C}$ for 20 to 24 hours.

Microorganisms	ATCC	Inoculum (CFU/ml)	Growth	Appearance of colony
<i>Escherichia coli</i>	25922	10^3	Good -Luxuriant	Pink with metallic sheen
<i>Enterobacter aerogenes</i>	13048	10^3	Good -Luxuriant	Pink to red (may have sheen)
<i>Proteus mirabilis</i>	14273	10^3	Good -Luxuriant	Colourless
<i>Staphylococcus aureus</i>	25923	10^3	Inhibited	-
<i>Enterococcus faecalis</i>	19433	10^3	None/poor	-

References

1. American Public Health Association, American Water Works Association and Water Pollution Control Federation: Standard Methods for the Examination of Water and Wastewater, 20th Ed., Washington. (1998).