

## TMV 174 - M-ENDO AGAR, LES(VEG.)

### INTENDED USE

For enumeration of coliforms in water using a twostep membrane filter technique.

### PRODUCT SUMMARY AND EXPLANATION

M-Endo Veg Agar LES is prepared by completely replacing animal based peptones with vegetable peptones which make the medium free of BSE/TSE risks. M-Endo Veg Agar LES is the modification M-Endo Agar LES (Lawrence, Experimental Station) which is formulated according to the formulation of McCarthy, Delaney and Grasso and is used for the enumeration of coliforms in water. Membrane filter technique for coliform enumeration is more reliable and precise than MPN multiple tube test. A two-stage process has been suggested for enrichment to get a non-toxic environment for maximum revival of the coliforms. This medium, like the conventional medium is based on the medium described by Endo for the differentiation of lactose fermenters from non-fermenters.

In the first step of enrichment, the pad is impregnated with Lauryl Tryptose Veg Broth. Membrane filter through which water sample is passed is aseptically placed on it and incubated without inverting for 2 hours at 35°C in a humid atmosphere. After incubation, the membrane filter is aseptically transferred to the M-Endo Veg Agar LES plate and incubated at 35°C for 24 hours. Alternatively, membrane filter pad can be placed inside the lid of petri plate of M-Endo Agar LES and then impregnated with 2 ml Lauryl Tryptose Veg Broth and incubated for 1 - 1½ hour at 35°C. In the second step, the prepared membrane filter is kept directly on the agar surface and incubated as described above. Presumptive coliforms produce golden green colonies with metallic sheen within 24 hours of incubation. If the inoculum is too heavy, the sheen will be suppressed. Sometimes non-coliform organisms may produce typical colonies with sheen, coliforms may also occasionally produce atypical colonies (dark red without sheen).

Coliform density calculation: Note the coliform density in terms of total coliforms/100 ml. Extrapolate the count using membrane filters with 20-80 coliform colonies but not more than 200 of all types per membrane.

The formula for calculating the count is as follows: Total coliform = coliform colonies x 100 colonies/100 ml of sample filtered.

### COMPOSITION

Ingredients	Gms / Ltr
Veg hydrolysate	3.7
Veg peptone	3.7
Veg hydrolysate No. 1	7.5
Yeast extract	1.2
Lactose	9.4
Dipotassium phosphate	3.3
Monopotassium phosphate	1.0
Sodium chloride	3.7
Synthetic detergent No. III	0.1
Sodium lauryl sulphate	0.05
Sodium sulphite	1.6
Basic fuchsin	0.8
Agar	15.0

### PRINCIPLE



Veg hydrolysate, veg hydrolysate No.1, Veg peptone and yeast extract provide essential nutrients especially nitrogenous for the coliforms. Lactose is the fermentable carbohydrate. Sodium sulphite, Synthetic detergent No. III and basic fuchsin inhibit the growth of gram-positive organisms. Phosphates buffer the medium. Coliforms ferment the lactose and form red colonies and similar coloration of the medium. Lactose non-fermenters form colourless colonies.

### INSTRUCTION FOR USE

- Dissolve 51.00 grams in 980 ml purified/distilled water.
- Heat to boiling the medium to dissolve completely. Do not autoclave.
- Cool to 45-50°C and add 20 ml of 95% ethanol aseptically.
- Mix and dispense 4 ml amounts into 60 mm Petri plates. In large plates, use sufficient medium to give 1.5 mm depth. Do not expose plates directly to sunlight.

Caution: Basic fuchsin is a potential carcinogenic, avoid inhalation and contamination on skin.

### QUALITY CONTROL SPECIFICATIONS

<b>Appearance of Powder</b>	: Light purple to purple coloured, homogeneous, free flowing powder
<b>Appearance of prepared medium</b>	: Red coloured, slightly opalescent gel forms in petri plates
<b>pH (at 25°C)</b>	: 7.2 ± 0.2

### INTERPRETATION

Cultural characteristics observed after incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Color of the colony (on membrane filter)	Incubation Temperature	Incubation Period
<i>Escherichia coli</i>	25922	50-100	Luxuriant	≥70%	red to black with sheen	35-37°C	20-24 Hours
<i>Klebsiella aerogenes</i>	13048	50-100	Luxuriant	≥70%	red (may have sheen)	35-37°C	20-24 Hours
<i>Salmonella serotype Typhi</i>	6539	50-100	Luxuriant	≥70%	Colorless to very light pink	35-37°C	20-24 Hours
<i>Staphylococcus aureus</i>	25923	50-100	Inhibited	0%	-	35-37°C	20-24 Hours
<i>Salmonella serotype Typhi</i>	14028	50-100	Luxuriant	≥70%	Colorless to very light pink	35-37°C	20-24 Hours

### PACKAGING:

In pack size of 100 gm and 500 gm bottles.



## STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.










**Product Deterioration:** Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

## DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

## REFERENCES

1. McCarthy J.A., Delaney J.E. and Grasso R., 1961, Water and Sewage Works, 108:238.
2. American Public Health Association, 1980, Standard Methods for the Examination of the Water and Wastewater, 15th ed. APHA, Inc., Washington, D.C.
3. Endo, 1904, Zentrabl. Bakteriologie. Abt. I. Orig., 35:109.

 Good Manufacturing Practices Certified	 Best Before	 Quantity	 Catalogue Number	 Manufacturer
 Temperature Unit	 Lot / Batch Number	 Consults Instructions for Use	 QR Code	

**NOTE:** Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.

**\*For Lab Use Only**  
Revision: 08 Nov., 2019