M-Endo HiVeg™ Agar LES MV1106

M-Endo HiVeg Agar LES is used for enumeration of coliforms in water using a two step membrane filtration method.

Composition ** :

Ingredients	Grams/Litre
HiVeg hydrolysate	3.7
HiVeg peptone	3.7
HiVeg 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

Final pH (at 25°C) 7.2 ± 0.2
** Formula adjusted, standardized to suit performance parameters.

Directions :

Suspend 51 grams in 980 ml distilled water. Heat to boiling to dissolve the medium completely. DO NOT AUTOCLAVE. Cool to 45°C and aseptically add 20 ml of 95% ethanol. Mix and dispense 4 ml amounts into 60 mm petri plates. In large plates, use sufficient medium to give an 1.5 mm depth. DO NOT EXPOSE PLATES TO DIRECT SUNLIGHT.
Caution : Basic fuchsin is a potential carcinogen and care must be taken to avoid inhalation and contamination of the skin.

Principle and Interpretation :

M-Endo HiVeg Agar LES is prepared by completely replacing animal based peptones with vegetable peptones which make the medium free of BSE/TSE risks. M-Endo HiVeg Agar LES is the modification M-Endo Agar LES (Lawrence, Experimental Station) which is formulated according to the formulation of McCarthy, Delaney and Grasso (1) and is used for the enumeration of coliforms in water (2). 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 (3).
HiVeg hydrolysate, HiVeg hydrolysate No.1, HiVeg 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 colouration of the medium. Lactose non-fermenters form colourless colonies.
In the first step of enrichment, the pad is impregnated with Lauryl Tryptose HiVeg Broth (MV080). 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 HiVeg 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 HiVeg Broth (MV080) 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 colonies/100 ml = coliform colonies x 100 / ml of sample filtered

Product Profile :

Vegetable based (Code MV)◉	Animal based (Code M)
MV1106 HiVeg hydrolysate HiVeg peptone HiVeg hydrolysate No. 1 Synthetic detergent No. III	**M1106** Casein enzymic hydrolysate Peptic digest of animal tissue Tryptose Sodium deoxycholate

Recommended for	:	Enumeration of coliforms in water using a two step membrane filtration method.
Reconstitution	:	51.0 g/l
Quantity on preparation (500g)	:	9.80 L
(100g)	:	1.96 L
pH (25°C)	:	7.2 ± 0.2
Supplement	:	95% ethanol
Sterilization	:	Boiling (DO NOT AUTOCLAVE)

Storage : Dry Medium - Below 30°C, Use freshly prepared Medium.

Quality Control :

Appearance of powder
Light purple to purple coloured, homogeneous, free flowing powder.
Gelling
Firm, comparable with 1.5% Agar gel.
Colour and Clarity
Red coloured, slightly opalescent gel forms in petri plates.

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Reaction

Reaction of 5.1% w/v aqueous solution with 2% v/v ethanol is pH 7.2 ± 0.2 at 25°C.

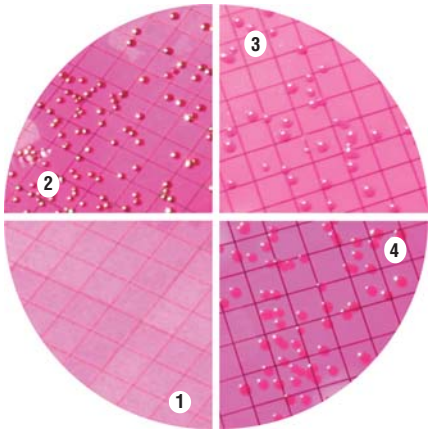
Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 20 - 24 hours.

Organisms (ATCC)	Inoculum (CFU)	Growth	Colour of colony
Enterobacter aerogenes (13048)	10 - 100	luxuriant	red to black with no sheen
Escherichia coli (25922)	10 - 100	luxuriant	red to black with sheen
Salmonella serotype Typhimurium(14028)	10 - 100	luxuriant	colourless to light pink
Salmonella serotype Typhi (6539)	10 - 100	luxuriant	colourless to slightly pink
Staphylococcus aureus (25923)	10 - 100	inhibited	-

References :

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



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1. Control
2. *Escherichia coli*
3. *Salmonella* serotype Typhimurium
4. *Enterobacter aerogenes*