



M-HD Endo Broth w/ BG

M1118

M-HD Endo Broth w/ BG is recommended for the detection of coliform in highly polluted waters using membrane filter technique.

Composition**

Ingredients	Gms / Litre
Casein enzymic hydrolysate	10.000
Peptic digest of animal tissue	10.000
Yeast extract	3.000
Lactose	20.000
Sodium deoxycholate	0.200
Sodium chloride	5.000
Dipotassium phosphate	6.000
Sodium sulphite	2.100
Brilliant green	0.140
Final pH (at 25°C)	7.5±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 56.44 grams in 1000 ml distilled water. Heat if necessary to dissolve the medium completely. DO NOT AUTOCLAVE. Dispense as desired. It is preferable to use the medium on the same day of preparation.

Principle And Interpretation

Coliform bacteria are traditionally described as a group of bacteria that are aerobic and facultatively anaerobic, gram-negative, non spore-forming, rod-shaped, fermenting lactose with gas and acid production at 35°C in 24 to 48 hours (1). The bacteria are classically used as indicators of faecal contamination or water pollution from sewage and thus are of sanitary significance. Coliform bacteria usually originate from the intestinal tract of warm blooded animals and may also originate from waters from wood industry, surfaces of red wood water tanks, biofilms within drinking water distribution systems (2).

M-HD Endo Broth w/ BG is formulated as per Hajna and Damon (3) and is used for detection of coliforms in highly polluted water using membrane filter technique (4). This medium is a modification of M-HD Endo Broth (M1118) with the basic fuchsin in the later being replaced with Brilliant green in the former.

M-HD Endo Broth w/ BG contains casein enzymic hydrolysate, peptic digest of animal tissue and yeast extract as the sources of essential nutrients. Lactose is the fermentable carbohydrate and energy source. Sodium deoxycholate and brilliant green form the selective system against non-coliform bacteria including *Shigella* species (4). Sodium chloride maintains osmotic equilibrium while phosphate buffers the medium. Brilliant green along with sodium sulphite forms the indicator system.

Sterile cotton absorbent pads are saturated with around 2 ml of M-HD Endo Broth w/ BG. Membrane filter through which the test water sample has been passed is aseptically placed on these cotton pads. Following an incubation at 35-37°C for 18-24 hours, lactose-fermenting coliforms will form green colonies with metallic sheen. Non-lactose fermenting coliforms will form colourless colonies.

Quality Control

Appearance

Cream to light green homogeneous free flowing powder

Colour and Clarity of prepared medium

Dark green coloured clear solution without any precipitate

Reaction

Reaction of 5.64% w/v aqueous solution at 25°C. pH : 7.5±0.2

pH

7.30-7.70

Cultural Response

M1118: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Colour of colony (on membrane filter)
Cultural Response			
<i>Escherichia coli</i> ATCC 25922	50-100	luxuriant	green with metallic sheen
<i>Enterobacter aerogenes</i> ATCC 13048	50-100	luxuriant	green
<i>Salmonella Typhi</i> ATCC 6539	50-100	luxuriant	colourless
<i>Staphylococcus aureus</i> ATCC 25923	$\geq 10^3$	inhibited	

Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label.

Reference

1. Dutka B. J., Chau A. S. Y., Coburn J., 1974, Water Res. 8 : 1047-1055
2. Hurst C. J., Knudsen G. R., McInerney M. J., Stetzenbach L. D., Walter M. V. (Eds.) 1997, Manual of Environmental Microbiology, ASM, Washington, D.C.
3. Hajna A. A. and Damon S. R., 1954, Public Health Rep., 69, 58
4. MacFaddin J. F. , 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore

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