Brilliant Green Bile Broth 2%

M121S

Brilliant Green Bile Broth 2% is recommended for the detection and confirmation of coliform bacteria in water, waste water, foods, milk and dairy products.

Composition**

Ingredients	Gms / Litre
Peptic digest of animal tissue	10.000
Lactose	10.000
Ox bile	20.000
Brilliant green	0.0133
Final pH (at 25°C)	7.2±0.1

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 40.01 grams in 1000 ml distilled water. Heat if necessary to dissolve the medium completely. Distribute in fermentation tubes containing inverted Durhams tubes and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Do not autoclave double strength broth.

Note : Where the number of organisms is expected to be low, larger quantities of the sample may be directly added to equal amounts of double strength medium in dilution bottle or flask.

Principle And Interpretation

Brilliant Green Bile Broth 2% is formulated as per American Public Health Association (1, 2, 3) for presumptive identification and confirmation of coliform bacteria (4, 5).

Brilliant green and Ox bile present in the medium inhibit gram-positive bacteria including lactose fermenting Clostridia (5). Production of gas from lactose fermentation is detected by incorporating inverted Durhams tube, indicates a positive evidence of faecal coliforms since nonfaecal coliforms growing in this medium do not produce gas. During examination of water samples, growth from presumptive positive tubes showing gas in Lactose Broth (M026) or Lauryl Tryptose Broth (M080) is inoculated in Brilliant Green Bile Broth 2% wherein gas formation within 48 ± 2 hours confirms the presumptive test (3).

Culture from positive presumptive liquid medium is streaked on Eosin Methylene Blue Agar Plates (M022S). Subsequently colonies form EMB plates are inoculated on Nutrient Agar plates (M561A) and Lactose Broth (M1003S) for further confirmation.

Gram-positive sporeformers may produce gas if the bile or brilliant green inhibition is weakened by food material.

Quality Control

Appearance
Cream to pale green coloured homogeneous free flowing powder

Colour and Clarity of prepared medium
Emerald green coloured clear solution without any precipitate.

Reaction
Reaction of 4.0% w/v aqueous solution at 25°C. pH : 7.2±0.1

pH
7.10-7.30

Cultural Response
Cultural characteristics observed after an incubation at 35-37°C for 18- 48 hours.

Cultural Response

Organism	Inoculum (CFU)	Growth	Gas
Cultural Response			
Bacillus cereus ATCC 10876	>=10^3	inhibited	
Enterobacter aerogenes ATCC 13048	50-100	luxuriant	Positive reaction
Escherichia coli ATCC 25922	50-100	luxuriant	Positive reaction
Enterococcus faecalis ATCC 19433	50-100	none-poor	Negative reaction
Staphylococcus aureus ATCC 25923	>=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. Greenberg A. E., Trussell R. R. and Clesceri L. S. (Eds.), 1985, Standard Methods for the Examination of Water and Wastewater, 16th ed., A.P.H.A., Washington D.C.
2. Speck M. (Ed.), 1985, Compendium of Methods for the Microbiological Examination of Foods, 2nd ed., A.P.H.A., Washington D.C.
3. Richardson G. (Ed.), 1985, Standard Methods for the Examination of Dairy Products, 15th ed, A.P.H.A., Washington, D.C.
4. McCrady and Langerin, 1932, J. Dairy Science, 15:321.
5. McCrady, 1937, Am. J. Publ. Health, 27:1243.

Revision : 2 / 2015