Technical Data

Brilliant Green Bile HiCynth™ Broth 2%

MCD121

Brilliant Green Bile HiCynth™ 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
HiCynth™ Peptone No.3*	28.000
Lactose	10.000
Synthetic detergent	2.000
Brilliant green	0.0133
Final pH (at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters
*Chemically defined peptone

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. For preparation of double strength it is recommended to heat the dissolved broth (80.02 grams per litre) at 100°C for 30 minutes.

Principle And Interpretation

Brilliant Green Bile Broth 2% is one of the most widely used medium for the detection of coliform bacteria in water, wastewater, foods, and milk and dairy products. This medium is formulated as per APHA (1, 2, 3) for the presumptive identification and confirmation of coliform bacteria (4, 5). This medium is also recommended by the ISO Committee for enumeration of coliforms by most probable number technique (6). Brilliant Green Bile HiCynth™ Broth 2% is the modification of the same using chemically defined peptone free from animal and vegetable peptones to avoid BSE/TSE risks associated with animal peptones.

HiCynth™ Peptone No.3 serves as a source of essential carbon and nitrogen nutrients, long chain amino acids, vitamins and other growth factors. Lactose is the fermentable carbohydrate. Synthetic detergent inhibits gram-positive bacteria whereas the gram-negative bacteria are inhibited by brilliant green. Production of gas from lactose fermentation is detected by incorporating inverted Durham's tube, which indicates the positive evidence of faecal coliform since non faecal coliforms growing in this medium do not produce gas. Further gas production in EC broth at 45°C used as a confirmation of faecal coliform. Gram-positive spore formers may produce gas if the bile or brilliant green inhibition is weakened by reaction with food material.

During examination of water samples, growth from presumptive positive tubes showing gas in Lactose Broth or Lauryl Tryptose Broth is inoculated in Brilliant Green Bile Broth 2%. Gas formation within 48 ± 2 hours confirms the presumptive test (1).

Quality Control

Appearance
Cream to pale green 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±0.2

pH
7.00-7.40

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

Cultural Response

Organism	Inoculum (CFU)	Growth	Gas

Bacillus cereus ATCC 10876	>=10^3	inhibited	
Escherichia coli ATCC 25922	50-100	good-luxuriant	positive reaction
Enterobacter aerogenes ATCC 13048	50-100	good-luxuriant	positive reaction
Enterococcus faecalis ATCC 29212	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., Eaton A. D. and Clesceri L. S., (Eds.), 2012, Standard Methods for the Examination of Water and Wastewater, 22nd ed., APHA, Washington, D.C.
2. Downes F. P. and Ito K. (Eds.) 2001, Compendium of Methods for the Microbiological Examination of Food. 4th Ed, APHA, Washington, D.C.
3. Richardson G., (Ed.), 1985, Standard Methods for the Examination of Dairy Products, 15th Ed, APHA, Washington, D.C.
4. McCrady and Langerin, 1932, J. Dairy Science, 15:321.
5. McCrady, 1937, Am. J. Publ. Health, 27:1243.
6. International Organization for Standardization (ISO), 1991, Draft ISO/DIS 4831.

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