



Technical Data

Brilliant Green Bile Broth

M121I

Intended Use:

Recommended for isolation and cultivation of coliform organisms from cream, yogurt and raw milk. The composition and performance criteria of this medium are as per the specifications laid down in ISO 4831:2006.

Composition**

Ingredients	Gms / Litre
Tryptone	10.000
Lactose monohydrate	10.000
Dehydrated bile	20.000
Brilliant green	0.0133
Final pH (at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 39.51 grams (the equivalent weight of dehydrated medium per liter) in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense the medium in quantities of 10ml in test tubes of approximately 16mm x 160mm containing Durham tubes. Sterilize in an autoclave set at 121°C for 15 minutes. Cool to 45-50°C.

Note: The Durham tube shall not contain air bubbles after sterilization.

Principle And Interpretation

Brilliant Green Bile Broth is formulated as per ISO 4831:2006(E) for confirmation of coliform bacteria (1) present in food samples or environmental samples in the area of food handling or food sampling.

Brilliant green and Dehydrated bile present in the medium inhibit gram-positive bacteria including lactose fermenting *Clostridia* (4). Production of gas from lactose fermentation is detected by incorporating inverted Durham's tube, indicates a positive evidence of faecal coliforms since nonfaecal coliforms growing in this medium do not produce gas. During examination of food samples or environmental samples, growth from presumptive positive tubes showing gas in Lauryl Tryptose Broth (M080) is inoculated in Brilliant Green Bile Broth wherein gas formation within 48 ± 2 hours confirms the presumptive test (1). Gram-positive spore-formers may produce gas if the bile or brilliant green inhibition is weakened by food material.

Type of specimen

Food samples

Specimen Collection and Handling:

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (5).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations :

1.This medium is general purpose medium and may not support the growth of fastidious organisms.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

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 3.95% 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.

Organism	Inoculum (CFU)	Growth	Gas
<i>Bacillus cereus</i> ATCC 10876	≥10 ⁴	inhibited	
<i>Escherichia coli</i> ATCC 25922 (00013*)	50-100	good-luxuriant	positive reaction
<i>Escherichia coli</i> ATCC 8739 (00012*)	50-100	good-luxuriant	positive reaction
<i>Enterobacter aerogenes</i> ATCC 13048 (00175*)	50-100	good-luxuriant	positive reaction
<i>Citrobacter freundii</i> ATCC 43864 (00006*)	50-100	good-luxuriant	positive reaction
<i>Enterococcus faecalis</i> ATCC 29212 (00087*)	50-100	none-poor	negative reaction
<i>Enterococcus faecalis</i> ATCC 19433 (00009*)	50-100	none-poor	negative reaction
<i>Staphylococcus aureus</i> subsp. <i>aureus</i> ATCC 25923 (00034*)	≥10 ⁴	inhibited	

Key : * - Corresponding WDCM numbers

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (2,3).

Reference

1. International Standard, ISO 4831:2006 (E). Microbiology of food and animal feeding stuff- Horizontal method for the detection and enumeration of coliforms- Most Probable number technique.
2. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
3. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock, D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
4. McCrady and Langerin, 1932, J. Dairy Science, 15:321.
5. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.

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Disclaimer :

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