

Instructions for Use

REF - **BM4181**

Brain Heart Infusion Broth with 10% Glycerol

Bottled Supplement

1. Intended Use

For *in vitro* diagnostic use only. BM4181 Brain Heart Infusion Broth with 10% Glycerol is suitable for preparing suspensions of bacterial and fungal isolates for frozen storage.

2. Composition*

Ingredient	g/L
Brain heart infusion solids	17.5
Enzymatic Digest of Casein	10.0
Dextrose	2.0
Sodium Chloride	5.0
Disodium Phosphate	2.5
Glycerol	100ml

*Adjusted/supplemented as needed to meet performance requirements

3. Summary and Explanation

Rosenow ⁽¹⁾ developed a medium that could be used to cultivate streptococci using a dextrose broth and brain tissue in 1919. Hayden ⁽²⁾ modified Rosenow's formula while working with dental pathogens in 1923. Modern Brain Heart Infusion Broth typically uses a solid infusion from porcine brain and heart rather than calf brain tissue, and uses disodium phosphate as a buffer, rather than the calcium carbonate used by Rosenow and Haden ^(1,2). It is a nutrient-rich medium and can therefore be used to culture a variety of fastidious organisms, including *streptococci*, *pneumococci*, and *meningococci*, which can be challenging to grow. This product has 10% glycerol added to the Brain Heart Infusion Broth.

4. Principle

The nitrogen, vitamin and carbon sources are supplied by the Brain-Heart infusion solids and peptone. Glucose serves as the carbohydrate source and sodium chloride aids in maintaining the osmotic balance. A phosphate buffer, disodium hydrogen phosphate, is incorporated to help neutralize any acids produced because of glucose utilization and thus maintain viability of the organisms. This formulation also has added glycerol to act as cryopreservative so the medium can be for the long-term frozen storage of microorganisms.

5. Physical Characteristics

	Appearance and Colour	pH
Medium	Light brown liquid	7.4 ± 0.2

6. Materials Provided

BM4181 can be provided in volumes of 2ml in packs of 50 x glass bijous; each bottle is ink-jet printed with (abbreviated) product name, product code, lot number and expiry date.

7. Materials Needed but not Provided

Standard microbiological laboratory materials e.g., sterile loops or swabs, collection containers, incubators, media for isolation and quality control organisms.

8. Specimens

Brain Heart Infusion Broth with 10% Glycerol is used for preparing microorganism isolates for long term frozen storage rather than as a primary isolation medium. Pure colonies of isolates from clinical specimens or food samples can be used to inoculate this product.

9. Test Procedures and Interpretation of results

Allow media to come to room temperature.

Use a sterile loop or swab to pick sufficient colonies of a pure culture of the isolate and inoculate the medium to achieve a heavily turbid suspension.

Transfer or aliquot the inoculated medium to a suitable cryovial and freeze at less than -70°C for the stable long-term storage of the isolate.

After storage, slowly defrost and inoculate to a suitable culture medium depending on the nutritional and growth requirements of the microorganism.

11. Quality Control

Organism	Incubation	Result (Specificity)
<i>E. coli</i> (NCTC 12241) <i>S. aureus</i> (NCTC 12981)	Incubate aerobically at 37 ± 1°C for 24-48 hours Incubate aerobically at 37 ± 1°C for 24-48 hours Subculture 1µl onto E&O PP0120 Columbia Blood agar and incubate plates anaerobically at 37 ± 1°C in for 18-24 hours.	Broth: Good turbidity. Subplate: Grey colonies Broth: Good turbidity. Subplate: White colonies

It is the responsibility of the user to perform Quality Control testing taking into consideration the intended use of the medium and in agreement with any local relevant guidelines (e.g., frequency, strains used, atmosphere, incubation temperature).

12. Performance

To fully verify BM4181 Brain Heart Infusion Broth with 10% Glycerol performance, samples were tested to assess colony morphology, recovery level and turbidity. Samples were inoculated and incubated at 37 ± 1°C for 18-24 hours. After, 1µl was subcultured onto E&O PP0120 Columbia Blood agar and plates incubated anaerobically at 37 ± 1°C in for 18-24 hours. All samples grew and showed good recovery and the correct morphology of the required test organisms: *Escherichia coli* (NCTC 12241) and *Staphylococcus aureus* (NCTC 12981). Therefore, it can be concluded that BM4181 Brain Heart Infusion Broth with 10% Glycerol, meets performance criteria when used according to manufacturer's instructions. Trend analysis data available upon request.

13. Limitations of the Media

- Acid producing organisms may overwhelm the buffering system, resulting in non-viable cultures of isolates if inoculated to the medium and incubated prior to transfer to frozen storage.
- Due to natural variation, some strains may grow poorly on this medium.

14. Precautions and Warnings

This product is considered non-hazardous under CLP regulations. Wear such PPE as recommended by laboratory COSSH assessment. During and after use, always handle all materials in a manner conforming to Good Laboratory Practices and consider that material under test should be regarded as a potential biohazard if mishandled.

Refer to Bottled Media Material Safety Data Sheet.

15. Storage conditions and Shelf life

Store BM4181 Brain Heart Infusion Broth with 10% Glycerol in the original bottle with the lid tightly closed at between 15 and 30°C in the dark. Kept under these conditions it may be used up to date of expiry shown on product label.

Dispose of in accordance with local and national authority requirements. Do not use media if it is contaminated, discoloured or out of date. Additionally, do not use medium if it has been stored inappropriately or the packaging has been damaged.

16. References

- Rosenow, E. (1919) Studies on Elective Localization Focal Infection with Special Reference to Oral Sepsis'. J. Dent. Research, 1, pp.205-249.
- Hayden, R. (1923) Elective localization in the eye of bacteria from infected teeth. Arch. Int. Med., 32, pp. 828-849.

Version History*

001 27/01/23 - New Document Created

*Note: minor typographical, grammatical, and formatting changes are not included in the revision history.



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TABLE OF APPLICABLE SYMBOLS

REF Catalogue number	LOT Batch code	IVD In vitro Diagnostic Medical Device	Manufacturer	Use by
Temperature limitation	Contents sufficient for <n> tests	Consult Instructions for Use	Keep away from direct light	Store in a dry place