

Brain Heart Infusion (Broth Media)

Brain Heart Infusion • Brain Heart Infusion with Supplements • Brain Heart Infusion without Dextrose • Brain Heart Infusion Broth, Modified

Intended Use

Brain Heart Infusion (BHI) is a general-purpose liquid medium used in the cultivation of fastidious and nonfastidious microorganisms, including aerobic and anaerobic bacteria, from a variety of clinical and nonclinical materials. It serves as a base for supplemented media containing 0.1% agar, Fildes enrichment or 6.5% sodium chloride. A supplemented pre-reduced formulation in tubes is especially recommended for the cultivation of anaerobes.

Summary and Explanation

Rosenow described brain-heart infusion broth prepared by adding pieces of brain tissue to meat infusion or beef extract-dextrose broth.¹ A variation of this medium appeared for many years in the National Formulary.² The current formulation is similar to the NF Brain Heart Infusion Broth, but the brain infusion component is composed of solids resulting from the drying of the liquid material and the heart infusion component has been replaced with a peptone of partially digested animal tissue.

BHI broth is used for the cultivation of a wide variety of microorganisms, including bacteria, yeasts and molds.

BHI broth, 0.5 mL per tube, is used for the cultivation of bacteria employed in the preparation of inocula for microdilution minimal inhibitory concentration (MIC) and identification (ID) test panels. When a large number of cells are inoculated into the small volume of broth, a bacterial culture rapidly reaches its stationary phase of growth.³ The medium is also used in 5-mL amounts per tube for the preparation of inocula in antimicrobial susceptibility test procedures. This volume and the 8-mL tubes also can be used for general purposes.

Fildes enrichment may be incorporated for the growth of fastidious organisms. With the addition of 0.1% agar, the medium is used for the cultivation of anaerobes. The medium pre-reduced in Hungate tubes is recommended for the cultivation of anaerobic microorganisms, particularly obligate anaerobes.

The broth medium that contains 6.5% sodium chloride is used to differentiate the enterococci from nonenterococcal group D streptococci by the 6.5% salt tolerance test.⁴

Brain Heart Infusion without Dextrose is a basal medium used with carbohydrates for fermentation studies.

Brain Heart Infusion, Modified differs from other formulations by the quantities of the ingredients and the substitution of pancreatic digest of casein for pancreatic digest of gelatin.

Principles of the Procedure

BHI Broth is a nutritious, buffered culture medium that contains infusions of brain and heart tissue and peptones to supply protein and other nutrients necessary to support the growth of fastidious and nonfastidious microorganisms. In the formulation containing 6.5% sodium chloride, the salt acts as a differential and/or selective agent by interfering with membrane permeability and osmotic and electrokinetic equilibria in salt-intolerant organisms. Fildes enrichment (peptic digest of sheep blood) is incorporated into one tubed formulation for the cultivation of fastidious microorganisms, such as *Haemophilus influenzae*.^{5,6} The addition of 0.1% agar aids in the cultivation of anaerobic microorganisms because its consistency yields conditions of reduced oxygen tension. The pre-reduced medium in Hungate tubes is based on Hungate methods of culturing anaerobic microorganisms outside of an anaerobic chamber.⁷ The tubes provide a reduced medium in

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User Quality Control

NOTE: Differences in the Identity Specifications and Cultural Response testing for media offered as both **Difco™** and **BBL™** brands may reflect differences in the development and testing of media for industrial and clinical applications, per the referenced publications.

Identity Specifications

Bacto™ Brain Heart Infusion

Dehydrated Appearance: Light tan, free-flowing, homogeneous.
 Solution: 3.7% solution, soluble in purified water upon boiling. Solution is light to medium amber, clear.
 Prepared Appearance: Light to medium amber, clear.
 Reaction of 3.7% Solution at 25°C: pH 7.4 ± 0.2

Difco™ Brain Heart Infusion without Dextrose

Dehydrated Appearance: Light tan, free-flowing, homogeneous.
 Solution: 3.5% solution, soluble in purified water upon boiling. Solution is light to medium amber, clear.
 Prepared Appearance: Light to medium amber, clear.
 Reaction of 3.5% Solution at 25°C: pH 7.4 ± 0.2

Cultural Response

Bacto™ Brain Heart Infusion or Difco™ Brain Heart Infusion without Dextrose

Prepare the medium per label directions. Inoculate and incubate at 35 ± 2°C for 18-48 hours.

| ORGANISM | ATCC™ | INOCULUM CFU | RECOVERY |
|---------------------------------|-------|----------------------------------|----------|
| <i>Neisseria meningitidis</i> | 13090 | 10 ² -10 ³ | Good |
| <i>Streptococcus pneumoniae</i> | 6305 | 10 ² -10 ³ | Good |
| <i>Streptococcus pyogenes</i> | 19615 | 10 ² -10 ³ | Good |

Identity Specifications

BBL™ Brain Heart Infusion

Dehydrated Appearance: Fine, homogeneous, free of extraneous material.
 Solution: 3.7% solution, soluble in purified water upon boiling. Solution is light to medium, yellow to tan, clear to slightly hazy.
 Prepared Appearance: Light to medium, yellow to tan, clear to slightly hazy.
 Reaction of 3.7% Solution at 25°C: pH 7.4 ± 0.2

BBL™ Brain Heart Infusion Broth, Modified

Dehydrated Appearance: Fine, homogeneous, free of extraneous material.
 Solution: 3.8% solution, soluble in purified water upon boiling. Solution is light to medium, yellow to tan, clear to slightly hazy.
 Prepared Appearance: Light to medium, yellow to tan, clear to slightly hazy.
 Reaction of 3.7% Solution at 25°C: pH 7.4 ± 0.2

Cultural Response

BBL™ Brain Heart Infusion or BBL™ Brain Heart Infusion Broth, Modified

Prepare the medium per label directions. Inoculate and incubate at 35 ± 2°C under appropriate atmospheric conditions for 7 days (incubate *C. albicans* at 20-27°C).

| ORGANISM | ATCC™ | INOCULUM CFU | RECOVERY BHI | RECOVERY BHI, MODIFIED |
|---------------------------------|-------|------------------|--------------|------------------------|
| <i>Bacteroides fragilis</i> | 25285 | ≤10 ⁴ | Good | Good |
| <i>Candida albicans</i> | 10231 | ≤10 ³ | Good | Good |
| <i>Enterococcus faecalis</i> | 29212 | ≤10 ³ | Good | N/A |
| <i>Neisseria meningitidis</i> | 13090 | ≤10 ³ | Good | Good |
| <i>Streptococcus pneumoniae</i> | 6305 | ≤10 ³ | Good | Good |
| <i>Streptococcus pyogenes</i> | 19615 | ≤10 ³ | Good | Good |

a self-contained, anaerobic tube sealed using a Hungate screw cap. The cap contains a butyl rubber septum stopper that permits inoculation and incubation without exposing the medium to air.

Formulae

Bacto™ Brain Heart Infusion

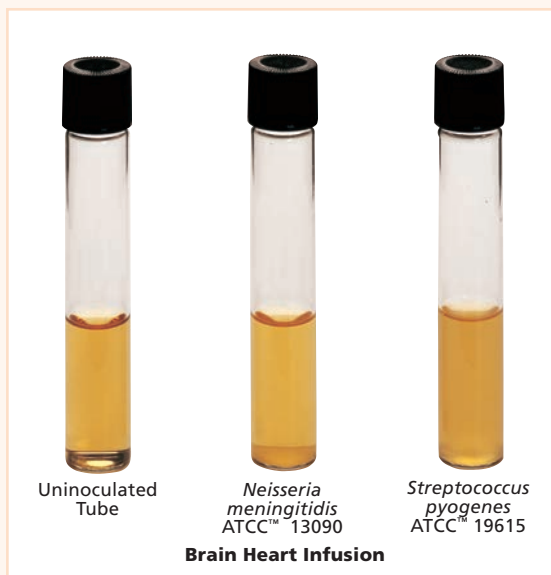
Approximate Formula* Per Liter
 Calf Brains, Infusion from 200 g 7.7 g
 Beef Heart, Infusion from 250 g 9.8 g
 Proteose Peptone 10.0 g
 Dextrose 2.0 g
 Sodium Chloride 5.0 g
 Disodium Phosphate 2.5 g

BBL™ Brain Heart Infusion

Approximate Formula* Per Liter
 Brain Heart, Infusion from (solids) 6.0 g
 Peptic Digest of Animal Tissue 6.0 g
 Pancreatic Digest of Gelatin 14.5 g
 Dextrose 3.0 g
 Sodium Chloride 5.0 g
 Disodium Phosphate 2.5 g

Difco™ Brain Heart Infusion without Dextrose

Approximate Formula* Per Liter
 Calf Brains, Infusion from 200 g 7.7 g
 Beef Heart, Infusion from 250 g 9.8 g
 Proteose Peptone 10.0 g
 Sodium Chloride 5.0 g
 Disodium Phosphate 2.5 g



BBL™ Brain Heart Infusion Broth, Modified

| | |
|-------------------------------------------|--------|
| Approximate Formula* Per Liter | |
| Brain Heart, Infusion from (solids) | 3.5 g |
| Peptic Digest of Animal Tissue | 15.0 g |
| Pancreatic Digest of Casein | 10.0 g |
| Dextrose | 2.0 g |
| Sodium Chloride | 5.0 g |
| Disodium Phosphate | 2.5 g |

*Adjusted and/or supplemented as required to meet performance criteria.

Directions for Preparation from Dehydrated Product

- Suspend the powder in 1 L of purified water:
 - Bacto™ Brain Heart Infusion – 37 g;
 - BBL™ Brain Heart Infusion – 37 g;
 - Difco™ Brain Heart Infusion without Dextrose – 35 g;
 - BBL™ Brain Heart Infusion Broth, Modified – 38 g.
 Mix thoroughly.
- Heat with frequent agitation and boil for 1 minute to completely dissolve the powder.
- Autoclave at 121°C for 15 minutes.
- Test samples of the finished product for performance using stable, typical control cultures.

Procedure

With liquid specimens, tubed media should be inoculated with 1-2 drops of the specimen using a sterile pipette. Swab specimens may be inserted into broth after inoculation of plated media.

Liquid media for anaerobic incubation should be reduced prior to inoculation by placing the tubes, with caps loosened, under anaerobic conditions for 18-24 hours prior to use. An efficient and easy way to obtain suitable anaerobic conditions is through the use of BBL™ GasPak™ EZ anaerobic systems or an alternative anaerobic system. Alternatively, liquid media may be reduced immediately prior to use by boiling with caps loosened and cooling with tightened caps to room temperature before inoculation.

Before inoculating Hungate tubes, disinfect the septum of the cap. To inoculate, insert needle of syringe containing specimen through the septum and inject the specimen into the medium. Withdraw the needle slowly to avoid introducing air into the tube.

For use in antimicrobial susceptibility testing, refer to appropriate references.⁸⁻¹⁰

Expected Results

Growth in the tubes is indicated by the presence of turbidity compared to an uninoculated control. If growth appears, cultures should be examined by Gram stain and subcultured onto appropriate media; e.g., a Trypticase™ Soy Agar with 5% Sheep Blood and/or Chocolate II Agar plate, EMB Agar or MacConkey II Agar plate. If anaerobes are suspected, subcultures should be incubated anaerobically, as in a GasPak EZ anaerobic system.

Enterococci will grow in the 6.5% NaCl broth within 24-48 hours. Nonenterococcal group D streptococci fail to grow in the medium after 48 hours of incubation.³

References

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- National Committee for Clinical Laboratory Standards. 2000. Approved Standard M2-A7, Performance standards for antimicrobial disk susceptibility tests, 7th ed. NCCLS, Wayne, Pa.

Availability

Bacto™ Brain Heart Infusion

AOAC BAM CCAM CMPH COMPF ISO MCM7 NCCLS
SMD SMWW USDA

| | | |
|----------|--------|--------------------|
| Cat. No. | 237400 | Dehydrated – 100 g |
| | 237500 | Dehydrated – 500 g |
| | 237200 | Dehydrated – 2 kg |
| | 237300 | Dehydrated – 10 kg |

BBL™ Brain Heart Infusion

AOAC BAM CCAM CMPH COMPF ISO MCM7 NCCLS
SMD SMWW USDA

| | | |
|----------|--------|-------------------------------------------------------------|
| Cat. No. | 211059 | Dehydrated – 500 g |
| | 211060 | Dehydrated – 5 lb (2.3 kg) |
| | 211061 | Dehydrated – 25 lb (11.3 kg) |
| | 221778 | Prepared Tubes, 0.5 mL (K Tubes) – Ctn. of 100 |
| | 221812 | Prepared Tubes, 5 mL (K Tubes) – Pkg. of 10 |
| | 221813 | Prepared Tubes, 5 mL (K Tubes) – Ctn. of 100 |
| | 220837 | Prepared Tubes, 8 mL (K Tubes) – Ctn. of 100 |
| | 297312 | Prepared Bottles, 100 mL – 1 bottle |
| | 296299 | Prepared Bottles, 400 mL – 1 bottle |
| | 297304 | Prepared Tubes, Pre-reduced (with Hungate Cap) – Pkg. of 10 |

BBL™ Brain Heart Infusion with 6.5% Sodium Chloride

| | | |
|----------|--------|-----------------------------|
| Cat. No. | 221785 | Prepared Tubes – Pkg. of 10 |
|----------|--------|-----------------------------|

BBL™ Brain Heart Infusion with 0.1% Agar

| | | |
|----------|--------|-----------------------------------------------|
| Cat. No. | 297640 | Prepared Tubes, 10 mL (D Tubes) – Ctn. of 100 |
|----------|--------|-----------------------------------------------|

BBL™ Brain Heart Infusion with Fildes Enrichment

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|----------|--------|-----------------------------------------------|
| Cat. No. | 297782 | Prepared Tubes, 5 mL (K Tubes) – Ctn. of 100* |
| | 297200 | Prepared Tubes, 9 mL (K Tubes) – Pkg. of 10* |

Difco™ Brain Heart Infusion without Dextrose

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|----------|--------|--------------------|
| Cat. No. | 250220 | Dehydrated – 10 kg |
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BBL™ Brain Heart Infusion Broth, Modified

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|----------|--------|--------------------|
| Cat. No. | 299070 | Dehydrated – 500 g |
|----------|--------|--------------------|

BBL™ Fildes Enrichment

| | | |
|----------|--------|----------------------------------------------|
| Cat. No. | 211866 | Prepared Tubes, 5 mL (K Tubes) – Pkg. of 10* |
| | 220810 | Prepared Bottles, 100 mL – 1 bottle* |
| | 220811 | Prepared Bottles, 100 mL – Pkg. of 6* |

*Store at 2-8°C.

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