



# BBL™ Brain Heart Infusion Agar (Deep Fill)

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## QUALITY CONTROL PROCEDURES

### I INTRODUCTION

Brain Heart Infusion Agar is a general purpose culture medium which supports the growth of a wide variety of bacterial and fungal species including many types of pathogens, such as streptococci and pneumococci.

### II PERFORMANCE TEST PROCEDURE

- Inoculate representative samples with the cultures diluted to contain 50–100 CFU per 0.1 mL.
  - Add 0.1 mL of the appropriate dilution to each plate and spread-inoculate using a sterile glass spreader.
  - Incubate the *Candida* and *Shigella* strains at  $35 \pm 2^\circ\text{C}$  in an aerobic atmosphere and the *Streptococcus* strain at  $35 \pm 2^\circ\text{C}$  in an aerobic atmosphere supplemented with 3–5% carbon dioxide.
- Examine plates after 18–24 and 48–72 h for amount of growth, colony size and pigmentation.
- Expected Results

Organisms	ATCC™	Recovery
* <i>Candida albicans</i>	10231	Moderate to heavy growth by 48 to 72 h. Colonies medium to large and white.
* <i>Shigella flexneri</i>	12022	Moderate to heavy growth by 48 to 72 h. Colonies medium to large, grayish-white, translucent, slightly convex and may be mucoid.
* <i>Streptococcus pneumoniae</i>	6305	Moderate to heavy growth. Colonies small, flattened, circular and translucent.

\*Recommended organism strain for User Quality Control.

### III ADDITIONAL QUALITY CONTROL

- Examine plates as described under "Product Deterioration."
- Visually examine representative plates to assure that any existing physical defects will not interfere with use.
- Determine the pH potentiometrically at room temperature for adherence to the specification of  $7.4 \pm 0.2$ .
- Note the firmness of plates during the inoculation procedure.
- Incubate uninoculated representative plates at  $35 \pm 2^\circ\text{C}$  for 72 h and examine for microbial contamination.

## PRODUCT INFORMATION

### IV INTENDED USE

Brain Heart Infusion (BHI) Agar is a general purpose medium suitable for the cultivation of a wide variety of organism types, including bacteria, yeasts and molds. The plates are deep-filled to reduce the effects of drying during prolonged incubation.

### V SUMMARY AND EXPLANATION

In the early years of bacteriology, meat infusions were utilized as the growth-supporting components in a large number of culture media. Although they were cumbersome to prepare, lacked consistency from batch to batch and were undefined as to their nutritive content, they enabled the cultivation of microorganisms in both solid and liquid media. As the state-of-the-art in enzymology and chemistry advanced, methods were developed for the preparation of peptones which were the result of enzymatic or acid hydrolysis of animal tissues or products and vegetable substances. These peptones currently are the major nutritional additives to culture media formulations, but infusions are still utilized in specific media.

Brain Heart Infusion Agar is one formulation in which meat infusion is used, although, unlike in the earlier days, the infusion components are solids resulting from the drying of the liquid infusion material rather than the liquid components themselves. Two peptones are also included as sources of nutrients.

This medium has proven to be effective in the cultivation of a wide variety of microorganisms, including many types of pathogens. It has served as the base medium for new culture media formulations when supplemented with sheep blood, cycloheximide, chloramphenicol, and/or gentamicin. BHI Agar currently is recommended as a general medium for aerobic bacteriology and for the primary recovery of fungi from clinical specimens.<sup>1,2</sup>

### VI PRINCIPLES OF THE PROCEDURE

BHI Agar derives its nutrients from the brain heart infusion, peptone and dextrose components. The peptones and infusion are sources of organic nitrogen, carbon, sulfur, vitamins and trace substances. Dextrose is the carbohydrate source which microorganisms utilize by fermentative action. The medium is buffered through the use of disodium phosphate.

### VII REAGENTS

#### Brain Heart Infusion Agar

Approximate Formula\* Per Liter Purified Water

Brain Heart, Infusion from (solids).....	8.0 g	Dextrose .....	2.0 g
Peptic Digest of Animal Tissue .....	5.0 g	Disodium Phosphate .....	2.5 g
Pancreatic Digest of Casein .....	16.0 g	Agar .....	13.5 g
Sodium Chloride .....	5.0 g		

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

#### Warnings and Precautions: For *in vitro* Diagnostic Use.

If excessive moisture is observed, invert the bottom over an off-set lid and allow to air dry in order to prevent formation of a seal between the top and bottom of the plate during incubation.

Pathogenic microorganisms, including hepatitis viruses and Human Immunodeficiency Virus, may be present in clinical specimens. "Standard Precautions"<sup>3-6</sup> and institutional guidelines should be followed in handling all items contaminated with

blood and other body fluids. After use, prepared plates, specimen containers and other contaminated materials must be sterilized by autoclaving before discarding.

**Storage Instructions:** On receipt, store plates in the dark at 2–8°C. Avoid freezing and overheating. Do not open until ready to use. Minimize exposure to light. Prepared plates stored in their original sleeve wrapping at 2–8°C until just prior to use may be inoculated up to the expiration date and incubated for recommended incubation times. Allow the medium to warm to room temperature before inoculation.

**Product Deterioration:** Do not use plates if they show evidence of microbial contamination, discoloration, drying, cracking or other signs of deterioration.

## VIII SPECIMEN COLLECTION AND HANDLING

Specimens suitable for culture may be handled using various techniques. For detailed information, consult appropriate texts.<sup>7,8</sup> Specimens should be obtained before antimicrobial therapy has been administered. Provision must be made for prompt delivery to the laboratory.

## IX PROCEDURE

**Material Provided:** Brain Heart Infusion Agar (Deep Fill)

**Materials Required But Not Provided:** Ancillary culture media, reagents, quality control organisms and laboratory equipment as required.

**Test Procedure:** Observe aseptic techniques.

The agar surface should be smooth and moist, but without excessive moisture.

Streak the specimen as soon as possible after it is received in the laboratory. The streak plate is used primarily to isolate pure cultures from specimens containing mixed flora.

Alternatively, if material is being cultured directly from a swab, roll the swab over a small area of the surface at the edge; then streak from this inoculated area.

Since many pathogens require carbon dioxide on primary isolation, plates may be incubated in an atmosphere containing approximately 5% CO<sub>2</sub>.

Incubate plates at 35 ± 2°C for 18–72 h.

**User Quality Control:** See “Quality Control Procedures.”

Quality Control requirements must be performed in accordance with applicable local, state and/or federal regulations or accreditation requirements and your laboratory's standard Quality Control procedures. It is recommended that the user refer to pertinent CLSI (formerly NCCLS) guidance and CLIA regulations for appropriate Quality Control practices.

## X RESULTS

After incubation most plates will show an area of confluent growth. Because the streaking procedure is, in effect, a “dilution” technique, diminishing numbers of microorganisms are deposited on the streaked areas. Consequently, one or more of these areas should exhibit isolated colonies of the organisms contained in the specimen. Further, growth of each organism may be semi-quantitatively scored on the basis of growth in each of the streaked areas.

## XI LIMITATIONS OF THE PROCEDURE

For identification, organisms must be in pure culture. Morphological, biochemical, and/or serological tests should be performed for final identification. Consult appropriate texts for detailed information and recommended procedures.<sup>7-12</sup>

## XII AVAILABILITY

Cat. No.	Description
221569	BBL™ Brain Heart Infusion Agar (Deep Fill), Pkg. of 20 plates
221570	BBL™ Brain Heart Infusion Agar (Deep Fill), Ctn. of 100 plates

## XIII REFERENCES

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