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BRAIN HEART INFUSION AGAR

Brain Heart Infusion (BHI) Agar is a general-purpose medical laboratory medium suitable for the cultivation of a wide variety of organism types, including bacteria, yeasts and molds.

REF: BS.1/BI01.100.0100 100 Gram REF: BS.1/BI01.250.01250 250 Gram REF: BS.1/BI01.500.0500 500 Gram

CLINICAL SIGNIFICANCE (1)

Brain Heart Infusion (BHI) Agar is highly nutritious and can support luxuriant growth of wide variety of microorganisms, in particular to culture streptococci, pneumococci and meningococci. It is a general purpose medium used for primary isolation of aerobic bacteria from clinical specimens.

METHOD PRINCIPLE (2-3)

BHI Agar is often used in food safety, water safety, and antibiotic sensitivity tests. It can be further enriched by the addition of blood or rendered selective by adding different antibiotics. Addition of 50 mg/l chloramphenicol or 40 mg/l streptomycin or a mixture of 50 mg/l gentamicin and 50 mg/l chloramphenicol along with 5-10% sterile defibrinated blood is often recommended for inhibition of bacteria and isolation of pathogenic systemic fungi. A mixture of cycloheximide (0.5 g/l) and chloramphenicol (0.05 g/l) is also used for selective isolation of pathogenic fungi (incubation at 25-30°C for 1-2 weeks). Some fungi may be inhibited on this medium with 10% sheep blood, gentamicin and chloramphenicol.

MEDIA COMPOSITION

BHI is made by combining an infusion from boiled bovine or porcine heart and brain with a variety of other nutrients. Proteose peptone and infusions used in the media serves as sources of carbon, nitrogen, vitamins, amino acids, along with essential growth factors. Dextrose is the energy source. Sodium chloride maintains the osmotic equilibrium of the medium while disodium phosphate buffers the medium. Defibrinated sheep blood added to the basal medium provides essential growth factors for the more fastidious fungal organisms.

Item	Concentration %
Brain infusion solids	24.04
Beef heart infusion solids	9.62
Proteose peptone	19.23
Sodium chloride	9.62
Glucose	3.85
Disodium phosphate	4.81
Agar	28.85

PRECAUTIONS AND WARNINGS

Media to be handled by entitled and professionally educated person. Do not ingest or inhale.

Good Laboratories practices using appropriate precautions should be followed in:

- Wearing personnel protective equipment (overall, gloves, glasses,).
- Do not pipette by mouth.
- In case of contact with eyes or skin; rinse immediately with plenty of soap and water. In case of severe injuries; seek medical advice immediately.
- Respect country requirement for waste disposal.
 \$56: dispose of this material and its container at hazardous or Special waste collection point.

S57: use appropriate container to avoid environmental contamination.

\$61: avoid release in environment.

For further information, refer to the BHI agar material safety data sheet.

MEDIA PREPARATION, STORAGE AND STABILITY (2)

BioScien BHI Agar dehydrated media are stable until expiration date stated on label when properly stored 10-30°C. Hydrated BHI Agar media is prepared by suspend 52 grams of the medium in one liter of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Sterilize in autoclave at 121°C for 15 minutes. Cool to 50°C, mix well and dispense into plates. For blood agar, cool to 50°C and enrich with 10% v/v sterile defibrinated blood. If desired, 20 units Penicillin and 40 µg Streptomycin per ml of medium may be added to make the medium selective for fungi. When the medium is solidified, invert the plates to avoid excess moisture. The prepared medium should be stored at 8-15°C.

Formula in g/L

infusion powder	12.500
BHI powder	5.000
Proteose peptone	10.000
Dextrose (Glucose)	2.000
Sodium chloride	5.000
Disodium phosphate	2.500
Agar	15.000

Final pH 7.4 \pm 0.2 at 25°C

Deterioration

The color of *BioScien* BHI Agar medium is cream to yellow homogeneous free flowing powder, dehydrated medium is clear light amber coloured to slightly opalescent gel. After addition of 5% v/v sterile defibrinated blood it turn to cherry red coloured, opaque gel forms in Petri plates. If there are any physical changes for powder or signs of deterioration (shrinking, cracking, or discoloration), and contaminations for hydrated media, discard the medium.

SPECIMEN COLLECTION AND PRESERVATION (4,5)

Clinical samples and blood(sputum, ETT, throat swab, conjunctival swab, ear swab, CSF, wound and pus swab, urine, urine catheter, and any effusion fluid), appropriate techniques for handling specimens as per established guidelines should be followed. After use, contaminated materials must be sterilized by autoclaving before discarding.

Note: for patient preparation follow Medical laboratory instruction

EQUIPMENT REQUIRED NOT PROVIDED

- · Sterile cups
- · Sterile petri-dishes
- Incubator
- Autoclave

CHARACTERISTICS OF THE COLONIES (1)

After the incubation period examine plates for organisms of interest. When examining primary plates a hand lens or stereoscopic microscope should be available for examining very small colonies. The different types of colonial morphology appearing on the agar plate should be noted as well as the number of each morphotype present. Hemolysis is a useful differential characteristic that is best viewed when a bright light is transmitted from behind the plate. Additional results such as pigment production and odor should also be recorded. Additional tests should be performed on isolated colonies from pure culture in order to complete identification.

- Certain pathogenic fungi may be inhibited on selective formulations of BHI therefore inoculation onto a non-selective medium is prescribed.
- Since nutritional requirements of organisms vary, some strains may be encountered that fail to grow or grow poorly on this medium

QUALITY CONTROL

To ensure adequate quality control, it is recommended that positive and negative control included in each run. If control values are found outside the defined range, check the system performance. If control still out of range please contact **BioScien** technical support.

BHI Agar	Expected results	
Streptococcus pneumoniae ATCC 6305	Good growth	
Pseudomonas aeruginosa ATCC 27853	Growth	
BHIA with Sheep Blood	Expected results	
Streptococcus pyogenes ATCC 19615	Growth, β-hemolysis	
Streptococcus pneumoniae ATCC 6305	Growth, α-hemolysis	
Staphylococcus aureus ATCC 25923	Growth	
Escherichia coli ATCC 25922	Growth	
Aspergillus niger ATCC 9642	Growth	
Candida albicans ATCC 10231	Growth	
BHIA w SB, Chloramphenicol & Cycloheximide	Expected results	
Aspergillus niger ATCC 9642	Growth	
Escherichia coli ATCC 25922	Inhibition	
Negative controls	Expected results	
Uninoculated medium	No change	

PERFORMANCE CHARACTERISTICS (4)

Performance of the medium from type cultures after incubation at a temperature of $35 \pm 2^{\circ}$ C, under 5 - 10% CO2, and observed after 24 - 72 hours. (It is recommended to grow Aspergillus brasiliensis and Saccharomyces cerevisiae aerobically at $30 \pm 2^{\circ}$ C).

Test Organisms	Growth without blood	Growth with 5% sheep blood	
Aspergillus brasiliensis ATCC 16404	Good	Good	
Neisseria meningitidis ATCC 13090	Moderate	Good	
Saccharomyces cerevisiae ATCC 9763	Good	Good	
Staphylococcus pneumoniae ATCC 6303	Good	Good	
Streptococcus pyogenes ATCC 19615	Moderate	Good	

REFERENCES

- Brain Heart Infusion Broth (Powder), US Biological. Archived from the original on 29 March 2014.
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- Isenberg, H.D. Clinical Microbiology Procedures Handb0ook. 2nd Edition. 8.
- 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.

	SYMBOLS IN PRODUCT LABELLING					
IVD	For in-vitro diagnostic use	\sum	Number of <n> test in the pack</n>			
LOT	Batch Code/Lot number	\triangle	Caution			
REF	Catalogue Number		Do not use if package is damaged			
1	Temperature Limitation	\mathbf{i}	Consult Instruction for use			
\square	Expiration Date					
~	Manufactured by					



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