

TECHNICAL DATA SHEET

MUELLER-HINTON BROTH

CULTURE OF MICROORGANISMS

1 INTENDED USE

Mueller Hinton Broth is a non-selective medium for the culture of a large number of microorganisms of varied origins, as well as for determining minimal inhibitory concentrations by the dilution method.

2 HISTORY

In work concerning the development of a medium capable of resisting autoclaving, Mueller and Hinton selected the complex medium of Gordon and Hine in an attempt to determine the essential components. The authors found that starch could replace pea extract in terms of nutritive value and also as a protective agent acting against toxic substances present in the medium. They subsequently found that pancreatic digest of meat could be replaced by acid hydrolysate of casein, thereby favoring the growth of gonococci and meningococci.

3 PRINCIPLES

The choice of ingredients is determined in order to obtain a very small quantity of thymine and thymidine, substances known to inhibit antibacterial activity of trimethoprim, a very small quantity of para-aminobenzoic acid (PABA) and its structural analogues, which antagonize the activity of sulfonamides.

Beef infusion and acid hydrolysate of casein are sources of amino acids and other nitrogenous substances favoring the growth of microorganisms.

Starch acts as a detoxifying agent.

4 TYPICAL COMPOSITION

The composition can be adjusted in order to obtain optimal performance.

For 1 liter of media :

Acid hydrolysate of Casein.....	17,5 g
Beef infusion.....	2,0 g
Soluble starch.....	1,5 g

pH of the ready-to-use media at 25 °C : 7,4 ± 0,2.

5 PREPARATION

- Dissolve 21,0 g of dehydrated media (BK108) in 1 liter of distilled or demineralized water.
- Slowly stir until complete dissolution.
- Dispense into tubes or vials.
- Sterilize in an autoclave at 121 °C for 15 minutes.
- Cool to room temperature.

✓ **Reconstitution :**
21,0 g/L

✓ **Sterilization :**
15 min a 121 °C

6 INSTRUCTIONS FOR USE

Cultures in tubes :

- Inoculate the medium with purified cultures or with other inocula containing mixed microflora.
- Incubate at the optimal temperature required in aerobic conditions or else in an anaerobiosis jar, depending on the bacteria to be cultured.

Determination of minimal inhibitory concentrations :

- Add serial dilutions of the stock solution of antibiotic to test to tubes containing the medium.
- Inoculate with a defined quantity of the culture of the microorganism to test.
- Incubate for 24 hours at 37°C.

7 RESULTS

Growth is determined by turbidity resulting from microbial division. The last tube with no microbial development contains antibiotic at a concentration which is the minimal inhibitory concentration. Multiplying this concentration by the corresponding dilution factor furnishes the concentration of active antibiotic.

8 QUALITY CONTROL

Dehydrated media : light beige powder, free-flowing and homogeneous.

Prepared media : amber solution, limpid.

Typical culture response after 24 hours of incubation at 37 °C, inoculum $\leq 10^2$ microorganisms :

Microorganisms		Growth
<i>Staphylococcus aureus</i>	WDCM 00034	Positive
<i>Streptococcus pyogenes</i>	ATCC® 19615	Positive

9 STORAGE / SHELF LIFE

Dehydrated media : 2-30 °C.

The expiration date is indicated on the label.

Prepared media in tubes or vials (*) : 180 days at 2-25 °C.

(*) Benchmark value determined under standard preparation conditions, following manufacturer's instructions.

10 PACKAGING

Dehydrated media :

500 g bottle BK108HA

11 BIBLIOGRAPHY

Mueller, J.H., and Hinton, J. 1941. Proc. Soc. Exp. Biol. Med., **48**: 330-333.

National Committee for Clinical Laboratory Standards. 1985. Approved standard: M7 A. Method for dilution antimicrobial susceptibility tests for bacteria that grow aerobically, NCCLS, Villanova, Pa.

Note for Guidance on minimising the risk of transmitting animal spongiform encephalopathy agents via human and veterinary medicinal products (EMA/410/01 Rev. 2 – October 2003) adopted by the Committee for Proprietary Medicinal Products (CPMP) and by the Committee for Veterinary Medicinal Products (CVMP) ; Official Journal of the European Union, 2004/C 24/03.

12 ADDITIONAL INFORMATION

The information provided on the labels take precedence over the formulations or instructions described in this document and are susceptible to modification at any time, without warning.

Document code : MUELLER HINTON BROTH_ENv8.
Creation date : 01-2003
Updated : 05-2016
Origin of revision : General update.