



## Mueller Hinton Broth

M391

Mueller Hinton Broth is recommended to determine the susceptibility of bacteria to sulphonamides by the tube dilution method. Also used for primary isolation of gonococci and meningococci.

### Composition\*\*

Ingredients	Gms / Litre
Beef, infusion from	300.000
Casein acid hydrolysate	17.500
Starch	1.500
Final pH ( at 25°C)	7.3±0.1

\*\*Formula adjusted, standardized to suit performance parameters

### Directions

Suspend 21 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Mix well and dispense into tubes as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Note: It is suggested to boil the medium before autoclaving to avoid settling of starch at the bottom.

### Principle And Interpretation

The Mueller Hinton formulation was originally developed as a simple, transparent agar medium for the cultivation of pathogenic *Neisseria* species (2). Other media were subsequently developed that replaced the use of Mueller Hinton Agar for the cultivation of pathogenic *Neisseria* species, but it became widely used in the determination of sulfonamide resistance of gonococci and other organisms. Mueller Hinton Broth is recommended for dilution antimicrobial susceptibility testing of all species of most commonly encountered aerobic and facultatively anaerobic bacteria (3, 1).

Beef infusion and casein acid hydrolysate provide nitrogenous compounds, carbon, sulphur and other essential nutrients. Starch acts as a protective colloid against toxic substances present in the medium. Starch hydrolysis yields dextrose, which serves as a source of energy. These ingredients are selected for low thymine and thymidine content as determined by MIC values for *Enterococcus faecalis* with sulfamethoxazoletrimethoprim (SXT). Calcium and magnesium ion concentrations are adjusted to provide the amounts recommended by CLSI to give the correct MIC values with aminoglycosides and *Pseudomonas aeruginosa* (3).

### Quality Control

#### Appearance

Cream to yellow homogeneous free flowing powder

#### Colour and Clarity of prepared medium

Light amber coloured clear solution in tubes

#### Reaction

Reaction of 2.1% w/v aqueous solution at 25°C. pH : 7.3±0.1

#### pH

7.20-7.40

#### Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

#### Cultural Response

Organism	Inoculum (CFU)	Growth
<b>Cultural Response</b> <i>Escherichia coli</i> ATCC 25922	50-100	good-luxuriant
<i>Haemophilus influenzae</i> ATCC 49247	50-100	good-luxuriant (in Mueller Hinton)

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<i>Neisseria gonorrhoeae</i> ATCC 49226	50-100	Chocolate Broth) good-luxuriant
<i>Pseudomonas aeruginosa</i> ATCC 27853	50-100	good-luxuriant
<i>Staphylococcus aureus</i> ATCC 25923	50-100	good-luxuriant
<i>Enterococcus faecalis</i> ATCC 19433	50-100	good-luxuriant
<i>Streptococcus pneumoniae</i> ATCC 6305	50-100	good-luxuriant (in Mueller Hinton Blood Broth)

## Storage and Shelf Life

Store below 30°C in tightly closed container and use freshly prepared medium. Use before expiry date on the label.

## Reference

1. Murray P. R., Baron J. H., Pfaller M. A., Tenover J. C. and Tenover F. C., (Ed.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C. , ,
2. Mueller J. H. and Hinton J., 1941, Proc. Soc. Exp. Biol. Med., 48:330.
3. National Committee for Clinical Laboratory Standards, 2000, Approved Standard: M7-A5. Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria that grow aerobically, 5th Ed., NCCLS, Wayne, Pa.

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