

Mueller Hinton Agar No. 2

Intended Use

Mueller Hinton Agar No. 2 is recommended for testing susceptibility of common and rapidly growing bacteria using antimicrobial discs by Kirby-Bauer method. Manufactured to contain low levels of thymine, thymidine, calcium and magnesium.

Summary

The Mueller Hinton formulation was originally developed as a simple, transparent agar medium for the cultivation of pathogenic *Neisseria* species. However, these organisms are now commonly isolated on selective media.

Mueller Hinton Agar is now used as a test medium for antimicrobial susceptibility testing. Mueller Hinton Agar is recommended for the diffusion of antimicrobial agents impregnated on paper disc through an agar gel as described in NCCLS (National Committee for Clinical Laboratory Standards), now CLSI (Clinical and Laboratory Standards Institute) approved standard. Mueller Hinton Agar No. 2 is used in susceptibility testing of rapidly growing aerobic and facultatively anaerobic bacteria from clinical specimens.

Principle

Thymine and thymidine inhibit sulfonamide and trimethoprim activity and calcium and magnesium interferes with the activity of aminoglycoside antibiotics. To overcome this problems Mueller Hinton Agar No. 2 is manufactured to contain low levels of thymine, thymidine and controlled levels of calcium and magnesium. MX Nutrients 1 and Casein acid hydrolysate provide nitrogenous compounds, vitamins, carbon, sulphur, amino acids and other essential nutrients in Mueller Hinton media. Starch act as a "protective colloid" against toxic substances present in the medium.

Formula*

Ingredients	g/L
MX Nutrients 1#	2.0
Casein Acid Hydrolysate	17.5
Starch, Soluble	1.5
Agar	17.0
Final pH (at 25°C)	7.3 ± 0.1

*Adjusted to suit performance parameters.

#Equivalent to intended performance of Beef heart infusion

Storage and Stability

Store dehydrated medium below 30°C in tightly closed container and the prepared medium at 2°C-8°C. Avoid freezing and overheating. Use before expiry date on the label. Once opened keep powdered medium closed to avoid hydration.

Type of Specimen

Clinical samples - Pure Cultures Isolated from Urine, Stool, Blood etc.

Specimen Collection and Handling

Ensure that all samples are properly labelled. Follow appropriate techniques for handling samples as per established guidelines. Some samples may require special handling, such as immediate refrigeration or protection from light, follow the standard procedure. The samples must be stored and tested within the permissible time duration. After use, contaminated materials must be sterilized by autoclaving before discarding.

Directions

1. Suspend 38.00 g of the powder in 1000 mL purified / distilled water.
2. Heat to boiling to dissolve the powder completely.
3. Sterilize by autoclaving at 121°C (15 psi) for 15 minutes as per validated cycle.
4. Cool to 45°C-50°C.
5. Mix well and pour into sterile petridishes or dispense as desired.

Quality Control

Dehydrated Appearance: Cream to yellow coloured, homogeneous, free flowing powder.

Prepared Appearance: Light yellow to amber coloured, clear to very slightly opalescent gel forms in petridishes.

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

Organism (ATCC)	Growth
<i>Escherichia coli</i> (25922)	Good
<i>Haemophilus influenzae</i> (49247)	Good (on Mueller Hinton chocolate agar)
<i>Neisseria gonorrhoeae</i> (49226)	Good
<i>Pseudomonas aeruginosa</i> Strain Boson 41501 (27853)	Good
<i>Staphylococcus aureus</i> subsp. <i>aureus</i> (25923)	Good
<i>Enterococcus faecalis</i> (29212)	Good
<i>Streptococcus pneumoniae</i> (6305)	Good (on Mueller Hinton blood agar)

Performance and Evaluation

Performance of the product is dependent on following parameters as per product label claim:

1. Directions
2. Storage
3. Expiry

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques.

Warranty

This product is designed to perform as described on the label and package insert. The manufacturer disclaims any implied warranty of use and sale for any other purpose.

Reference

1. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Yolken R. H., (Ed.), 2007, Manual of Clinical Microbiology, 9th 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.
4. NCCLS Approved Standard: ASM-2, 1979, Performance Standards for Antimicrobial disc Susceptibility Tests, 2nd Ed., National Committee for Clin. Lab. Standards.
5. Bauer A. W., Kirby W. M., Sherris J. L. and Turck M., 1966, Am. J. Clin. Pathol., 45:493.
6. Present Status and Future Work, WHO Sponsored collaborative study, Chicago, Oct. 1967.
7. Ericsson H. M. and Sherris J. L., 1971, Acta Pathol. Microbiol., Scand. Sect B Suppl., 217:1.
8. National Committee for Clinical Laboratory Standards, 1986, Proposed Standards, M6-P, NCCLS, Villanova, Pa. Koch A. E. and Burchall J. J., 1971, Appl. Microbiol., 22: 812.
9. Ferone R. Bushby R. M., Burchall J. J., Moore W. D., Smith D., 1975, Antimicrob. Agents chemotherap., 7 : 91.
10. Pollock H. M., Minshew B. H., Kenney M. A., Schoenknecht F. D., 1978, Antimicrob. Agents Chemotherap.; 14:360.
11. DAmato R. F., and Thornsberry C., 1979, Curr. Microbiol., 2 : 135.
12. Data on file: Microxpress®, A Division of Tulip Diagnostics (P) Ltd.

Product Presentation:

Cat No.	Product description	Pack Size
201130660500	Dehydrated Culture Media	500 g
205130980100	Ready Prepared Plate (90 mm)	100 Plates

 Temperature Limit	 Manufacturer	 LOT	Batch Code	 Date of Manufacture	 This way up	 RO Received on
REF Catalogue Number	 Consult Instructions for use	 Use-by Date	 HC	Hygroscopic keep container tightly closed	OO Opened on	

Revision: 0725/VER-03

Disclaimer

Information provided is based on our inhouse technical data on file, it is recommended that user should validate at his end for suitable use of the product.