Biomed Global

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Mueller Hinton II with 5% Sheep Blood Agar| Ready-to-use Media

a product by Biomed MDX

IVD

Intended Use:

A medium for drug susceptibility testing of fastidious bacteria as standardized by the Clinical and Laboratory Standards Institute (CLSI).

Principle of the Procedure:

Mueller-Hinton II agar with 5% sheep blood is a nutrient-rich medium specifically formulated for antimicrobial susceptibility testing of fastidious bacteria, for example streptococci. The Mueller-Hinton II base ensures standardized and reproducible antibiotic diffusion, while the added sheep blood provides essential growth factors for these more demanding organisms. This combination allows for accurate determination of antibiotic effectiveness by observing zones of inhibition around antibiotic-impregnated discs.

Product Summary:

Mueller Hinton II Agar with 5% Sheep Blood is primarily used for antimicrobial susceptibility testing, particularly with disc diffusion or gradient strip methods. While suitable for rapidly growing aerobes, it may require supplementation with 5% sheep blood for more fastidious bacteria.

Formulation* (PER LITER):

Beef Extract	**2.0g**
Acid Hydrolysate of Casein	**17.5g**
Starch	**1.5g**
Agar	**17.0g**
Sheep Blood	**50ml**

pH 7.3 +/- 0.1

*Adjust and/or supplemental as required to meet performance criteria

Procedure

Materials Provided

90mm Mueller Hinton II Agar with 5% Sheep Blood Agar.

Materials Required But Not Provided

Ancillary culture media, reagents, and laboratory equipment as required.

Test Procedure

1. For routine susceptibility tests, the inoculum may be prepared by making a direct saline or broth suspension of colonies selected from an 18- to 24-h nonselective nutrient agar plate, e.g., blood agar.
2. Pick three to five colonies from fresh nonselective agar plate and suspend in the broth. (Colonies should not be more than 24 hrs).
3. Adjust the suspension to the turbidity of the 0.5 McFarland standard.
4. Within 15 minutes, dip the sterile swab into the suspension.
5. Inoculate the surface of the plate uniformly on the plate to achieve uniform growth.
6. Allow inoculum to be absorbed.
7. Place the appropriate discs onto the respective cultures. Positioned the discs in the centers.
8. Incubate plates aerobically at 35 ± 2°C within 18 to 24 hours.
9. Observe the plates by measuring the complete zones of inhibition diameters by using a caliper.
10. Zone sizes should fall within the ranges of acceptable zone diameter quality control limits specified by the Clinical and Laboratory Standards Institute (CLSI).
11. These limits are published in CLSI M100™ Performance Standards for Antimicrobial Susceptibility Testing.

Results

The diameters of the zones of inhibition surrounding the antibiotic discs should be measured and compared to the reference values provided

in the Clinical and Laboratory Standards Institute (CLSI) document M100™. The susceptibility of specific organisms can then be categorized as resistant, intermediate, or susceptible.

Quality Control

Inoculate representative samples with the following strains. Incubate the inoculated plates at 35 ± 2°C for 18 to 24 hrs. to allow colonies to develop on the medium.

Strains	ATCC®
Streptococcus pneumoniae	49619
Uninoculated plate	-

Zone sizes and MIC ranges should fall within the acceptable ranges specified by the ISO/TS 16782 Technical Specification and/or CLSI document M100™. These ranges are periodically updated, so refer to the latest CLSI guidelines for up-to-date recommendations.

Transportation:

Temperature fluctuations may occur during transportation. However, these fluctuations do not affect the performance, quality, or safety of the media.

Storage and Shelf Life:

Upon receipt, store plates at 2 to 8°C, in their original sleeve wrapping until just before use. Avoid freezing and overheating.

The plates may be inoculated up to the expiration date (see package label) and incubated for the recommended incubation times.

Warning and Precautions:

For in vitro diagnostic use. For Professional Use Only. Do Not. Reuse.

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

Limitations of the Procedure

This medium is for laboratory use only and is not intended for the diagnosis of disease or other conditions. Identifications are presumptive and colonies should be identified using appropriate methods

Reference

1. International Organization for Standardization. 2016. Clinical laboratory testing — Criteria for acceptable lots of dehydrated Mueller-Hinton agar and broth for antimicrobial susceptibility testing (ISO/TS 16782:2016).
2. Clinical and Laboratory Standards Institute (CLSI). 2024. Performance Standards for Antimicrobial Susceptibility Testing, 34th ed. CLSI document M100. Wayne, PA: Clinical and Laboratory Standards Institute.
3. Zimbro, M. J., Power, D. A., Miller, S. M., Wilson, G. E., & Johnson, J. A. (Eds.). (2009). Difco™ and BBL™ manual: Manual of microbiological culture media (2nd ed.). Becton, Dickinson and Company.

Packaging Symbol

Symbol	Definition
REF	Catalogue number
IVD	In Vitro Diagnostic Medical Device
LOT	Batch code
	Date of manufacture
	Temperature limit
	Use-by date
	Keep away from sunlight
	Do not re-use
	Fragile, handle with care
	Consult instructions for use or consult electronic instructions for use
	Do not use if packaging damaged and consult instructions for use
	Manufacturer

Further Information:

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