Mueller Hinton Agar Plate w/ 5% Sheep Blood

MP1806

Intended Use:

Recommended for determination of susceptibility of *Streptococcus* species to antimicrobial agents.

Composition**

Ingredients	g / L
HM infusion from 300 g #	2.000
Acicase ##	17.500
Starch	1.500
Agar	17.000
Sheep Blood	50.000 ml

**Formula adjusted, standardized to suit performance parameters

Equivalent to Beef, infusion from

Equivalent to Casein acid hydrolysate

Directions

Either streak, inoculate or surface spread the test inoculum (50-100 CFU) aseptically on the plate.

Principle And Interpretation

The Mueller Hinton formulation was originally developed as a simple, transparent agar medium for the cultivation of pathogenic *Neisseria* species (1). Mueller Hinton Agar is now used as a test medium for antimicrobial susceptibility testing (2). Mueller Hinton Agar is recommended for the diffusion of antimicrobial agents impregnated on paper disc through an agar gel as described in CLSI and EUCAST approved Standard (3,4).

Kirby-Bauer et al recommended this medium for performing antibiotic susceptibility tests using a single disc of high concentration (5). WHO Committee on Standardization of Susceptibility Testing has accepted Mueller Hinton Agar for determining the susceptibility of microorganisms because of its reproducibility (6). The mediumplates supplemented with 5% Sheep blood is recommended for determination of susceptibility of *Streptococcus* species to antimicrobial agents.

HM infusion B from and Acicase 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 sulfamethoxazole trimethoprim (SXT)

The Kirby-Bauer procedure is based on agar diffusion of antimicrobial substances impregnated on paper discs. This method employs disc with a single concentration of antimicrobial agent and the zone diameters observed are correlated with minimum inhibitory concentration (MIC) values (7,8,9). A standardized suspension of the organism is swabbed over the entire surface of the medium. Paper discs impregnated with specific amounts of antimicrobial agents are then placed on the surface of the medium, incubated and zones of inhibition around each disc are measured. The susceptibility is determined by comparing with CLSI & EUCAST standards (2,4).

Type of specimen

Clinical samples - Isolated microorganisms from throat, wound sample,etc. According to CLSI & EUCAST, use only pure cultures. Clinical swab samples may be used directly, but should be confirmed with repeated test with pure cultures.

Specimen Collection and Handling

A standardized suspension of the organism is swabbed over the entire surface of the medium. Paper discs impregnated with specific amounts of antimicrobial agents are then placed on the surface of the medium, incubated and zones of inhibition around each disc are measured. The susceptibility is determined by comparing with CLSI standards (10). The various factors, which influence disc diffusion susceptibility tests, are agar depth, disc potency, inoculum concentration, pH of the medium and beta-lactamase production by test organisms (3,10). After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precaution

In Vitro diagnostic use only. For professional use only. Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling clinical specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. This medium is recommended for susceptibility testing of pure cultures only.
2. Inoculum density may affect the zone size. Heavy inoculum may result in smaller zones or too less inoculum may result in bigger zones.
3. As antimicrobial susceptibility is carried with antibiotic disc, proper storage of the disc is desired which may affect the potency of the disc.
4. Under certain circumstances, the in vitro results of antibiotic susceptibility may not show the same in vivo.
5. Each lot of the medium has been tested for the organisms specified on the COA.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Sterile Mueller Hinton Agar Plate w/ 5% Sheep Blood in 90 mm disposable plates with convex surface and absence of black particles/cracks/bubbles.

Colour

Red coloured medium

Quantity of medium

25 ml of medium in 90 mm disposable plate

pH

7.10- 7.50

Sterility Check

Passes release criteria

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 24-48 hours with added 5% w/v sterile sheep blood.

Organism	Growth	Recovery	Haemolysis	Clindamycin CD 2 mcg	Erythromycin E 15mcg	Vancomycin VA 30 mcg
Streptococcus pyogenes ATCC 19615	luxuriant	>=70%	beta			
Streptococcus pneumoniae ATCC 49619	luxuriant	>=70%	alpha	19 -25 mm	25 -30 mm	20 -28 mm

Storage and Shelf Life

On receipt store between 2-8°C. Use before expiry date on the label. Product performance is best if used within stated expiry period.

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 clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (12,13).

Reference

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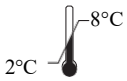
IVD ***In vitro* diagnostic medical device**



Storage temperature

EC REP CEpartner4U, Esdoornlaan 13,
3951DB Maarn, NL
www.cepartner4u.eu

CE **CE Marking**

Do not use if package is damaged

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