



Mueller Hinton Agar (i23118)

Mueller Hinton Agar is recommended for antimicrobial disc diffusion susceptibility testing of common, rapidly growing bacteria by the Bauer-Kirby method, as standardized by the CLSI.

Industry: Clinical

Principles & Uses

Mueller Hinton Agar, originally designed for cultivating pathogenic *Neisseria* species, has found its primary use in antimicrobial susceptibility testing. This transparent agar medium is valued for its batch consistency, low inhibitors for specific antibiotics, and support for the growth of many non-fastidious bacterial pathogens. Kirby-Bauer's influential work recommended it for antibiotic susceptibility testing, and it's been embraced by organizations like the WHO and CLSI for its reproducibility.

The culture medium's components, peptones, offer essential nutrients. Starch acts as a protective colloid, and starch hydrolysis yields dextrose for energy. The Kirby-Bauer procedure utilizes this medium in the agar diffusion method with paper discs impregnated with antimicrobial agents. Zone diameters around the discs correspond to minimum inhibitory concentration (MIC) values, determining susceptibility based on CLSI standards.

However, Mueller Hinton Agar isn't suitable for slowgrowing organisms, anaerobes, or capnophiles. Blood can be added to enhance growth, but this might affect susceptibility results for *enterococci* to aminoglycosides. In the broader context, antimicrobial susceptibility testing plays a critical role in guiding effective treatment for infectious diseases, aiding therapeutic decisions by assessing the sensitivity or resistance of pathogenic bacteria to antimicrobial compounds.

Composition (gr/L)

Acid Hydrolysate of Casein 17.5, Beef Extract 2, Starch 1.5, Agar 13. Final pH at 25°C 7.4 ± 0.2

Preparation from dehydrated Powder

Suspend 34 g of the powder in 1 Liter of purified water. Mix thoroughly. Autoclave at 121°C for 15 minutes. DO NOT OVERHEAT.

OPTIONAL: Cool medium to 45-50°C and aseptically add 5% sterile defibrinated sheep blood. Pour cooled Mueller Hinton agar into sterile Petri dishes on a level, horizontal surface to give a uniform depth of about 4 mm and cool to room temperature.

Quality Control

Dehydrated Appearance: Beige, free-flowing, homogeneous.

Prepared Appearance: Light to medium amber, slightly opalescent.

Reaction of 3.4% Solution at 25°C: pH 7.4 ± 0.2

Cultural Response

Using the organisms listed below, inoculate plates, add antibiotic disks and incubate as recommended by CLSI. Measure zone diameters and compare to the CLSI recommended zone ranges.

Test disks	E. Coli (25922)	S. aureus (25923)	P. aeruginosa (27853)
Ampicillin 10 μg	16-22	27-35	-
Tetracyclin 30 μg	18-25	19-28	-
Gentamicin 10 µg	19-26	19-27	16-21
Polymyxin B 300 IU	12-17	7-13	-

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Enterococcus faecalis (left). *Staphylococcus aureus* (right). The background has been darkened to enhance the visibility of bacterial growth.

Storage

Keep the container at 15-30 °C. Store prepared medium at 2-8 °C.