



Mueller Hinton Agar | AS-1299

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

Mueller Hinton Agar is a common culture medium that is frequently used for antimicrobial susceptibility testing (AST). It was created initially to cultivate pathogenic *Neisseria* species, but its versatility for a wide variety of non-fastidious bacteria makes it a good choice for industry standard for AST.

Peptones serve as the main source of nutrients, while starch provides a protective colloid. Its dependability in assessing bacterial sensitivity to antibiotics is attributed to its stable composition and minimal presence of compounds that inhibit antibiotic activity.

The standardized Kirby-Bauer disk diffusion method uses Mueller Hinton Agar and involves putting disks treated with antibiotics on the surface of the agar. The antibiotic's effectiveness against the test organism is correlated with the diameter of the ensuing inhibition zones.

Mueller Hinton Agar has limitations while being useful for a wide range of bacteria, such as its inability for growth of fastidious organisms and the possible conflict between some antibiotic activity with blood supplements.

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

Dissolve 34 g of the powder into 1 liter distilled water. Autoclave at 121 °C for 15 minutes. DO NOT OVERHEAT.

It is optional to add 5% sterile defibrinated sheep blood to the medium. for this purpose, the autoclaved medium must be cooled to 45 - 50 °C.

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

Microbial Test Results

Inoculate plates with organisms listed below. Put antibiotic disks on the surface of the agar and incubate as recommended by CLSI. Measure zone diameters and compare to the CLSI recommended zone ranges.



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

Storage

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