Technical Information

Mueller Hinton MiVeg Agar No. 2

Product Code :VM2084

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

Composition

Ingredients	Gms / Litre
MiVeg hydrolysate	17.500
MiVeg infusion	2.000
Starch, soluble	1.500
Agar	17.000
Final pH (at 25°C)	7.3±0.2

** Formula adjusted, standardized to suit performance parameters.

Principle & Interpretation

Mueller Hinton MiVeg Agar No. 2 is prepared by using Miveg hydrolysate and Miveg infusion in place of casein enzymic hydrolysate and Beef infusion, thereby making the medium BSE/TSE risk free. The goal of susceptibility test is to predict through an in vitro assessment the likelihood of successfully treating a patient's infection with a particular antimicrobial agent (1). This medium is the modification of Mueller Hinton Agar originally developed for the cultivation of pathogenic *Neisseria* species (2). Bauer, Kirby et al recommended Mueller Hinton Agar for performing antibiotic susceptibility tests using a high concentration single disc (5). It 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 (4).

This medium with addition of 5% sheep blood and Mueller Hinton Chocolate Agar have been recommended for antimicrobial susceptibility testing of *Streptococcus pneumoniae* and *Haemophilus influenzae* . Thymine and thymidine inhibit Sulfonamide and Trimethoprim (9,10) activity and calcium and magnesium (11,12) interferes with activity of aminoglycoside antibiotics. It is manufactured with low levels of thymine, thymidine and controlled levels of calcium and magnesium to overcome problems mentioned above.

This medium contains MiVeg Hydrolysate and MiVeg Infusion which supplies nitrogenous compounds, carbon, sulphur and other essential nutrients. Starch, soluble 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). Calcium and magnesium ion concentrations are adjusted to provide the amounts recommended by CLSI to give the correct MIC values with aminoglycosides and *Pseudomonas aeruginosa* (3).

The Bauer-Kirby procedure is based on the agar diffusion of antimicrobial substances impregnated on paper discs. This method employs discs with a single concentration of antimicrobial agent and zone diameters are correlated with minimal inhibitory concentration (MIC) values (2,3,7). A standardized suspension of the organism is inoculated by swabbing over the entire surface of the medium. Then paper discs impregnated with specified amounts of antimicrobial agents are placed on the surface of the medium. The plates incubated at 35-37°C for 24 hours and after incubation zones of inhibition around each disc are measured. The susceptibility is determined by comparing with NCCLS Standards (8). 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 (7).

Methodology

Suspend 38 grams of powder media in 1000 ml distilled water. Mix thoroughly. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Mix well and dispense as desired.

Quality Control

Physical Appearance

Cream to yellow homogeneous free flowing powder

Gelling

Firm, comparable with 1.7% agar gel.

Colour and Clarity of prepared medium

Light amber coloured clear to slightly opalescent gel forms in Petri Plates

Reaction

Reaction of 3.8 % w/v aqueous solution pH: 7.3 ±0.2 at 25°C

pH range

7.10-7.50

Cultural Response/Characteristics

Cultural characteristics observed after an incubation at 35-37°C for 24-48 hours

Organisms (ATCC)	Inoculum (CFU)	Growth	Recovery
Escherichia coli ATCC25922	50-100	good-luxuriant	>=70%
Haemophilus influenza ATCC 35056	50-100	good-luxuriant (chocolate Agar)	>=70%
Pseudomonas aeruginosa ATCC 27853	50-100	good-luxuriant	>=70%
Staphylococcus aureus ATCC 25923	50-100	good-luxuriant	>=70%
Streptococcus faecalis ATCC 29212	50-100	good-luxuriant	>=70%
Streptococcus pneumonia ATCC 6303	50-100	good-luxuriant (Blood Agar)	>=70%

Storage and Shelf Life

Dried Media: Store below 30°C in tightly closed container and use before expiry date as mentioned on the label.
Prepared Media: 2-8^0 in sealable plastic bags for 2-5 days.

Further Reading

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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.
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8. National Committee for Clinical Laboratory Standards, 1986, Proposed Standards, M6-P, NCCLS, Villanova, Pa.
09. Koch A. E. and Burchall J. J., 1971, Appl. Microbiol., 22: 812.
10. Ferone R. Bushby R. M., Burchall J. J., Moore W. D., Smith D., 1975, Antimicrob. Agents chemotherap., 7 : 91.
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12. DAmato R. F., and Thornsberry C., 1979, Curr. Microbiol., 2 : 135.

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