Technical Information

Hisitest Agar

Product Code: DM 1485A

Application: - Hisitest Agar is used for determination of antibiotic susceptibility of fastidious microorganisms.

Composition**

Ingredients	Gms / Litre
Casein enzymic hydrolysate	11.000
Peptone	3.000
Sodium chloride	3.000
Dextrose	2.000
Starch	1.000
Buffer salt	3.300
Nucleoside basis	0.020
Thiamine	0.00002
Agar	8.000
Final pH (at 25°C)	7.4±0.2

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

Hi-Sensitivity Test Agar is developed for antimicrobial susceptibility tests It gives a reproducible, semi-defined medium in which the mineral contents have been stabilized. The thymine and thymidine content is very low thus making it most suitable for testing antimicrobial activity of sulphonamides. It supports the growth of majority of micro-organisms without further supplementation. Casein enzymic hydrolysate, peptone, dextrose, vitamins supply nitrogen, carbon compoundsand other essential growth nutrients.

This medium has been designed to overcome the problems occurring in Mueller-Hinton media as (1 - 7)

1. Different M.I.C. values in the broth and agar versions of the medium.

2. Agar shows antagonistic effect towards tetracycline.

3. High levels of sulphonamide and trimethoprim antagonists.

4. Poor reproducibility with different manufacturers peptones.

5. Poor growth supporting ability for Streptococci and variable growth rates with Gram-positive organisms.

Some mutant strains which are totally dependent on thymine and thymidine for their growth, will not grow on Sensitivity Test Agar due to very low levels of these compounds in this medium as they are the naturally occurring antagonist of Trimethoprim. Care must be taken to recognize these strains (8, 9, 10). Some pathogenic organisms are nutritionally dependent due to intrinsic demands for special growth factors. Supplemental nutrients can be added to Sensitivity Test Agar to improve the growth of these organisms (1). The following nutrients can be used.

Nutrient	Micro-organism
1. Laked Blood (5% v/v)	Neisseria and Streptococcus
2. Fildes Peptic Digest of Blood (5% v/v)	Haemophilus
3. Menadione (0.5 mcg/ml) & Thiamine (2mcg/ml)	Dwarf colonies of S.aureus and coliform bacteria
4. Pyridoxine Hydrochloride (1mcg/ml)	Symbiotic Streptococci

Methodology

Suspend 31.32 grams of dehydrated powder media in 1000 ml distilled water. Mix thoroughly & heat to boil to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C. Shake well before pour into sterile Petri

Quality Control

Appearance
Light yellow coloured homogeneous free flowing powder
Gelling
Firm, comparable with 0.8% Agar gel.
Colour and Clarity
Light yellow coloured clear to slightly opalescent gel forms in Petri plates.
Reaction
Reaction 3.13% w/v aqueous solution at 25°C. pH : 7.4±0.2
pH Range
7.20-7.60
Cultural Response
DM 1485A: Cultural characteristics observed after an incubation at 35 - 37°C for 18 - 24 hours.

Organism	Inoculum (CFU)	Growth	Recovery
Bacillus subtilis ATCC 6633	50-100	good-luxuriant	>=70%
Bacteroides vulgatus ATCC 8482	50-100	good-luxuriant	>=70%
Enterococcus faecalis ATCC 29212	50-100	good-luxuriant	>=70%
Salmonella Typhimurium ATCC 14028	50-100	good-luxuriant	>=70%
Staphylococcus aureus ATCC 25923	50-100	good-luxuriant	>=70%
Streptococcus pyogenes ATCC 19615	50-100	good-luxuriant	>=70%

Storage and Shelf Life

Dried Media: Store below 8°C in tightly closed container and the prepared medium at 2- 8°C. Use before expiry date on the label.
Prepared Media: 2-8° in sealable plastic bags for 2-5 days.

Further Reading

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4. Duncan I.B.R., 1974, Antimicrobial agents & Chemotherapy, 5:9.
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8. Tanner E.I. and Bullin C.H., 1974, J. Clin. Path., 27:565.
9. Thomas M. and Bond L., 1973, Med. Lab. Technol., 30:277.
10. Barker J., Healing D. and Hutchinson J.G.P., 1972, J. Clin. Path., 25:1086.

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- User must ensure suitability of the product(s) in their application prior to use.
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