Technical Information

Antibiotic Assay Medium No.1 (Seed Agar)

Product Code : DM1003

Application: - Antibiotic Assay Medium No.1 (Seed Agar) is used in the microbiological assay of beta-lactam and other antibiotics.

Composition**

Ingredients	Gms / Litre
Peptic digest of animal tissue (Peptone)	6.000
Casein enzymic hydrolysate	4.000
Yeast extract	3.000
Beef extract	1.500
Dextrose	1.000
Agar	15.000
Final pH (at 25°C)	6.6±0.2

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

The potency of an antibiotic can be determined by chemical, physical and biological methods. An assay is performed to determine the ability of an antibiotic to kill or inhibit the growth of living microorganisms. Biological tests offer the most convenient means of performing an assay [(1)], since a reduction in the antimicrobial activity of a specific antibiotic reveals changes that is not usually displayed by chemical methods [(2)]. Antibacterial susceptibility testing may be performed by either dilution (turbidimetric) or diffusion methods. The choice of methodology is based on many factors, including ease of performance, flexibility and use of automated or semi-automated devices for both identification and susceptibility testing [(3)]. Grove and Randall have elucidated antibiotic assays and media in their comprehensive treatise on antibiotic assays [(4)]. Antibiotic Assay Medium No.1 is used in the microbiological assay of ß-lactam and other antibiotics. These media are prepared according to the specifications detailed in various pharmacopoeias [(2-6)] and by the FDA [(7)].

Freshly prepared plates should be used for antibiotic assays. Test organisms are inoculated in sterile seed agar cooled to 40-45^{o} C and spread evenly over the surface of solidified base agar. After incubation the concentration of the antibiotic being assayed is determined by measuring the zone of inhibition obtained, with that of reference standard antibiotic. All conditions in the microbiological assay must be carefully controlled. The use of standard culture media in the test is one of the important steps for good results.

Methodology

Suspend 30.5 grams of powder media in 1000 ml distilled water. Shake well & heat to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Advice : Recommended as a inoculum medium for Amikacin, Bacitracin, Cap reomycin, Cephalothin, Cephaperin, Chloramphenicol, Chlortetracycline, Cloxacillin Cycloserine, Colistimethate sodium, Colistin, Demeclocycline, Dihydrostreptomycin, Erythromycin, Framycetin, Gentamicin,Kanamycin, Kanamycin B, Kanamycin sulphate, Lymecycline, Methacycline, Nafcillin, Neomycin, Netilmicin, Novobiocin, Oxytetracycline, Paromomycin, Penicillin-G, Rifamycin sodium Rolitetracycline, Sisomycin Spiramycin, Streptomycin Tetracycline, Tobramycin, Troleandomycin, Tylosin.

Quality Control

Physical Appearance
Cream to yellow homogeneous free flowing powder

Gelling
Firm,comparable with 1.5% Agar gel

Colour and Clarity of prepared medium
Yellow coloured, clear to slightly opalescent gel forms in Petri plates.

Reaction
Reaction of 3.05% w/v aqueous solution at 25°C. pH : 6.6±0.2

pH range 6.40-6.80

Cultural Response/ characteristics
DM 1003: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Inoculum medium	Assay medium	Inoculum & Assay medium
Bacillus subtilis ATCC 6633 *Bordetella bronchiseptica ATCC 4617*	50-100	luxuriant	>=70%	Framycetin, Josamycin,Josamycin propionate, Kanamycin B,Spiramycin, Streptomycin,Vancomycin Colistimethate sodium,Colistin,Polymyxin B	Streptomycin , Vancomycin	
Escherichia coli ATCC *10536*	50-100	luxuriant	>=70%	Chloramphenicol		
Bacillus cereus var mycoides ATCC *11778*	50-100	luxuriant	>=70%	Oxytetracycline, Tetracycline		
Bacillus pumilis ATCC *14884*	50-100	luxuriant	>=70%	Chlortetracycline,Framycetin, Kanamycin sulphate		
Klebsiella pneumoniae ATCC *10031*	50-100	luxuriant	>=70%	Capreomycin,Dihydrostreptomycin, Neomycin,Streptomycin, Troleandomycin		
Micrococcus luteus ATCC *9341*	50-100	luxuriant	>=70%	Erythromycin, Erythromycin Rifamycin sodium		
Micrococcus luteus ATCC *10240*	50-100	luxuriant	>=70%			Bacitracin
Pseudomonas aeruginosa ATCC *25619*	50-100	luxuriant	>=70%	Carbenicillin		
Staphylococcus aureus ATCC *29737* Staphylococcus epidermidis ATCC 12228	50-100	luxuriant	>=70%	Amikacin,Cephothin, Cephapirin, Chlotetracycline, Cloxacillin, Cycloserine,Demeclocycline ,Doxycycline, Kanamycin, Methacycline, Nafcillin, Oxytetracycline,Penicillin G, Rolitetracycline, Tetracycline, Tobramycin, Tylosin Gentamicin, Neomycin,Netilmicin, Novobiocin,Sisomycin,Paromomycin	Cephalothin, Cephapirin, Nafcillin,Penicillin-G	Cloxacillin

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° in sealable plastic bags for 2-5 days.

Further Reading

1. Pelczar M. J. Jr., Reid R. D., Chan E. C. S., 1977, Microbiology, 4th Edi, Tata McGraw-Hill Publishing Company Ltd, New Delhi

2. The United States Pharmacopoeia, 2009, The United States Pharmacopoeial Convention,Rockville, MD.

3. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Yolken R. H., (Eds.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.

4. Grove and Randall, 1955, Assay Methods of Antibiotics Medical Encyclopedia, Inc.New York.

5. European Pharmacopoeia, 2009, European Department, for the Quality of Medicines

6. British Pharmacopoeia, 2009,The Stationery office British Pharmacopoeia

7. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs, FDA, CFR, 1983 Title 21, Part 436, Subpart D, Washington, D.C.: U.S. Government Printing Office, paragraphs 436, 100-436, 106, p. 242- 259 (April 1).

Disclaimer :

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