Antibiotic Assay Medium A (No. 1) (Seed Agar)

Intended Use
Antibiotic Assay Medium A (No. 1) (Seed Agar) is used for determining antibiotic potency by microbiological assay techniques as per USP/IP.

Summary
The potency or the activity of an antibiotic can be determined by chemical, physical and biological means. Biological tests offer the most convenient means of performing an assay, since a reduction in the antimicrobial activity of a specific antibiotic reveals changes not usually displayed by chemical methods. Antibacterial susceptibility testing may be performed by either dilution (turbidimetric) or diffusion methods. The choice of methodology is often based on many factors, including relative ease of performance, flexibility and use of automated or semi-automated devices for both identification and susceptibility testing. Grove and Randall have elucidated those antibiotic assays and media in their comprehensive treatise on antibiotic assays. 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 and by the FDA.
Freshly prepared plates should be used for antibiotic assays. Test organisms are inoculated in sterile seed agar cooled to 40°C-45°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.

Principle
Nutrients and growth factors are supplied by the ingredients like Peptone, Pancreatic Digest of Casein, yeast extract and cara beef extract. Dextrose is supplemented as a carbon and energy source.
Cylinder Plate Assay: This method is based on the diffusion of an antibiotic solution from a cylinder placed on the surface of an inoculated agar medium. The diameter of a zone of inhibition after incubation depends, in part, on the concentration or activity of the antibiotic. This method is used in the assay of commercial preparations of antibiotics, as well as in the quantitative determination of antibiotics in body fluids, animal feeds and other materials.
Turbidimetric Assay: The turbidimetric method is based on the inhibition of growth of a microbial culture in a fluid medium containing a uniform solution of an antibiotic. Turbidimetric determinations have the advantage of requiring a short incubation period, providing test results after 3 or 4 hours. However, the presence of solvents or other inhibitory materials may influence turbidimetric assays more markedly than cylinder plate assays. Use of this method is appropriate only when test samples are clear.

Formula*
Ingredients	**g/L**
Peptone	6.0
Pancreatic Digest of Casein	4.0
Yeast Extract	3.0
Cara Beef Extract#	1.5
Dextrose	1.0
Agar	15.0
Final pH (at 25°C)	6.6 ± 0.1

*Adjusted to suit performance parameters
Equivalent to Beef Extract

Storage and Stability
Store dehydrated medium below 30°C in tightly closed container and the prepared medium at 2°C-8°C. Avoid freezing and overheating. Use before expiry date on the label. Once opened keep powdered medium closed to avoid hydration.

Type of specimen
Pharmaceutical sample

Specimen Collection and Handling

Ensure that all samples are properly labelled.
Follow appropriate techniques for handling samples as per established guidelines.
Some samples may require special handling, such as immediate refrigeration or protection from light, follow the standard procedure The samples must be stored and tested within the permissible time duration.
After use, contaminated materials must be sterilized by autoclaving before discarding.

Directions

1. Suspend 30.5 g of the powder in 1000 mL purified / distilled water and mix thoroughly.
2. Boil with frequent agitation to dissolve the powder completely.
3. Sterilize by autoclaving at 121°C (15 psi) for 15 minutes as per validated cycle.
4. Cool to 45°C-50°C.
5. Pour into Sterile petridishes as desired.

Quality Control

Dehydrated Appearance: Light yellow coloured, homogeneous, free flowing powder.
Prepared Appearance: Yellow coloured, slightly opalescent gel forms in petridishes.
Growth Promotion Test: Growth promotion is carried out in accordance with the harmonized method of USP/EP/JP and growth is observed after an incubation at 18-24 hours at 30°C-35°C.
Growth Promoting Properties: The test results observed are within the specified temperature and shortest period of time specified in the test, inoculating ≤100 cfu of appropriate microorganism at 30°C-35°C for 18 hours.

Organism (ATCC)	Growth	Antibiotic Assayed by	
		a) Cylinder Plate Method	b) Turbidimetric Method
Bordetella bronchiseptica (4617)	Good	Colistimethate, Colistin, Polymyxin B	-
Escherichia coli (10536)	Good	-	Chloramphenicol
Klebsiella pneumoniae subsp. *pneumoniae* (10031)	Good	-	Capreomycin, Troleandomycin, Dihyrostreptomycin, Neomycin
Kocuria rhizophila Strain PCI 1001 (9341)	Good	Erythromycin	-
Micrococcus luteus (10240)	Good	Bacitracin	-
Pseudomonas aeruginosa (25619)	Good	Carbenicillin	-
Staphylococcus aureus (29737)	Good	Cloxacillin, Nafcillin, Penicillin G	Chlotetracycline, Tetracycline,Oxytetracycline

Performance and Evaluation

Performance of the product is dependent on following parameters as per product label claim:

1. Directions
2. Storage
3. Expiry

Precautions / Limitations

In vitro diagnostic use only. Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling clinical specimens. Freshly prepared plates must be used or it may result in erroneous results.

Warranty

This product is designed to perform as described on the label and package insert. The manufacturer disclaims any implied warranty of use and sale for any other purpose.

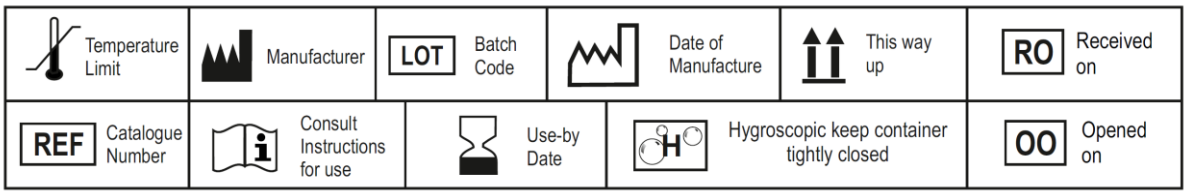
Reference

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 U. S. Pharmacopoeial Convention, Rockville, MD.

3. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H., Yolken R. H., (Eds.), 2003. Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.
4. Grove & 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 & Methods of Assay of Antibiotics & Antibiotic containing Drugs, FDA, CFR, 1983. Part 436(D), Washington, D.C.: U.S. Govt. Printing Office, paragraphs 436, 100-436, 106, p. 242- 259
8. Data on file: Microxpress®, A Division of Tulip Diagnostics (P) Ltd.

Product Presentation:

Cat No.	Product description	Pack Size
201010130100	Dehydrated Culture Media	100 g
201010130500	Dehydrated Culture Media	500 g
203010440012	Ready Prepared Slant	12 Slants



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Disclaimer

Information provided is based on our inhouse technical data on file, it is recommended that user should validate at his end for suitable use of the product.