

TM 1748 – ANTIBIOTIC ASSAY MEDIUM NO. 10 (as per USP)

INTENDED USE

For antibiotic assay of Carbenicillin, Colistimethate sodium and Polymyxin-B.

PRODUCT SUMMARY AND EXPLANATION

This medium is used as seed agar for assay of Polymyxin B, Colistimethate sodium, Colistin and Carbenicillin. The medium composition is in accordance to USP and CFR and numerically identical with the name assigned by Groove and Randall. Polymixins are reported to have slow diffusion in agar giving smaller zone of inhibition. Hence the reduced agar concentration (1.2%) in this medium improves the diffusion of polymixin in the medium.

Polysorbate 80 is reported to function synergistically with polymixins on spheroplasts of *Pseudomonas aeruginosa*. Polysorbate 80 enhances the penetration of Polymyxin to the cytoplasmic membrane and hence is an appropriate ingredient in the medium used for assay of Polymyxin. Freshly prepared plates should be used for antibiotic assays. Test organisms are inoculated in sterile seed agar pre-cooled to 40-45°C and spread evenly over the surface of solidified base agar. All conditions in the microbiological assay must be controlled carefully. The use of standard culture media in the test is one of the important steps for good results.

COMPOSITION

Ingredients	Gms / Ltr
Tryptone	17.000
Soya peptone	3.000
Dextrose	2.500
Sodium chloride	5.000
Dibasic potassium phosphate	2.500
Agar	12.000

PRINCIPLE

Combination of tryptone and soya peptone provide essential nutrients for the growth of test organisms. Dextrose provides the carbon source, enhances the growth of test organism. Phosphates in the medium enhances buffering action and sodium chloride maintains osmotic equilibrium.

INSTRUCTION FOR USE

- Dissolve 42 grams in 1000 ml purified/distilled water containing 10 ml of Polysorbate 80.
- Heat to boiling to dissolve the medium completely.
- Sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes. Cool to 45-50°C.
- Mix well and pour into sterile Petri plates.

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder	: Cream to yellow homogeneous free flowing powder.
Appearance of prepared medium	: Medium amber coloured clear to very slightly opalescent gel forms in Petri plates.
pH (at 25°C)	: 7.2±0.1

INTERPRETATION

Cultural characteristics observed after incubation.



Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Antibiotics assayed	Incubation Temperature	Incubation Period
<i>Bordetella bronchiseptica</i>	4617	50-100	Luxuriant	>=70%	Colistimethate sodium, Colistin, Polymyxin B	32-37°C	18-24 Hours
<i>Pseudomonas aeruginosa</i>	25619	50-100	Luxuriant	>=70%	Carbenicillin	32-37°C	18-24 Hours

PACKAGING:

In pack size of 500 gm bottles.

STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.










Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

REFERENCES

1. United States Pharmacopoeia / National Formulary 2011, US Pharmacopoeial Convention, Inc., Rockville, MD.
2. 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).
3. Grove and Randall, 1955, Assay Methods of Antibiotics Medical Encyclopaedia, Inc. New York.
4. Barry, 1991, Procedure and theoretical considerations for testing antimicrobial agents in agar media. Antibiotics in Laboratory medicine, New York pp 3
5. Brown & Winsley, 1968.. J Gen Microbiol. 1968 50(3) Supplix.

 GMP Good Manufacturing Practices Certified	 Best Before	 Quantity	 Catalogue Number	 Manufacturer
 Temperature Unit	 Lot / Batch Number	 Consults Instructions for Use	 QR Code	

NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.

***For Lab Use Only**
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