



Antibiotic Assay Medium No. 6

M223

Intended Use:

Recommended for induction of spore production in *Bacillus subtilis* strains used in antibiotic assay.

Composition**

Ingredients	Gms / Litre
Tryptone	17.000
Soya peptone	3.000
Sodium chloride	5.000
Dextrose (Glucose)	2.500
Dipotassium hydrogen phosphate	2.500
Manganese sulphate	0.030
Final pH (at 25°C)	7.0±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 30.03 grams in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Principle And Interpretation

Antibiotic Assay media are used in the performance of antibiotic assays. Grove and Randall have elucidated those antibiotic assays and media in their comprehensive treatise on antibiotic assays (1). Schmidt and Moyer have reported the use of antibiotic assay medium for the liquid formulation used in the performance of antibiotic assay (5). These media are prepared as per FDA (4).

Tryptone and Soya peptone provides the nutrients and growth factors. Dextrose provides as energy source. Dipotassium hydrogen phosphate provides the buffering system. Manganese sulphate helps in the early initiation of *Bacillus* species.

Type of specimen

Pharmaceutical preparations

Specimen Collection and Handling

For pharmaceutical samples follow appropriate techniques for handling specimens as per established guidelines (2,3). After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

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 specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may result in erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Light amber coloured clear solution may contain slight precipitate.

Reaction

Reaction of 3.0% w/v aqueous solution at 25°C. pH : 7.0±0.2

pH

6.80-7.20

Cultural Response

Cultural characteristics observed after an incubation at different temperatures for 6 days.

Organism	Inoculum (CFU)	Growth	Incubated at	Spores
<i>Bacillus cereus</i> ATCC 10876	50-100	luxuriant	30°C	positive
<i>Bacillus stearothermophilus</i> ATCC 7953	50-100	luxuriant	55°C	positive
<i>Bacillus subtilis</i> ATCC 6633 subsp. <i>spizizenii</i> (00003*)	50-100	luxuriant	35°C	positive
<i>Bacillus pumilus</i> ATCC 14884	50-100	luxuriant	35°C	positive

Key : (*)Corresponding WDCM numbers.

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (2,3).

Reference

1. Grove and Randall, 1955, Assay Methods of Antibiotics Medical Encyclopedia, Inc, New York.
2. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
3. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
4. 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 pg 242-259 (April 1).
5. Schmidt and Moyer, 1944; J. Bact, 47:199.

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Disclaimer :

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