



## Antibiotic Assay Medium E

MM006

Antibiotic Assay Medium E is used for microbiological assay of Framycetin and Kanamycin B using *Bacillus subtilis* in accordance with Indian Pharmacopoeia

### Composition\*\*

Ingredients	Gms / Litre
Peptone	6.000
Yeast extract	3.000
Beef extract	1.500
Agar	15.000
pH after sterilization	7.9±0.1

\*\*Formula adjusted, standardized to suit performance parameters

### Directions

Suspend 25.5 grams in 1000 ml purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

*Advice : Recommended for the Microbiological assay of Framycetin and Kanamycin B .*

### Principle And Interpretation

This medium is used in the assay of commercial preparations of antibiotics, as well as for antibiotics in body fluids, feeds etc. Medium composition is in accordance to the specifications detailed in the IP, FDA (1,2). Peptone, yeast and beef extract provide necessary growth nutrients for the test organisms like *Bacillus subtilis* . This medium provides solidified substratum for growth of organisms. The pH-7.9 maintained in this medium- provides optimum growth conditions for *Bacillus subtilis* (3). This medium is used to prepare the base as well as seed layer in the microbiological assay of antibiotics such as Framycetin and Kanamycin B. To perform the antibiotic assay the Base Agar should be prepared on the same day as the test. For the cylinder method, a base layer of 21 ml is required. Once the base medium has solidified, seed layer inoculated with the standardized test culture can be overlaid. Even distribution of the layer is important.

### Quality Control

#### Appearance

Cream to yellow coloured homogeneous free flowing powder

#### Gelling

Firm, comparable with 1.5% Agar gel

#### Colour and Clarity of prepared medium

Medium amber coloured slightly opalescent gel forms in Petri plates.

#### pH

7.80-8.00

#### Cultural Response

MM006: Cultural characteristics observed after an incubation at 32-35°C for 5 days.

Organism	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed
<i>Bacillus subtilis</i> ATCC 6633	50-100	good-luxuriant	>=70%	Framycetin, Kanamycin B.

### Storage and Shelf Life

Store below 30°C in tightly closed container and use freshly prepared medium . Use before expiry date on the label.

## Reference

1. Indian Pharmacopeia 2010, Ministry of Health and Family welfare, Government of India, New Delhi.
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. Stearn and Stearn, J Bacteriol. 1933. 26(1): 37-55.

Revision : 2 / 2015

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