



## Antibiotic Assay Medium B

MM005

Antibiotic Medium B is used for microbiological assay of antibiotics 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	6.55±0.05

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

### Directions

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

### Principle And Interpretation

This medium is commonly used as base agar for microbiological agar diffusion assays for wide variety of antibiotics. These agar diffusion assays can be performed by cylinders, punched-hole or paper disc tests. This medium is prepared according to the specifications detailed in the IP and CFR(1,2).

Peptone, yeast extract and beef extract nitrogenous, vitamins and mineral requirement for the growth of test organisms for the test organisms. This medium provides solidified substratum for growth of organisms. This medium is widely used to prepare the base layer in the microbiological assay of antibiotics such as bacitracin, novobiocin and penicillin.

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 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

Amber coloured slightly opalescent gel forms in Petri plates.

#### pH

6.50-6.60

#### Cultural Response

MM005: Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed
<i>Bacillus subtilis</i> ATCC 6633	50-100	luxuriant	≥70%	Spiramycin

### 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 Pharmacopoeia 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).

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