



Antibiotic Assay Medium No.35

MU798

Antibiotic Assay Medium No.35 is used for the microbiological assay of Bleomycin using *Mycobacterium smegmatis* as a test organism in accordance to United States Pharmacopoeia.

Composition**

Ingredients	Gms / Litre
Peptone	10.000
Beef extract	10.000
Sodium chloride	3.000
Agar	17.000
Final pH (at 25°C)	7.0±0.1

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 40 grams in 1000 ml R-water/ purified/distilled water containing 10 gms glycerol. 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 Bleomycin .

Principle And Interpretation

This medium is formulated in accordance with USP and CFR (1,2). This medium is employed widely as base agar for agar diffusion assay of Bleomycin using *Mycobacterium smegmatis* .

The nutrients essential for growth of test organism is provided by peptone and beef extract in this medium. Agar provides excellent solid substratum for support and overlaying of seed agar, for the assay of Bleomycin. Addition of glycerol is important for slow and continuous provision of carbon to the test organism.

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 homogeneous free flowing powder

Gelling

Firm, comparable with 1.7% agar gel.

Colour and Clarity of prepared medium

Medium amber coloured clear to slightly opalescent gel forms in Petri plates.

Reaction

Reaction of 4.0% w/v aqueous solution containing 1% glycerol. pH : 7.0±0.1

pH

6.90-7.10

Cultural Response

MU798: Cultural characteristics observed after an incubation at 35-37 for 18-48 hours .

Organism	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed
<i>Mycobacterium smegmatis</i> ATCC 607	50-100	luxuriant	>=50%	Bleomycin

Storage and Shelf Life

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

Reference

1. United States Pharmacopoeia 2009, 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).

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