



## Antibiotic Assay Medium D

M556B

### Intended Use:

Recommended for the microbiological assay of Erythromycin using *Klebsiella pneumoniae* as a test organism in accordance with BP.

### Composition\*\*

Ingredients	Gms / Litre
HMH extract #	1.500
Yeast extract	1.500
Casitose ##	5.000
Glucose monohydrate	1.000
Sodium chloride	3.500
Dipotassium hydrogen phosphate	3.680
Potassium dihydrogen phosphate	1.320
Potassium nitrate	2.00
pH after sterilization	7.0±0.1

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

# Equivalent to Heart extract

## Equivalent to Peptone casein

### Directions

Suspend 19.4 grams (the equivalent weight of dehydrated medium) in 1000 ml R-water/ purified/distilled water. Heat with frequent agitation to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Adjust the pH of the medium, using freshly prepared buffer solution as recommended by the British pharmacopoeia for the antibiotic assayed.

### Principle And Interpretation

This medium is formulated in accordance to British Pharmacopoeia (1). This medium is widely used for turbidimetric assay of erythromycin esolate using *Klebsiella pneumoniae* as test organism. Turbidimetric methods for determining the potency of antibiotics are inherently more accurate and more precise than comparable agar diffusion procedures.

Combination of Casitose, HMH extract and yeast extract supply nutrients and essential mineral and growth factors for enhanced microbial growth. Potassium nitrate serves as inorganic source of nitrogen for the growth of test organism. Sodium chloride maintains the osmotic equilibrium while phosphates are incorporated in the medium to provide good buffering action. Glucose monohydrate serves as the carbon and energy source for faster growth.

Turbidimetric antibiotic assay is based on the change or inhibition of growth of a test microorganism in a liquid medium containing a uniform concentration of an antibiotic.

### Type of specimen

Pharmaceutical sample

### Specimen Collection and Handling:

For pharmaceutical sample 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 medium plates must be used or it may result in erroneous results.
2. Use of this method is appropriate only when test samples are clear.

## 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 coloured homogeneous free flowing powder

### Colour and Clarity of prepared medium

Light yellow coloured clear solution without any precipitate.

### Reaction

Reaction of 1.94% w/v aqueous solution. pH : 7.0±0.1

### pH

6.90-7.10

### Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Serial dilution with
<i>Klebsiella pneumoniae</i> ATCC 10031	50-100	luxuriant	Erythromycin estolate

## Storage and Shelf Life

The product should be stored in a tightly closed container and the prepared medium at room temperature before the expiry date on the label. Upon opening, products should be properly stored dry after tightly capping the bottle to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation in dry ventilated areas protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Product performance is best if used within the stated expiry period.

## References

1. British Pharmacopoeia, 2011, The Stationery Office, British Pharmacopoeia

### Disclaimer :

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