Antibiotic Assay Medium D

Antibiotic Assay Medium D is used for the microbiological assay of Erythromycin estolate using *Klebsiella pneumoniae*

**Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heart extract</td>
<td>1.500</td>
</tr>
<tr>
<td>Yeast extract</td>
<td>1.500</td>
</tr>
<tr>
<td>Peptone-Casein</td>
<td>5.000</td>
</tr>
<tr>
<td>Glucose monohydrate</td>
<td>1.000</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>3.500</td>
</tr>
<tr>
<td>Dipotassium hydrogen phosphate</td>
<td>3.680</td>
</tr>
<tr>
<td>Potassium dihydrogen phosphate</td>
<td>1.320</td>
</tr>
<tr>
<td>Potassium nitrate</td>
<td>2.000</td>
</tr>
</tbody>
</table>

Final pH (at 25°C) 7.0±0.2

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

**Directions**

Suspend 19.40 grams (the equivalent weight of dehydrated medium per litre) in 1000 ml purified/distilled water. Heat if necessary to dissolve the medium completely. Dispense and sterilize by autoclaving at 15lbs pressure (121°C) for 15 minutes.

Adjust the pH of the medium, using freshly prepared buffer solution as recommended by the European/British Pharmacopoeia for the antibiotic assayed.

**Principle And Interpretation**

Antibiotic Assay Medium D is used for the microbiological assay of Erythromycin estolate using *Klebsiella pneumoniae*. Grove and Randall have elucidated the antibiotic assays and media in their comprehensive treatise on antibiotic assays.

Turbidimetric methods for determining the potency of antibiotics are inherently more accurate and more precise than comparable agar diffusion procedures.

Combination of peptone, heart extract and yeast extract supplies nutrients and essential mineral and growth factors for enhanced microbial growth. Potassium nitrate serves as an 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. Use of this method is appropriate only when test samples are clear.

**Quality Control**

**Appearance**
Cream to yellow 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 at 25°C, pH: 7.0±0.2

**pH**
6.80-7.20

**Cultural Response**
M556: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Inoculum (CFU)</th>
<th>Growth</th>
<th>Serial dilution with</th>
</tr>
</thead>
</table>
**Klebsiella pneumoniae**
*ATCC 10031*

50-100 luxuriant Erythromycin stearate

**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**

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