Antibiotic Assay Medium No. 8

Antibiotic Assay Medium No. 8 is used for microbiological assay of Vancomycin in accordance with United States Pharmacopoeia.

**Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peptone</td>
<td>6.000</td>
</tr>
<tr>
<td>Yeast extract</td>
<td>3.000</td>
</tr>
<tr>
<td>Beef extract</td>
<td>1.500</td>
</tr>
<tr>
<td>Agar</td>
<td>15.000</td>
</tr>
<tr>
<td>pH after sterilization</td>
<td>5.9±0.1</td>
</tr>
</tbody>
</table>

**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 Vancomycin.*

**Principle And Interpretation**

The composition of this medium is in accordance to USP and CFR (1,2) and identical numerically with the name assigned by Grove and Randall (3).

Peptone, yeast and beef extract provide essential nutritional requirement for the test organisms. This medium provides solidified substratum for growth of organisms. This medium provides the optimal pH 5.9 for assay of tetracycline as these antibiotics are stable at slightly lower pH (4). This pH condition also supports the growth of test organisms. This medium is also used as base and seed agar medium for agar diffusion assay for mitomycin, mithramycin, plicamycin and Vancomycin (5).

To perform the antibiotic assay the Base Agar should be prepared on the same day as the test. The potency of an antibiotic can be demonstrated by its inhibitory effect on microorganisms under suitable conditions. 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**

Light amber coloured slightly opalescent gel forms in Petri plates.

**pH**

5.80-6.00

**Growth Promotion Test**

As per United States Pharmacopoeia

**Cultural Response**

MU041: Cultural characteristics observed after an incubation at 32 - 35°C for 18 24 hours.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Inoculum (CFU)</th>
<th>Growth</th>
<th>Recovery</th>
<th>Antibiotics assayed</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Bacillus subtilis ATCC 6633</em> 50-100</td>
<td>luxuriant</td>
<td>&gt;=70%</td>
<td></td>
<td>Vancomycin</td>
</tr>
</tbody>
</table>

Please refer disclaimer Overleaf.
**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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