Antibiotic Assay Medium C

Antibiotic Assay Medium C is used as the broth medium in turbidimetric assay of a wide variety of antibiotics in accordance with European 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>Beef extract</td>
<td>1.500</td>
</tr>
<tr>
<td>Yeast extract</td>
<td>3.000</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>3.500</td>
</tr>
<tr>
<td>Glucose monohydrate</td>
<td>1.000</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>pH after sterilization</td>
<td>*7.0±0.1</td>
</tr>
</tbody>
</table>

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

*While assaying Josamycin & Josamycin sulphate adjust the pH to 8.0 ±0.1

**Directions**

Suspend 19.9 grams of dehydrated media in 1000 ml R-water/ purified /distilled water. Heat if necessary 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 European pharmacopoeia for the antibiotic assayed.

Advice: Recommended for the microbiological assay of Colistimethate sodium, Dihydrostreptomycin sulphate, Erythromycin estolate, Framycetin sulphate, Gentamicin sulphate, Gramicidin, Kanamycin acid sulphate, Kanamycin monosulphate, Neomycin sulphate, Rifamycin sodium, Spiramycin, Streptomycin sulphate, Tylosin, Tylosin tartarate, Tyrothricin and Vancomycin hydrochloride.

**Principle And Interpretation**

This medium is used in turbidimetric assay of several antibiotics. The composition of the medium is in accordance to the specifications detailed in the European Pharmacopoeia (1). Turbidimetric methods for determining the potency of antibiotics are inherently more accurate and more precise than comparable agar diffusion procedures (2).

Peptone, beef extract and yeast extract provide essential nutrients and growth factors for enhanced microbial growth. 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 (3). 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

**pH**
6.90-7.10

**Cultural Response**
ME555: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.
Organism | Inoculum (CFU) | Growth | Serial dilution with
--- | --- | --- | ---
Escherichia coli ATCC 9637 | 50-100 | luxuriant | Colistimethate sodium
Escherichia coli ATCC 10536 | 50-100 | luxuriant | Rifamycin sodium
Enterococcus hirae ATCC 10541 | 50-100 | luxuriant | Gramicidin, Tyrothricin
Klebsiella pneumoniae ATCC 10031 | 50-100 | luxuriant | Streptomycin sulphate
Staphylococcus aureus ATCC 6538p | 50-100 | luxuriant | Framycetin sulphate, Genatamicin sulphate, Gramicidin, Kanamycin monosulphate, Kanamycin acid sulphate, Neomycin sulphate, Spiramycin; For Josamycin & Josamycin propionate- adjust the pH of the medium to 8.0 ± 0.1, For Vancomycin hydrochloride incubate at 35-37°C.
Staphylococcus aureus ATCC 9144 | 50-100 | luxuriant | Tylosin, Tylosin tartarate

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. European Pharmacopoeia, 2011, European Department, for the Quality of Medicines.

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