Acetobacter Broth (Mannitol)

Intended Use:
Recommended as a cultivation medium for mannitol positive Acetobacter species.

Composition

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peptone</td>
<td>3.000</td>
</tr>
<tr>
<td>Yeast extract</td>
<td>5.000</td>
</tr>
<tr>
<td>Mannitol</td>
<td>25.000</td>
</tr>
<tr>
<td>Final pH (at 25°C)</td>
<td>7.4±0.2</td>
</tr>
</tbody>
</table>

**Formula adjusted, standardized to suit performance parameters

Directions
Suspend 33.0 grams in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense in test tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Principle And Interpretation

Acetic acid bacteria are found in fruits with high carbohydrate concentration, which is selective for yeasts, that produce ethanol. This ethanol forms the substrate for acetic acid bacteria and may oxidize ethanol to acetic acid (7). Various synthetic and maintenance media for Acetobacter cultures have been cited (1). A typical maintenance medium is Acetobacter Broth (1).

Acetobacter Broth is formulated as per Manual of Microbiological Methods (5) and used for the maintenance of Acetobacter species utilizing mannitol (2).

Peptone, yeast extract in the medium provides nitrogen, vitamins and minerals necessary to support bacterial growth. Mannitol acts as energy source. Calcium carbonate acts as a buffer.

Type of specimen
Food samples - fruits

Specimen Collection and Handling
For food samples, follow appropriate techniques for sample collection and processing as per guidelines (6). 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. Further biochemical and serological tests must be carried out for complete identification.
2. Some organism may show poor growth due to nutritional variation.

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 homogeneous free flowing powder

Colour and Clarity of prepared medium
Yellow coloured, clear to slightly opalescent solution in tubes

Reaction

Please refer disclaimer Overleaf.
Reaction of 3.3% w/v aqueous solution at 25°C. pH : 7.4±0.2

pH
7.20-7.60

Cultural Response

<table>
<thead>
<tr>
<th>Organism</th>
<th>Inoculum (CFU)</th>
<th>Growth</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acetobacter hansenii ATCC</td>
<td>50-100</td>
<td>luxuriant</td>
</tr>
<tr>
<td>ATCC 35959</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Acetobacter pasteurianus</td>
<td>50-100</td>
<td>luxuriant</td>
</tr>
<tr>
<td>ATCC 6033</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (3,4).

Reference

1. Asai, 1968, Univ. of Tokyo Press, Tokyo, Japan and Univ. Park Press, Baltimore, MD.
2. Catalogue of Bacteria and Bacteriophages, 1992, 18th Ed., American Type Culture Collection, Rockville, MD.