Acetobacter Broth (Glucose)

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

Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yeast extract</td>
<td>10.000</td>
</tr>
<tr>
<td>Calcium carbonate</td>
<td>10.000</td>
</tr>
<tr>
<td>Dextrose (Glucose)</td>
<td>3.000</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 23.0 grams in 1000 ml purified/distilled water. Heat if necessary and dispense in test tubes, taking care to distribute calcium carbonate evenly. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Note: Due to presence of Calcium carbonate, the prepared medium forms opalescent solution with white precipitate.

Principle And Interpretation
Acetobacter species are aerobic, gram negative organisms. 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 glucose (2).

Yeast extract in the medium provides nitrogen, vitamins and minerals necessary to support bacterial growth. Glucose 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
Light amber coloured, clear to slightly opalescent solution with heavy white precipitate forms in tubes.

Reaction
Reaction of 2.3% w/v aqueous solution at 25°C. pH : 7.4±0.2

pH
7.20-7.60

Cultural Response
Cultural characteristics observed after an incubation at 35-37°C for 24-48 hours.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Inoculum (CFU)</th>
<th>Growth</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acetobacter aceti ATCC 15973</td>
<td>50-100</td>
<td>luxuriant</td>
</tr>
<tr>
<td>Acetobacter liquifaciens ATCC 14835</td>
<td>50-100</td>
<td>luxuriant</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.