Bile Salts Agar

Intended use
Recommended for isolation and identification of bile tolerant bacteria responsible for food poisoning. It is recommended by BIS committee under the specifications IS:5887(Part V) -1976.

Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peptone</td>
<td>10.000</td>
</tr>
<tr>
<td>HM extract #</td>
<td>5.000</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>5.000</td>
</tr>
<tr>
<td>Sodium taurocholate</td>
<td>5.000</td>
</tr>
<tr>
<td>Agar</td>
<td>15.000</td>
</tr>
<tr>
<td>Final pH ( at 25°C)</td>
<td>8.5±0.2</td>
</tr>
</tbody>
</table>

**Formula adjusted, standardized to suit performance parameters

# - Equivalent to Meat extract

Directions
Suspend 40 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. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

Principle And Interpretation
Bile Salt Agar is formulated as recommended by BIS (1) for isolation, identification and enumeration of *Vibrio cholerae*. *Vibrio* species, like many other gram-negative bacteria, grow in the presence of relatively high levels of bile salts (2). On Bile Salt Agar colonies of Vibrio's have a distinctive appearance which may be seen by growing a known strain of *Vibrio cholerae* and comparing it with *Escherichia coli* (1). Suspicious growths may be tested by slide agglutination using polyvalent cholera typing serum.

The medium contains peptone, HM extract which provides carbonaceous, nitrogenous compounds and other essential growth nutrients. Sodium chloride buffers the medium well. Sodium taurocholate inhibits most of the gram-negative organisms.

Type of specimen
Food samples

Specimen Collection and Handling:
For food samples, follow appropriate techniques for sample collection and processing as per guidelines (1,4)

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. Due to nutritional variations, some strains may show poor growth
2. Further biochemical and serological tests must be carried out for confirmation.

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
Light yellow coloured homogeneous free flowing powder
Gelling
Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium
Light yellow coloured clear to slightly opalescent gel forms in Petri plates.

Reaction
Reaction of 4.0% w/v aqueous solution at 25°C. pH : 8.5±0.2

pH
8.30-8.70

Cultural Response
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>Recovery</th>
</tr>
</thead>
<tbody>
<tr>
<td># Klebsiella aerogenes ATCC 13048 (00175*)</td>
<td>50-100</td>
<td>luxuriant</td>
<td>&gt;=50 %</td>
</tr>
<tr>
<td>Escherichia coli ATCC 25922 (00013*)</td>
<td>50-100</td>
<td>luxuriant</td>
<td>&gt;=50%</td>
</tr>
<tr>
<td>Salmonella Typhi ATCC 6539</td>
<td>50-100</td>
<td>luxuriant</td>
<td>&gt;=50%</td>
</tr>
<tr>
<td>Staphylococcus aureus subsp.aureus ATCC 25923 (00034*)</td>
<td>&gt;=10⁴</td>
<td>inhibited</td>
<td>0%</td>
</tr>
<tr>
<td>Vibrio cholerae ATCC 15748</td>
<td>50-100</td>
<td>luxuriant</td>
<td>&gt;=50%</td>
</tr>
</tbody>
</table>

Key : (#) Enterobacter aerogenes, (*) Corresponding WDCM numbers.

Storage and Shelf Life
Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°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 (2,3).

Reference