MacConkey Agar w/ 0.15% Bile salts, CV and NaCl

Intended Use:
Recommended for the selective isolation and differentiation of coliform organisms and other enteric pathogens from clinical and non-clinical samples.

Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gelatin peptone</td>
<td>17.000</td>
</tr>
<tr>
<td>Tryptone</td>
<td>1.500</td>
</tr>
<tr>
<td>Peptone</td>
<td>1.500</td>
</tr>
<tr>
<td>Lactose</td>
<td>10.000</td>
</tr>
<tr>
<td>Bile salts</td>
<td>1.500</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>5.000</td>
</tr>
<tr>
<td>Neutral red</td>
<td>0.030</td>
</tr>
<tr>
<td>Crystal violet</td>
<td>0.001</td>
</tr>
<tr>
<td>Agar</td>
<td>15.000</td>
</tr>
<tr>
<td>Final pH (at 25°C)</td>
<td>7.1±0.2</td>
</tr>
</tbody>
</table>

**Formula adjusted, standardized to suit performance parameters

Directions
Suspend 51.53 grams in 1000 ml purified/distilled water. Heat to boiling with gentle swirling to dissolve the agar completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Avoid overheating. Cool to 45 - 50°C and pour into sterile Petri plates. The surface of the medium should be dry when inoculated.

Principle And Interpretation
MacConkey agars are slightly selective and differential plating media mainly used for the detection and isolation of gram-negative organisms from clinical (10), dairy (14), food (5,11), water (1), pharmaceutical (3,12) and industrial sources (15). It is also recommended for the selection and recovery of the Enterobacteriaceae and related enteric gram-negative bacilli. USP recommends this medium for use in the performance of Microbial Limit Tests (12).

These agar media are selective since the concentration of bile salts, which inhibit gram-positive microorganisms, is low in comparison with other enteric plating media. The medium M081, which corresponds with, that recommended by APHA can be used for the direct plating of water samples for coliform bacilli, for the examination of food samples for food poisoning organisms (11) and for the isolation of Salmonella and Shigella species in cheese (14). Other than that this medium is also used for count of coli-aerogenes bacteria in cattle and sheep faeces (9), the count of coli-aerogenes and non-lactose fermenters in poultry carcasses (9), bacterial counts on irradiated canned minced chicken (13) and the recognition of coli-aerogenes bacteria during investigations on the genus Aeromonas (4).

MacConkey Agar is the earliest selective and differential medium for cultivation of enteric microorganisms from a variety of clinical specimens (7,8). The original medium contains protein, bile salts, sodium chloride and two dyes. The selective action of this medium is attributed to crystal violet and bile salts, which are inhibitory to most species of gram-positive bacteria. Gram-negative bacteria usually grow well on the medium and are differentiated by their ability to ferment lactose. Lactose-fermenting strains grow as red or pink colonies and may be surrounded by a zone of acid precipitated bile. The red colour is due to production of acid from lactose, absorption of neutral red and a subsequent colour change of the dye when the pH of medium falls below 6.8. Lactose non-fermenting strains, such as Shigella and Salmonella are colourless, transparent and typically do not alter appearance of the medium.

Peptone, Tryptone and gelatin peptone are sources of nitrogen, carbon, long chain amino acids and other nutrients. Lactose is a fermentable carbohydrate, Sodium chloride maintains the osmotic equilibrium. Bile salts and crystal violet are selective agents that inhibit growth of gram-positive organisms. Neutral red is the pH indicator dye.

Type of specimen
Clinical - faeces, urine and other pathological material, foodstuffs and dairy samples, water samples, pharmaceutical samples.
Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (6,10).
For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (5,11,14.).
For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards.(1)
For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per guidelines.(3,12) After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

In Vitro diagnostic use. 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 clinical specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Though the medium is recommended for selective isolation, further biochemical and serological testing must be carried out for further confirmation.
2. The surface of the medium should be dry when inoculated.

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 to pink homogeneous free flowing powder

Gelling
Firm comparable with 1.35% Agar gel.

Colour and Clarity of prepared medium
Red with purplish tinge coloured clear to slightly opalescent gel forms in Petri plates.

Reaction
Reaction of 4.95% w/v aqueous solution at 25°C. pH : 7.1±0.2

pH
6.90-7.30

Cultural Response
Cultural response was observed after an incubation at 30-35°C for 18-72 hours. Recovery rate is considered as 100% for bacteria growth on Soybean Casein Digest Agar.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Inoculum (CFU)</th>
<th>Growth</th>
<th>Recovery</th>
<th>Colour of colony</th>
</tr>
</thead>
<tbody>
<tr>
<td>Corynebacterium diptheriae type gravis</td>
<td>&gt;=10⁴</td>
<td>inhibited</td>
<td>0%</td>
<td></td>
</tr>
<tr>
<td>Shigella flexneri ATCC 12022 (00126*)</td>
<td>50 -100</td>
<td>fair to good</td>
<td>30 -40 %</td>
<td>colourless</td>
</tr>
<tr>
<td>Salmonella Paratyphi A ATCC 9150</td>
<td>50 -100</td>
<td>luxuriant</td>
<td>&gt;=50 %</td>
<td>colourless</td>
</tr>
<tr>
<td>Salmonella Abony NCTC 6017 (00029*)</td>
<td>50 -100</td>
<td>luxuriant</td>
<td>&gt;=50 %</td>
<td>colourless</td>
</tr>
<tr>
<td>Proteus vulgaris ATCC 13315</td>
<td>50 -100</td>
<td>luxuriant</td>
<td>&gt;=50 %</td>
<td>colourless</td>
</tr>
<tr>
<td>Salmonella Typhi ATCC 6539</td>
<td>50 -100</td>
<td>luxuriant</td>
<td>&gt;=50 %</td>
<td>colourless</td>
</tr>
<tr>
<td>Staphylococcus epidermidis ATCC 12228 (00036*)</td>
<td>&gt;=10⁴</td>
<td>inhibited</td>
<td>0%</td>
<td></td>
</tr>
<tr>
<td>Escherichia coli ATCC 8739 (00012*)</td>
<td>50 -100</td>
<td>luxuriant</td>
<td>&gt;=50 %</td>
<td>pink-red with bile precipitate</td>
</tr>
</tbody>
</table>
### Technical Data

#### Staphylococcus aureus subsp. aureus ATCC 6538 (00032*)
- Growth: inhibited
- Percentage: 0%

#### Salmonella Paratyphi B ATCC 8759
- Growth: luxuriant
- Percentage: ≥50 %
- Colour: colourless

#### Escherichia coli ATCC 25922 (00013*)
- Growth: luxuriant
- Percentage: ≥50 %
- Colour: pink to red with bile precipitate

#### Escherichia coli NCTC 9002
- Growth: luxuriant
- Percentage: ≥50 %
- Colour: pink to red with bile precipitate

#### # Klebsiella aerogenes ATCC 13048 (00175*)
- Growth: luxuriant
- Percentage: ≥50 %

#### Salmonella Typhimurium ATCC 14028 (00031*)
- Growth: luxuriant
- Percentage: ≥50 %

#### Enterococcus faecalis ATCC 29212 (00087*)
- Growth: colourless to pale pink

#### Salmonella Enteritidis ATCC 13076 (00030*)
- Growth: luxuriant
- Percentage: ≥50 %

#### Staphylococcus aureus subsp. aureus ATCC 25923 (00034*)
- Growth: inhibited
- Percentage: 0%

Key: * Corresponding WDCM numbers
- # Formerly known as Enterobacter aerogenes

### 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 clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (6,10).

### Reference

12. The United States Pharmacopoeia, 2018, The United States Pharmacopeial Convention, Rockville, M.D.

Revision: 05/2019

Please refer disclaimer Overleaf.
In vitro diagnostic medical device

CE Marking

Storage temperature

10°C - 30°C

Do not use if package is damaged

HiMedia Laboratories Pvt. Limited,
23 Vadhani Industrial Estate,
LBS Marg, Mumbai-86, MS, India

CE Partner 4U, Esdoornlaan 13, 3951 DB Maarn The Netherlands,
www.cepartner4u.eu

Disclaimer:
User must ensure suitability of the product(s) in their application prior to use. Products conform solely to the information contained in this and other related HiMedia™ publications. The information contained in this publication is based on our research and development work and is to the best of our knowledge true and accurate. HiMedia™ Laboratories Pvt Ltd reserves the right to make changes to specifications and information related to the products at any time. Products are not intended for human or animal or therapeutic use but for laboratory, diagnostic, research or further manufacturing use only, unless otherwise specified. Statements contained herein should not be considered as a warranty of any kind, expressed or implied, and no liability is accepted for infringement of any patents.