**Intended Use:**
MacConkey HiCynth™ Agar w/ CV and NaCl is 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>HiCynth™ Peptone No.3*</td>
<td>17.000</td>
</tr>
<tr>
<td>HiCynth™ Peptone No.5*</td>
<td>3.000</td>
</tr>
<tr>
<td>Lactose</td>
<td>10.000</td>
</tr>
<tr>
<td>Synthetic detergent</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><strong>Final pH (at 25°C)</strong></td>
<td>7.1±0.2</td>
</tr>
</tbody>
</table>

*Formula adjusted, standardized to suit performance parameters
*Chemically defined peptones

**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). MacConkey HiCynth™ Agar w/ CV and NaCl is the modification of regular Macconkey agar prepared by replacing animal and vegetable peptones with chemically defined peptones to avoid BSE/TSE risks associated with animal peptones.

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 synthetic detergent, 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. 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 colorless, transparent and typically do not alter appearance of the medium. HiCynth™ Peptone No.3 and HiCynth™ Peptone No.5 provide the necessary nitrogen compounds, carbon, long chain amino acids, vitamins and also some trace ingredients necessary for the growth of bacteria. Lactose is a fermentable carbohydrate. Sodium chloride maintains the osmotic equilibrium. Synthetic detergent 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><em>Corynebacterium diphtheriae type gravis</em></td>
<td>&gt;=10⁴</td>
<td>inhibited</td>
<td>0%</td>
<td>colourless</td>
</tr>
<tr>
<td><em>Shigella flexneri ATCC 12022 (00126</em>)*</td>
<td>50 -100</td>
<td>fair to good</td>
<td>30 -40%</td>
<td>colourless</td>
</tr>
<tr>
<td><em>Salmonella Paratyphi A ATCC 9150</em></td>
<td>50 -100</td>
<td>luxuriant</td>
<td>&gt;=50%</td>
<td>colourless</td>
</tr>
<tr>
<td><em>Salmonella Abony NCTC 6017 (00029</em>)*</td>
<td>50 -100</td>
<td>luxuriant</td>
<td>&gt;=50%</td>
<td>colourless</td>
</tr>
<tr>
<td><em>Proteus vulgaris ATCC 13315</em></td>
<td>50 -100</td>
<td>luxuriant</td>
<td>&gt;=50%</td>
<td>colourless</td>
</tr>
<tr>
<td><em>Salmonella Typhi ATCC 6539</em></td>
<td>50 -100</td>
<td>luxuriant</td>
<td>&gt;=50%</td>
<td>colourless</td>
</tr>
<tr>
<td><em>Staphylococcus epidermidis ATCC 12228 (00036</em>)*</td>
<td>&gt;=10⁴</td>
<td>inhibited</td>
<td>0%</td>
<td></td>
</tr>
<tr>
<td><em>Escherichia coli ATCC 8739 (00012</em>)*</td>
<td>50 -100</td>
<td>luxuriant</td>
<td>&gt;=50%</td>
<td>pink-red with bile precipitate</td>
</tr>
</tbody>
</table>
### Technical Data

<table>
<thead>
<tr>
<th>Organism</th>
<th>Growth (µl)</th>
<th>Colouration</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Staphylococcus aureus</em> subsp.aureus ATCC 6538</td>
<td>&gt;=10⁴</td>
<td>inhibited</td>
<td>0%</td>
</tr>
<tr>
<td><em>Salmonella Paratyphi B</em> ATCC 8759</td>
<td>50 -100</td>
<td>luxuriant</td>
<td>&gt;=50 %</td>
</tr>
<tr>
<td><em>Escherichia coli ATCC 25922 (00013</em>)*</td>
<td>50 -100</td>
<td>luxuriant</td>
<td>&gt;=50 %</td>
</tr>
<tr>
<td><em>Escherichia coli NCTC 9002</em></td>
<td>50 -100</td>
<td>luxuriant</td>
<td>&gt;=50 %</td>
</tr>
<tr>
<td># Klebsiella aerogenes ATCC 13048 (00175*)</td>
<td>50 -100</td>
<td>luxuriant</td>
<td>&gt;=50 %</td>
</tr>
<tr>
<td><em>Salmonella Typhimurium ATCC 14028 (00031</em>)*</td>
<td>50 -100</td>
<td>luxuriant</td>
<td>&gt;=50 %</td>
</tr>
<tr>
<td><em>Enterococcus faecalis ATCC 29212 (00087</em>)*</td>
<td>50 -100</td>
<td>none - poor</td>
<td>&lt;=10 %</td>
</tr>
<tr>
<td><em>Salmonella Enteritidis ATCC 13076 (00030</em>)*</td>
<td>50 -100</td>
<td>luxuriant</td>
<td>&gt;=50 %</td>
</tr>
<tr>
<td><em>Staphylococcus aureus</em> subsp.aureus ATCC 25923 (00034*)</td>
<td></td>
<td>inhibited</td>
<td>0%</td>
</tr>
</tbody>
</table>

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. 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 : 02/ 2019
HiMedia Laboratories

In vitro diagnostic medical device

CE Marking

Storage temperature

10°C - 30°C

Do not use if package is damaged

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