Deoxycholate Citrate HiCynth™ Agar

Intended Use:
Recommended for the isolation of enteric pathogens particularly *Salmonella* and *Shigella* species.

**Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>HiCynth™ Peptone No.2*</td>
<td>12.000</td>
</tr>
<tr>
<td>HiCynth™ Peptone No.6*</td>
<td>11.000</td>
</tr>
<tr>
<td>Lactose</td>
<td>10.000</td>
</tr>
<tr>
<td>Synthetic detergent</td>
<td>2.000</td>
</tr>
<tr>
<td>Neutral red</td>
<td>0.020</td>
</tr>
<tr>
<td>Sodium citrate</td>
<td>20.000</td>
</tr>
<tr>
<td>Ferric ammonium citrate</td>
<td>2.000</td>
</tr>
<tr>
<td>Agar</td>
<td>13.500</td>
</tr>
<tr>
<td>Final pH (at 25°C)</td>
<td>7.5 ± 0.2</td>
</tr>
</tbody>
</table>

**Formula adjusted, standardized to suit performance parameters**

* Chemically defined peptones

**Directions**
Suspend 70.52 grams in 1000 ml of purified / distilled water. Heat to boiling to dissolve the medium completely. DO NOT AUTOCLAVE. Avoid excessive heating as it is detrimental to the medium. Cool to 45-50°C. Mix well and pour into sterile petri plates.

**Principle And Interpretation**
Deoxycholate Citrate Agar is prepared as per the modified formula of Leifson (4). This medium is used for the isolation and maximum recovery of intestinal pathogens belonging to *Salmonella* and *Shigella* groups from foods (5). Deoxycholate Citrate HiCynth™ Agar is modified by replacing animal and vegetable peptones with chemically defined peptones to avoid risks associated with animal peptones. However, it is recommended to use less inhibitory medium when *Shigella* have to be isolated (1). The selectivity of this medium permits the use of fairly heavy inocula without danger of overgrowth of *Shigella* and *Salmonella* by other microflora. For the routine examination of stool and urine specimens, it is suggested that other media such as MacConkey HiCynth™ Agar (MCD082), Bismuth Sulphite HiCynth™ Agar (MCD027) etc. be used in conjunction with this medium.
HiCynth™ Peptone No.2 and HiCynth™ Peptone No.6 provides nitrogenous and carbonaceous compounds, long chain amino acids, vitamins and other nutrients. Coliform bacteria and gram-positive bacteria are inhibited or greatly suppressed due to synthetic detergent, sodium citrate and ferric ammonium citrate. Dipotassium phosphate buffers the medium. Lactose helps in differentiating enteric bacilli, as lactose fermenters produce red colonies while lactose non-fermenters produce colorless colonies. Coliform bacteria, if present form pink colonies on this medium. The degradation of lactose causes acidification of the medium surrounding the relevant colonies and the pH indicator neutral red changes its colour to red. The reduction of ferric ammonium citrate to iron sulfide is indicated by the formation of black iron sulfide. *Salmonella* and *Shigella* species do not ferment lactose but *Salmonella* may produce H2S, forming colorless colonies with or without black centers. Citrate and iron (Fe) combination has a strong hydrolyzing effect on agar when the medium is heated, producing a soft and unelastic agar. If autoclaved the agar becomes soft and almost impossible to streak.

**Type of specimen**
Clinical- faeces; Food samples
Specimen Collection and Handling
For Clinical samples follow appropriate techniques for handling specimens as per established guidelines (2,3).
For Food samples follow appropriate techniques for handling specimens as per established guidelines (5).
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. Further Biochemical identification is required for confirmation of species.
2. Due to nutritional variations some organisms may show poor growth.
3. Surface colonies of non-lactose fermenters often absorb a little colour (pinkish) from the medium and organisms may be mistaken for coliforms (4).

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 pinkish beige homogeneous free flowing powder
Gelling
Firm, comparable with 1.35% Agar gel.
Colour and Clarity of prepared medium
Reddish orange coloured, clear to slightly opalescent gel forms in Petri plates
Reaction
Reaction of 7.05% w/v aqueous solution at 25°C. pH : 7.5±0.2
pH
7.30-7.70
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>
<th>Colour of Colony</th>
<th>H₂S</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Enterococcus faecalis</em> ATCC 29212 (00087*)</td>
<td>&gt;=10⁴</td>
<td>inhibited</td>
<td>0%</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Escherichia coli</em> ATCC 25922 (00013*)</td>
<td>50-100</td>
<td>poor</td>
<td>20-30%</td>
<td>pink</td>
<td>negative reaction,</td>
</tr>
<tr>
<td><em>Salmonella Enteritidis</em> ATCC 13076 (00030*)</td>
<td>50-100</td>
<td>good-luxuriant</td>
<td>&gt;=50%</td>
<td>colourless</td>
<td>positive reaction,</td>
</tr>
<tr>
<td><em>Salmonella Typhimurium</em> ATCC 14028 (00031*)</td>
<td>50-100</td>
<td>good-luxuriant</td>
<td>&gt;=50%</td>
<td>colourless</td>
<td>black centered colonies, positive reaction,black centered colonies</td>
</tr>
<tr>
<td><em>Shigella flexneri</em> ATCC 12022 (00126*)</td>
<td>50-100</td>
<td>good</td>
<td>40-50%</td>
<td>colourless</td>
<td></td>
</tr>
<tr>
<td><em>Escherichia coli</em> ATCC 8739 (00012*)</td>
<td>50-100</td>
<td>poor</td>
<td>20-30%</td>
<td>pink</td>
<td>negative reaction</td>
</tr>
<tr>
<td><em>Escherichia coli</em> NCTC 9002</td>
<td>50-100</td>
<td>poor</td>
<td>20-30%</td>
<td>pink</td>
<td>negative reaction</td>
</tr>
</tbody>
</table>
Salmonella Abony NCTC 6017 (00029*)
50-100 good-luxuriant >=50% colourless positive reaction, black centered colonies

Staphylococcus aureus subsp. aureus ATCC 25923 (00034*)
>=10^4 inhibited 0%

Key: (*) 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. 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 (2,3).

Reference


Revision: 03/2019

In vitro diagnostic medical device

CE Marking

Storage temperature

Do not use if package is damaged

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Disclaimer:

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