HiCrome™ Enrichment HiCynth™ Broth Base for EC O157:H7

**Intended Use**
Recommended for isolation and selective differentiation of *Escherichia coli* O157:H7 from food, environmental and clinical samples by chromogenic method.

**Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>HiCynth™ peptone No.1#</td>
<td>10.000</td>
</tr>
<tr>
<td>Sorbitol</td>
<td>10.000</td>
</tr>
<tr>
<td>Synthetic detergent No.1</td>
<td>1.500</td>
</tr>
<tr>
<td>Chromogenic mixture</td>
<td>1.300</td>
</tr>
<tr>
<td>Final pH (at 25°C)</td>
<td>7.1±0.2</td>
</tr>
</tbody>
</table>

**Directions**
Suspend 11.4 grams in 500 ml purified / distilled water. Heat if necessary to dissolve the medium completely. Sterilize by autoclaving at 15lbs pressure (121°C) for 15 minutes. For selective isolation of *E.coli* O157:H7, aseptically add the rehydrated contents of 1vial of HiCrome™ EC O157:H7 Selective Supplement I (FD230) to sterile, molten and cooled (at 45-50°C) medium. Mix well and dispense into sterile test tubes.

**Principle And Interpretation**

March and Ratnam (3) reported the inability of *Escherichia coli* O157:H7 to ferment sorbitol while developing Sorbitol MacConkey medium. Subsequently Thomson et al (5) observed the absence of ß-glucuronidase activity in *E.coli* O157:H7 from a variety of samples by direct culture. HiCrome™ Enrichment HiCynth™ Broth Base for EC O157:H7 is a modification of HiCrome™ Enrichment Broth Base for EC O157:H7 wherein animal based peptones are completely replaced with chemically defined peptones to avoid BSE/TSE/GMO risks associated with animal peptones.

The medium contains HiCynth™ peptone No.1 that provides nitrogenous, carbonaceous compounds, long chain amino acids, vitamins and other essential growth nutrients. Sorbitol is the fermentable carbohydrate. Synthetic detergent No.1 inhibits most of the gram-positive organisms. Addition of tellurite (FD230) makes the medium more specific and selective. The bluish colour development by colonies of *E.coli* and *Klebsiella* in the medium is due to the enzymes ß-glucuronidase and ß-D-galactosidase that cleaves the chromogenic substrates present in chromogenic mixture respectively. However *E.coli* O157:H7 gives a purple colour to the medium due to the absence of ß-glucuronidase and its inability to ferment sorbitol.

**Type of specimen**
Clinical samples : stool samples; Food samples.

**Specimen Collection and Handling**

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (1,2). For food samples follow appropriate techniques for sample collection and processing as per guidelines (4). After use, contaminated materials must be sterilized by autoclaving before discarding.

**Warning and Precautions**

In Vitro diagnostic use only. 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. β-glucuronidase is present in 97% of *E. coli* strains, however few *E. coli* may be negative.
2. Some species may show poor growth due to nutritional variations.

**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 yellow coloured, clear solution which may have slight precipitate.

**Reaction**
Reaction of 2.28% w/v aqueous solution at 25°C. pH : 7.1±0.2

**pH**
6.90-7.30

**Cultural Response**
Cultural characteristics observed with added HiCrome™ EC O157:H7 Selective Supplement(FD230), 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>Colour of Medium</th>
<th>Growth (after addition of FD230)</th>
<th>Colour of Medium (after addition of FD230)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Klebsiella pneumoniae</em> ATCC 13883 (00097*)</td>
<td>50-100</td>
<td>good-luxuriant</td>
<td>bluish-green may show slight precipitation of growth</td>
<td>good</td>
<td>bluish green may show slight precipitation of growth</td>
</tr>
<tr>
<td><em>Escherichia coli</em> ATCC 25922 (00013*)</td>
<td>50-100</td>
<td>good-luxuriant</td>
<td>blue may show slight precipitation of growth</td>
<td>inhibited</td>
<td>-</td>
</tr>
<tr>
<td><em>Enterococcus faecalis</em> ATCC 29212 (00087*)</td>
<td>&gt;=10³</td>
<td>inhibited</td>
<td>-</td>
<td>inhibited</td>
<td>-</td>
</tr>
<tr>
<td><em>Escherichia coli</em> O157:H7 (NCTC 12900) (00014*)</td>
<td>good-luxuriant</td>
<td>purple may show slight precipitation of growth</td>
<td>good-luxuriant</td>
<td>purple may show slight precipitation of growth</td>
<td></td>
</tr>
<tr>
<td><em>Cronobacter sakazakii</em> ATCC 12868</td>
<td>50-100</td>
<td>good-luxuriant</td>
<td>white may show slight precipitation of growth</td>
<td>none-poor</td>
<td>colourless may show slight precipitation of growth</td>
</tr>
<tr>
<td><em>Staphylococcus aureus</em> ATCC 25923 (00034*)</td>
<td>&gt;=10³</td>
<td>inhibited</td>
<td>-</td>
<td>inhibited</td>
<td>-</td>
</tr>
<tr>
<td><em>Salmonella Enteritidis</em> ATCC 13076 (00030*)</td>
<td>good-luxuriant</td>
<td>light green may good show slight precipitation of growth</td>
<td>light green may show slight precipitation of growth</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td><em>Shigella flexneri</em> ATCC 12022 (00126*)</td>
<td>50-100</td>
<td>good</td>
<td>colourless</td>
<td>inhibited</td>
<td>-</td>
</tr>
</tbody>
</table>

Key : * Corresponding WDCM numbers, # Formerly known as *Enterobacter sakazakii*.

**Storage and Shelf Life**

Store dehydrated powder and prepared medium on receipt at 2-8°C. Use before expiry period 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 (1,2).

**Reference**