Tryptone Broth, HiVeg™ (Tryptone Water, HiVeg™)

**Intended use**
Tryptone Broth, HiVeg™ (Tryptone Water, HiVeg™) is used for the detection of indole producing microorganisms.

**Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>HiVeg™ hydrolysate</td>
<td>10.000</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>5.000</td>
</tr>
<tr>
<td>Final pH (at 25°C)</td>
<td>7.5±0.2</td>
</tr>
</tbody>
</table>

**Directions**
Dissolve 15 grams in 1000 ml purified/distilled water. Heat if necessary to dissolve the medium completely. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

**Principle And Interpretation**
Tryptone Broth, HiVeg™ is prepared by using HiVeg™ Hydrolysate in place of tryptone which makes the medium free of BSE/TSE risks. HiVeg™ hydrolysate is a good substrate for indole production because of its high tryptophan content. Thus it can be used as an alternative of tryptone. Tryptone Water is recommended by APHA (1) for detection of indole production by coliforms, which is a key feature in differentiation of bacteria. A slight modification of Tryptone Water (M463I) is recommended by ISO committee (4) for the same purpose. This test demonstrates the ability of certain bacteria to hydrolyze the amino acid tryptophan to indole mediated by suitable enzymes. Liberated indole gets accumulated in the medium (2). The indole produced can be detected by either Kovac's or Ehrlich's reagent (3). Indole combines with the aldehyde present in the above reagent to give red colour in the alcohol layer. The alcohol layer extracts and concentrates the red colour complex.

**Type of specimen**
Water samples, Clinical samples- Faeces, Urine.

**Specimen Collection and Handling**
For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (1). For clinical samples follow appropriate techniques for handling specimens as per established guidelines (5, 6). 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. Biochemical confirmation is necessary for complete identification of the organisms.
**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**
Yellow coloured clear solution

**Reaction**
Reaction of 1.5% 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>Indole production</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Escherichia coli ATCC 25922</em></td>
<td>50-100</td>
<td>luxuriant</td>
<td>Positive, red ring at the interface of the medium</td>
</tr>
<tr>
<td><em>Klebsiella aerogenes ATCC 13048</em></td>
<td>50-100</td>
<td>luxuriant</td>
<td>Negative, no colour development / cloudy ring</td>
</tr>
<tr>
<td><em>Klebsiella pneumoniae ATCC 13883</em></td>
<td>50-100</td>
<td>luxuriant</td>
<td>Negative, no colour development / cloudy ring</td>
</tr>
</tbody>
</table>

Key : (#) Formerly known as *Enterobacter aerogenes*  (*) Corresponding WDCM numbers.

**Storage and Shelf Life**

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°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 (5,6).

**Reference**