Buffered HiCynth™ Peptone Water

**Intended use**
Recommended is used as pre-enrichment medium for increasing the recovery of injured *Salmonella* species from foods prior to selective enrichment and isolation. The composition and performance criteria of this medium are as per the applications laid down in ISO 6579-2017.

**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>
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<tr>
<td>Sodium chloride</td>
<td>5.000</td>
</tr>
<tr>
<td>Disodium hydrogen phosphate.12H2O</td>
<td>9.000</td>
</tr>
<tr>
<td>Potassium dihydrogen phosphate</td>
<td>1.500</td>
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<tr>
<td>pH ( at 25°C)</td>
<td>7.0±0.2</td>
</tr>
</tbody>
</table>

**Directions**
Suspend 20.07 grams (equivalent weight of dehydrated medium) in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense as desired and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

**Principle And Interpretation**
Microorganisms that are subjected to environmental stresses may become structurally or metabolically damaged or injured. These microorganisms are unable to replicate in selective environments. Therefore these injured organisms must be resuscitated or permitted to repair the damage by incubation in an appropriate, non-selective environment (7). Edel and Kampelmacher (2) noted that sub-lethal injury to *Salmonella* may occur in many food preservation processes. Enriching injured cells in Lactose Broth (pH 6.9) may be further detrimental to their recovery (1). Pre-enrichment in Buffered Peptone No.1 Water (M14941) at 35°C for 18-24 hours results in repair of injured cells (6). The buffering system prevents bacterial damage due to change in the pH of the medium. Recently ISO committee has also recommended this pre-enrichment medium for the detection of *Enterobacteriaceae* from food stuffs and other materials (3).

Buffered HiCynth™ Peptone Water is prepared by completely replacing animal or vegetable based peptones with chemically defined peptone to avoid BSE/TSE/GMO risks associated with animal peptones. HiCynth™ Peptone No. 1 supplies nitrogen and carbon compounds, long chain amino acids, vitamins and other essential nutrients. Phosphates buffers the medium. Sodium chloride maintains osmotic balance.

Inoculate 10 grams specimen in 50 ml of Buffered HiCynth™ Peptone Water (MCD1494I) and incubate at 35°C for 18 hours. Transfer 10 ml from this medium to 100 ml of Mueller Kauffman Tetrathionate Novobiocin Broth Base (M1496I) and Rappaport Vassiliadis Soya Broth (RVS Broth) (M1491) and incubate at 43°C for 24-48 hours and then subculture on selective media like XLD Agar, Modified (M031I). Examine the plates for colonies of *Salmonella* species.

**Type of specimen**
Food and dairy samples

**Specimen Collection and Handling**
For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (3). After use, contaminated materials must be sterilized by autoclaving before discarding.

**Warning and Precautions**
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 specimens. Safety guidelines may be referred in individual safety data sheets.

**Limitations :**
1. Due to nutritional variations some strains may show poor growth.

**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 without any precipitate

**Reaction**
Reaction of 2.0% w/v aqueous solution at 25°C. pH : 7.0±0.2

**pH**
6.80-7.20

**Cultural Response**
Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.(Recovery is observed on XLD Agar, M031I)

<table>
<thead>
<tr>
<th>Organism</th>
<th>Inoculum (CFU)</th>
<th>Growth</th>
<th>Recovery</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Salmonella Enteritidis</em> ATCC 13076 (00030*)</td>
<td>50-100</td>
<td>luxuriant</td>
<td>&gt;=50%</td>
</tr>
<tr>
<td><em>Salmonella Typhi ATCC 6539</em></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>Escherichia coli ATCC 25922 (00013</em>)*</td>
<td>50-100</td>
<td>fair-good</td>
<td>30-40%</td>
</tr>
<tr>
<td><em>Pseudomonas aeruginosa ATCC 27853 (00025</em>)*</td>
<td>50-100</td>
<td>luxuriant</td>
<td>&gt;=50%</td>
</tr>
</tbody>
</table>

Key : * 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. 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 sample must be decontaminated and disposed of in accordance with current laboratory techniques (4,5).

**Reference**

**Disclaimer:**
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