**Intended Use:**
For enrichment of *Shigella* from pharmaceutical products in accordance with Indian pharmacopoeia 2017.

**Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Polypeptone peptone</td>
<td>20.000</td>
</tr>
<tr>
<td>Glucose</td>
<td>1.000</td>
</tr>
<tr>
<td>Sodium citrate</td>
<td>2.000</td>
</tr>
<tr>
<td>Sodium deoxycholate</td>
<td>0.500</td>
</tr>
<tr>
<td>Di-potassium hydrogen phosphate</td>
<td>4.000</td>
</tr>
<tr>
<td>Mono potassium dihydrogen phosphate</td>
<td>1.500</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>5.000</td>
</tr>
</tbody>
</table>

**Final pH (at 25°C)**: 7.0±0.2

**Directions**
Suspend 34.00 grams in 1000 ml purified/distilled water. Mix well and allow to stand for 15 minutes. With continuous stirring bring gently to boil and maintain at boiling till completely dissolved. DO NOT AUTOCLAVE. Dispense in sterile test tubes or flasks as desired.

**Principle And Interpretation**
GN Broth is recommended by the Indian Pharmacopoeia (1) for the selective isolation of *Shigella* species with subsequent isolation on a selective medium, XLD Agar (MH031). Croft and Miller isolated more strains of *Shigella* from rectal swabs using this medium (2). Taylor and Schelhart showed the superiority of GN Broth to selenite enrichment media for isolation of *Shigella* (3). Hajna (4, 5) also suggested the enrichment of organisms from rectal swabs in this medium 1-6 hours before plating on solid media.

The medium contains polypeptone peptone, which provides amino acids and other nitrogenous substances to support bacterial growth. The combination of sodium citrate and sodium deoxycholate inhibit gram-positive and some gram-negative bacteria such as coliforms. Phosphates serve as a buffering system. Sodium chloride maintains osmotic equilibrium. *Proteus, Pseudomonas* and coliforms do not overgrow *Salmonella* and *Shigella* in GN Broth during the first 6 hours of incubation. This enrichment broth should be used in conjunction with selective and nonselective plating media to increase the probability of isolating pathogens (6,7,8).

**Type of specimen**
Pharmaceutical samples

**Specimen Collection and Handling**
For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per guidelines (1). 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. Further isolation and biochemical tests must be carried out for confirmation.
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 amber coloured, clear to slightly opalescent solution in tubes.

Reaction
Reaction of 3.9% w/v aqueous solution at 25°C. pH : 7.0±0.2

pH
6.80-7.20

Cultural Response
Cultural characteristics observed after inoculation in GN Broth and then subculture on XLD Agar and incubation at 30-35°C for 24-48 hours.

Growth promoting properties
Growth promotion was carried out in accordance with the method of IP.Clearly visible growth of microorganism comparable to that previously obtained with previous tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating <=100 cfu (at 30-35°C for <=24 hours).

Inhibitory properties
No growth of the test microorganism occurs for the specified temperature for not less than longest period of time specified inoculating >100 cfu (at least 100 cfu) (at 30-35°C for >=48 hours).

Cultural Response

<table>
<thead>
<tr>
<th>Organism</th>
<th>Inoculum (CFU)</th>
<th>Growth in GN broth</th>
<th>Recovery on XLD Agar</th>
<th>Recovery</th>
<th>Colour of colony</th>
</tr>
</thead>
<tbody>
<tr>
<td>Growth promoting</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Shigella boydii ATCC 8700</td>
<td>50 -100</td>
<td>good</td>
<td>good-luxuriant</td>
<td>&gt;=50 %</td>
<td>red translucent</td>
</tr>
<tr>
<td>Inhibitory</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Staphylococcus aureus</td>
<td>&gt;=10³</td>
<td>inhibited</td>
<td></td>
<td>&lt;=0 %</td>
<td></td>
</tr>
<tr>
<td>ATCC 6538 (00193*)</td>
<td></td>
<td></td>
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<td></td>
<td></td>
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</tbody>
</table>

* - Corresponding WDCM numbers

Storage and Shelf Life
Store below 30°C in a tightly closed container and the prepared medium at 2-8°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 (6,7).

Reference
1. Indian Pharmacopoeia, 2017, Ministry of Health and Family Welfare, Govt. of India.,