GN Broth • GN Broth, Hajna

Intended Use
GN Broth is used for the selective enrichment of *Salmonella* and *Shigella*.

Summary and Explanation
GN (Gram Negative) Broth was developed by Hajna as an enrichment medium for the recovery of *Salmonella* and *Shigella* from clinical and nonclinical specimens.\(^1\) Croft and Miller succeeded in isolating more *Shigella* strains by use of this medium, rather than by direct streaking.\(^3\) Taylor and Schelhart reported that GN Broth enhanced the isolation of enteric pathogens, producing a 53% increase in *Shigella* and a 36% increase in *Salmonella* as compared to direct streaking.\(^4\) In another study, Taylor and Schelhart showed that GN Broth was superior to selenite enrichment media for the isolation of *Shigella*.\(^5\) GN Broth currently is recommended for use in the microbiological examination of foods.\(^6\)

Principles of the Procedure
Peptones provide amino acids and other nitrogenous substances to support bacterial growth. Mannitol and dextrose are sources of energy. Mannitol is provided in a higher concentration than dextrose to enhance the growth of mannitol-fermenting species, such as *Salmonella* and *Shigella*, and limit the growth of *Proteus* and other dextrose-fermenting bacteria. Phosphate buffers are incorporated to maintain the pH of the medium. Sodium citrate and sodium desoxycholate are added to inhibit gram-positive and some gram-negative bacteria. *Proteus, Pseudomonas* and coliforms do not overgrow *Salmonella* and *Shigella* in GN Broth during the first 6 hours of incubation.

User Quality Control

NOTE: Differences in the Identity Specifications and Cultural Response testing for media offered as both Difco™ and BBL™ brands may reflect differences in the development and testing of media for industrial and clinical applications, per the referenced publications.

### Identity Specifications

#### Difco™ GN Broth, Hajna
- **Dehydrated Appearance**: Off-white to light tan, free-flowing, homogeneous.
- **Solution**: 3.9% solution, soluble in purified water. Solution is light amber, clear to slightly opalescent.
- **Prepared Appearance**: Light amber, clear to slightly opalescent.
- **Reaction of 3.9% Solution at 25°C**: pH 7.0 ± 0.2

#### BBL™ GN Broth
- **Dehydrated Appearance**: Fine, dry, homogeneous, free of extraneous material.
- **Solution**: 3.9% solution, soluble in purified water. Solution is pale to medium, tan to yellow, clear to slightly hazy.
- **Prepared Appearance**: Pale to medium, tan to yellow, clear to slightly hazy.
- **Reaction of 3.9% Solution at 25°C**: pH 7.0 ± 0.2

### Cultural Response

#### Difco™ GN Broth, Hajna
Prepare the medium per label directions. Inoculate and incubate at 35 ± 2°C for 18-24 hours.

<table>
<thead>
<tr>
<th>ORGANISM</th>
<th>ATCC*</th>
<th>INOCULUM CFU</th>
<th>RECOVERY</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Enterococcus faecalis</em></td>
<td>19433</td>
<td>10⁰-2×10⁴</td>
<td>None to poor</td>
</tr>
<tr>
<td><em>Escherichia coli</em></td>
<td>25922</td>
<td>10⁰-10⁴</td>
<td>Good</td>
</tr>
<tr>
<td><em>Salmonella enterica</em></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>subsp. <em>Typhimurium</em></td>
<td>14028</td>
<td>10⁰-10⁴</td>
<td>Good</td>
</tr>
<tr>
<td><em>Shigella flexneri</em></td>
<td>12022</td>
<td>10⁰-10⁴</td>
<td>Good</td>
</tr>
</tbody>
</table>

#### BBL™ GN Broth
Prepare the medium per label directions. Inoculate and incubate at 35 ± 2°C; subculture to MacConkey II Agar after 6 hours and again after 18-24 hours of incubation. Incubate subculture plates at 35 ± 2°C for 18-24 hours.

<table>
<thead>
<tr>
<th>ORGANISM</th>
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<td>14028</td>
<td>10⁰-10⁴</td>
<td>Good</td>
</tr>
<tr>
<td><em>Shigella sonnei</em></td>
<td>9290</td>
<td>10⁰-10⁴</td>
<td>Good</td>
</tr>
</tbody>
</table>
**Formulae**

**Difco™ GN Broth, Hajna**

Approximate Formula* Per Liter

- Pancreatic Digest of Casein ....................................... 12.0 g
- Proteose Peptone No. 3 ............................................... 8.0 g
- Dextrose ..................................................................... 1.0 g
- D-Mannitol ................................................................. 2.0 g
- Sodium Citrate ............................................................ 5.0 g
- Sodium Desoxycholate ................................................ 0.5 g
- Dipotassium Phosphate ............................................... 4.0 g
- Monopotassium Phosphate ......................................... 1.5 g
- Sodium Chloride ......................................................... 5.0 g

**BBL™ GN Broth**

Approximate Formula* Per Liter

- Pancreatic Digest of Casein ....................................... 10.0 g
- Peptic Digest of Animal Tissue ................................... 10.0 g
- Dextrose ..................................................................... 1.0 g
- D-Mannitol ................................................................. 2.0 g
- Sodium Citrate ............................................................ 5.0 g
- Sodium Desoxycholate ................................................ 0.5 g
- Dipotassium Phosphate ............................................... 4.0 g
- Monopotassium Phosphate ......................................... 1.5 g
- Sodium Chloride ......................................................... 5.0 g

*Adjusted and/or supplemented as required to meet performance criteria.

**Directions for Preparation from Dehydrated Product**

1. Suspend 39 g of the powder in 1 L of purified water. Mix thoroughly.
2. Dispense and autoclave at 121°C for 15 minutes.
3. Alternatively, the broth may be steamed for 30 minutes at 100°C.
4. Test samples of the finished product for performance using stable, typical control cultures.

**Procedure**

Inoculate the broth as soon as possible after the specimen arrives at the laboratory. Swab specimens may be inserted directly into the broth. For stool specimens, use 1 g of feces or 1 mL of liquid stool per tube. Consult appropriate references for information about the processing and inoculation of other clinical specimens or food samples. 

Incubate the tubes with loosened caps at 35 ± 2°C and subculture onto selective and differential media after 6-8 hours of incubation and again after 18-24 hours of incubation.

**Expected Results**

Growth in broth media is indicated by turbidity compared to an uninoculated control. Subculture onto appropriate selective and differential media to isolate pathogens for identification.

**Limitation of the Procedure**

Enrichment broths should not be used as the sole isolation medium. They are to be used in conjunction with selective and nonselective plating media to increase the probability of isolating pathogens, especially when they may be present in small numbers. Consult references for detailed information and recommended procedures.

**References**


**Availability**

**Difco™ GN Broth, Hajna**

<table>
<thead>
<tr>
<th>BS12</th>
<th>Cat. No. 248610</th>
<th>Dehydrated – 500 g</th>
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<td>BS12</td>
<td>Cat. No. 211279</td>
<td>Dehydrated – 500 g</td>
</tr>
<tr>
<td></td>
<td>221729</td>
<td>Prepared Tubes, 8 mL (K Tubes) – Pkg. of 10*</td>
</tr>
<tr>
<td></td>
<td>221730</td>
<td>Prepared Tubes, 8 mL (K Tubes) – Ctn. of 100*</td>
</tr>
</tbody>
</table>

*Store at 2-8°C.