Broth Medium I (Tetrathionate Bile Brilliant Green Broth)  

M1255B

Tetrathionate Bile Brilliant Green Broth (Broth Medium I) is used for isolation and identification of *Salmonellae* in accordance with British Pharmacopoeia, 2009.

**Composition**

*Ingredients* | **Gms / Litre**
--- | ---
Peptone | 8.600
Ox bile dried | 8.000
Sodium chloride | 6.400
Calcium carbonate | 20.000
Potassium tetrathionate | 20.000
Brilliant green | 0.070

**pH** after heating (at 25°C) | 7.0±0.2

**Formula adjusted, standardized to suit performance parameters**

**Directions**

Suspend 63.07 grams in 1000 ml purified/distilled water. Heat just to boiling. DO NOT AUTOCLAVE OR REHEAT. Dispense as desired.

*Note: Due to presence of Calcium Carbonate, the prepared medium forms opalescent solution with white precipitate.*

**Principle And Interpretation**

Tetrathionate Bile Brilliant Green Broth cited as Broth Medium I is prepared as per the recommendation of British Pharmacopoeia. (1) Is used for isolation and identification of *Salmonella* species in the tests prescribed for sterility checking in the Pharmacopoeia. It is also used to detect *Salmonella* from pharmaceutical, foods, water and other materials of sanitary importance.

Peptone provides nitrogenous nutrients to the *Salmonellae*. Brilliant green and ox-bile inhibit both gram-positive as well as some selected gram-negative organisms. Potassium tetrathionate inhibits normal flora of faecal specimens. Sodium chloride helps in maintaining osmotic equilibrium. Calcium carbonate neutralizes the acids produced by reduction of tetrathionate.

Medium is not suitable for growth of *Salmonella* Typhi and *Salmonella* Paratyphi (2). The sample is initially enriched in Broth Medium I and incubated at 43-45°C for 18-24 hours. The enriched sample is then subcultured on any two of following Agar medium J, Agar medium K and Agar medium L.

**Quality Control**

**Appearance**

Light yellow to pale green homogeneous free flowing powder.

**Colour and Clarity of prepared medium**

Bluish green coloured opalescent solution with white precipitate.

**pH**

6.80-7.20

**Cultural Response**

M1255B: Cultural characteristics observed after enrichment in Broth Medium I at 41-43°C for 18-24 hours, and then subcultured on Agar Medium K (M031B) and Agar Medium L (M016B) and incubated at 35-37°C for specified period.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Inoculum (CFU)</th>
<th>Growth</th>
<th>Observed Lot value (CFU)</th>
<th>Recovery</th>
<th>Colour of Colony</th>
<th>Incubation period</th>
</tr>
</thead>
</table>

**Cultural Response**

Please refer disclaimer Overleaf.
## Growth on Agar Medium

### K
- **Salmonella Typhimurium ATCC 14028**
  - 50 -100 luxuriant
  - 25 -100 >=50 % red with black centres
  - 18 -72 hrs
- **Salmonella Abony NCTC 6017**
  - 50 -100 good-luxuriant
  - 25 -100 >=50 % red with black centres
  - 18 -72 hrs
- **Salmonella Enteritidis ATCC 50 -100**
  - luxuriant
  - 25 -100 >=50 % red with black centres
  - 18 -72 hrs
- **Staphylococcus aureus ATCC 6538**
  - >=10³ inhibited
  - 0 0%
  - >=72 hrs
- **Escherichia coli ATCC 8739 50 -100**
  - fair
  - 10 -30 20 -30 % yellow
  - 18 -72 hrs

### L
- **Salmonella Typhimurium ATCC 14028**
  - 50 -100 luxuriant
  - 25 -100 >=50 % pinkish white
  - 18 -72 hrs
- **Salmonella Abony NCTC 6017**
  - 50 -100 luxuriant
  - 25 -100 >=50 % pinkish white
  - 18 -72 hrs
- **Salmonella Enteritidis ATCC 50 -100**
  - luxuriant
  - 25 -100 >=50 % pinkish white
  - 18 -72 hrs
- **Staphylococcus aureus ATCC 6538**
  - >=10³ inhibited
  - 0 0%
  - >=72 hrs
- **Escherichia coli ATCC 8739 50 -100**
  - fair
  - 10 -30 20 -30 % yellow
  - 18 -72 hrs

## Storage and Shelf Life
Store below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label.

## Reference

Revision : 1 / 2011

**Disclaimer:**

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