Slanetz and Bartley Medium w/o TTC

Slanetz and Bartley Medium is recommended for detection and enumeration of faecal Streptococci by membrane filtration technique.

**Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tryptose</td>
<td>20.000</td>
</tr>
<tr>
<td>Yeast extract</td>
<td>5.000</td>
</tr>
<tr>
<td>Dextrose</td>
<td>2.000</td>
</tr>
<tr>
<td>Disodium phosphate</td>
<td>4.000</td>
</tr>
<tr>
<td>Sodium azide</td>
<td>0.400</td>
</tr>
<tr>
<td>Agar</td>
<td>15.000</td>
</tr>
<tr>
<td>Final pH (at 25°C)</td>
<td>7.2±0.2</td>
</tr>
</tbody>
</table>

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

**Directions**

Suspend 46.4 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. DO NOT AUTOCLAVE OR OVERHEAT. Excessive heating is detrimental. If desired add sterile 1% TTC solution (FD057) to the medium.

Warning: Sodium azide has a tendency to form explosive metal-azides with plumbing materials. It is advisable to use enough water to flush off the disposables.

**Principle And Interpretation**

Slanetz and Bartley Medium was originally devised by Slanetz and Bartley (1) for the detection and enumeration of Enterococci by membrane filtration technique. It can be also used as a direct plating medium (2, 3). M612A media formulation is devoid of triphenyl tetrazolium chloride that is present in M612.

Tryptose and yeast extract serves as the source of essential nutrients to the organisms. The medium is highly selective for Enterococci. Sodium azide has inhibitory effect on gram-negative organisms. If Triphenyl Tetrazolium Chloride(TTC) is added to the medium, it is reduced to the insoluble formazan inside the bacterial cell forming dark red-coloured colonies. When the medium is incubated at higher temperature (44-45°C), all red or maroon colonies can be considered as presumptive Enterococci (4, 5).

The Department of Health (6) has recommended similar medium with TTC to be used for enumeration of Enterococci in water supplies. Water is filtered through a membrane filter, which is then placed on the surface of the Slanetz and Bartley Medium plates and incubated at 35°C for 4 hours and then at 44-45°C for 44-48 hours. Red or maroon colonies are counted as Enterococci. The preliminary incubation at 35°C helps for the recovery of stressed organisms. Not all the species reduce TTC; hence pale colonies also should be considered.

For analyzing food samples, it is homogenized and diluted with normal saline to give countable 15-150 colonies on each Petri plate when spread on agar surface and incubated at 35°C for 48 hours. Pink or dark red colonies with a narrow whitish border are counted (3).

**Quality Control**

**Appearance**
Cream to yellow homogeneous free flowing powder

**Gelling**
Firm, comparable with 1.5% Agar gel

**Colour and Clarity of prepared medium**
Light yellow coloured clear to slightly opalescent gel forms in Petri plates

**Reaction**

Please refer disclaimer Overleaf.
Reaction of 4.64% w/v aqueous solution at 25°C. pH : 7.2±0.2

**pH**
7.00-7.40

**Cultural Response**
M612A: Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours with added TTC solution (FD057).

<table>
<thead>
<tr>
<th>Organism</th>
<th>Inoculum (CFU)</th>
<th>Growth</th>
<th>Recovery</th>
<th>Colour of colony</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Escherichia coli</em> ATCC 25922</td>
<td>&gt;=10³</td>
<td>inhibited</td>
<td>0%</td>
<td></td>
</tr>
<tr>
<td><em>Enterococcus faecalis</em> ATCC 50-100</td>
<td>good-luxuriant</td>
<td>&gt;=50%</td>
<td>red or maroon</td>
<td></td>
</tr>
</tbody>
</table>

**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**