**Mannitol Motility Nitrate Medium**

Mannitol Motility Nitrate Medium is used for studying mannitol fermentation, nitrate reduction and motility of bacteria.

### Composition**

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Casein enzymic hydrolysate</td>
<td>10.000</td>
</tr>
<tr>
<td>Potassium nitrate</td>
<td>1.000</td>
</tr>
<tr>
<td>Mannitol</td>
<td>7.500</td>
</tr>
<tr>
<td>Phenol red</td>
<td>0.040</td>
</tr>
<tr>
<td>Agar</td>
<td>3.500</td>
</tr>
<tr>
<td>Final pH (at 25°C)</td>
<td>7.6±0.2</td>
</tr>
</tbody>
</table>

**Formula adjusted, standardized to suit performance parameters

### Directions

Suspend 22.04 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Dispense into test tubes. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool the medium in an upright position.

### Principle And Interpretation

Mannitol Motility Nitrate Medium is designed to differentiate bacteria on the basis of their motility, ability to ferment mannitol and reduce nitrate (1). The highly nutritious casein enzymic hydrolysate supports luxuriant growth of bacteria. Semisolid nature of the medium due to 0.35% agar helps to detect motility. Motile bacteria produce diffused growth throughout the medium while non motile bacteria grow only along the line of inoculation. Combination of mannitol and phenol red helps differentiation of mannitol fermenting bacteria which turns the medium yellow.

Reduction of nitrate is generally an anaerobic respiration in which an organism derives its oxygen form nitrate. Members of *Enterobacteriaceae* characteristically reduce nitrate to nitrite which reacts with sulfanilic acid and dimethyl-1-napthylamine to produce the red colour.

### Quality Control

**Appearance**

Light yellow to pink homogeneous free flowing powder

**Gelling**

Semisolid, comparable with 0.35% Agar gel.

**Colour and Clarity of prepared medium**

Red coloured clear to slightly opalescent semisolid gel forms in tubes

**Reaction**

Reaction of 2.20% w/v aqueous solution at 25°C. pH : 7.6±0.2

**pH**

7.40-7.80

### Cultural Response

M1320: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Growth</th>
<th>Mannitol fermentation</th>
<th>Motility</th>
<th>Nitrate reduction</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Escherichia coli ATCC 35218</em></td>
<td>luxuriant</td>
<td>Positive reaction, yellow growth away from stabline causing turbidity</td>
<td>Positive,</td>
<td>Positive reaction red colour developed within 1-2 minutes</td>
</tr>
</tbody>
</table>
### Proteus vulgaris ATCC 13315
- **luxuriant**
- **Negative reaction, no colour change or red**
- **Positive reaction, growth away from stabline causing turbidity**
- **Positive reaction, red colour developed within 1-2 minutes**

### Salmonella Typhi ATCC 6539
- **luxuriant**
- **Positive reaction, yellow colour**
- **Positive reaction, growth away from stabline causing turbidity**
- **Positive reaction, red colour developed within 1-2 minutes**

### Shigella sonnei ATCC 25931 luxuriant
- **Positive reaction, yellow colour**
- **Negative, growth along the stabline, surrounding medium remains clear**
- **Positive reaction, red colour developed within 1-2 minutes**

### Staphylococcus aureus ATCC 25923
- **luxuriant**
- **Positive reaction, yellow colour**
- **Negative, growth along the stabline, surrounding medium remains clear**
- **Positive reaction, red colour developed within 1-2 minutes**

### Staphylococcus epidermidis ATCC 12228
- **luxuriant**
- **Negative reaction, no colour change or red**
- **Negative, growth along the stabline, surrounding medium remains clear**
- **Positive reaction, red colour developed within 1-2 minutes**

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

### Reference

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Disclaimer:

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