Fermentation Medium for Staphylococcus and Micrococcus, w/ 0.2% Agar

Fermentation Medium for Staphylococcus and Micrococcus, w/ 0.2% Agar is used for studying fermentation by *Staphylococcus* species in accordance with FDA BAM, 1998.

**Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tryptone</td>
<td>10.000</td>
</tr>
<tr>
<td>Yeast extract</td>
<td>1.000</td>
</tr>
<tr>
<td>Glucose</td>
<td>10.000</td>
</tr>
<tr>
<td>Bromocresol purple</td>
<td>0.040</td>
</tr>
<tr>
<td>Agar</td>
<td>2.000</td>
</tr>
<tr>
<td>Final pH (at 25°C)</td>
<td>7.0±0.2</td>
</tr>
</tbody>
</table>

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

**Directions**

Suspend 23.04 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Dispense in tubes and sterilize by autoclaving at 10 lbs pressure (115°C) for 20 minutes. Allow tubed medium to cool in an upright position.

**Principle And Interpretation**

Fermentation Medium for Staphylococcus and Micrococcus, w/ 0.2% Agar is used for studying the fermentation characteristics of *Staphylococcus* species in accordance with FDA BAM, 1998 (1). Staphylococci and Micrococi are the most frequently encountered cocci in the clinical laboratory. Both are gram positive and catalase positive. Ability to ferment glucose has served as the basis for differentiating staphylococci from the micrococci that lacks the ability to ferment glucose (2). *Staphylococcus aureus* is a primary pathogen, which may be associated with severe infection. Micrococi are generally strict aerobes and can reduce nitrate. Fermentation Medium for *Staphylococcus* and *Micrococcus* is recommended for differentiation of these two organisms on the basis of glucose fermentation (3, 4).

According to the BAM protocol, total plate count of the suspected sample is carried out using Baird Parker Agar (M043). Suspected colonies of *S. aureus* are inoculated into Fermentation Medium for *Staphylococcus* and *Micrococcus*, w/ 0.2% Agar. Make sure that the inoculum reaches the bottom of the tube. Overlay the surface of agar with a 25mm layer of sterile paraffin oil. Incubate the tubes for 5 days at 37°C. Acid production is indicated by the change in colour of the medium to yellow, indicating presence of *S. aureus*. Run controls simultaneously.

Tryptone and yeast extract provide necessary nitrogenous nutrients for the organisms. Glucose is the fermentable carbohydrate source in the medium. Bromo cresol purple is the pH indicator. Incorporation of small amount of agar in this medium helps to create anaerobic condition in the depths of the tubes.

**Quality Control**

**Appearance**

Light yellow to greenish yellow homogeneous free flowing powder

**Gelling**

Semisolid, comparable with 0.2% Agar gel.

**Colour and Clarity of prepared medium**

Purple coloured clear to slightly opalescent gel forms in tubes as butts

**Reaction**

Reaction of 2.3% w/v aqueous solution at 25°C. pH : 7.0±0.2

**pH**

6.80-7.20
Cultural Response
M827F: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Inoculum (CFU)</th>
<th>Growth</th>
<th>Acid production</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Micrococcus luteus</em> ATCC 10240</td>
<td>50-100</td>
<td>good-luxuriant</td>
<td>negative reaction, no colour change</td>
</tr>
<tr>
<td><em>Staphylococcus aureus</em> ATCC 25923</td>
<td>50-100</td>
<td>good-luxuriant</td>
<td>positive reaction, yellow colour</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