Lysine Iron Agar Slant in tubes

Intended use
Recommended for detection of enteric organism especially *Salmonella Arizonae*, based on their ability to decarboxylate or deaminate lysine and to form hydrogen sulphide (H₂S).

Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peptone</td>
<td>5.000</td>
</tr>
<tr>
<td>Yeast extract</td>
<td>3.000</td>
</tr>
<tr>
<td>Dextrose (Glucose)</td>
<td>1.000</td>
</tr>
<tr>
<td>L-Lysine</td>
<td>10.000</td>
</tr>
<tr>
<td>Ferric ammonium citrate</td>
<td>0.500</td>
</tr>
<tr>
<td>Sodium thiosulphate</td>
<td>0.040</td>
</tr>
<tr>
<td>Bromocresol purple</td>
<td>0.020</td>
</tr>
<tr>
<td>Agar</td>
<td>15.000</td>
</tr>
<tr>
<td>Final pH (at 25°C)</td>
<td>6.7±0.2</td>
</tr>
</tbody>
</table>

**Formula adjusted, standardized to suit performance parameters

Directions
Streak the test inoculum aseptically into the slant and incubate at appropriate conditions. Incubate the slants at 30-35°C for 18-24 hours.

Principle And Interpretation
Lysine Iron Agar was developed by Edwards and Fife (1) to detect lactose fermenting *Salmonella* species. *Salmonella* are known to decarboxylate lysine rapidly and produce large amounts of hydrogen sulphide (2, 7). This medium is a sensitive medium for the detection of lactose fermenting and lactose non-fermenting *Salmonella* species. Many strains of this group ferment lactose very rapidly thus suppressing H₂S production on Triple Sugar Iron Agar (M021). So there is a possibility that the organisms frequently found in food poisoning outbreaks could be overlooked. Thatcher and Clark (8) described the isolation of *Salmonella* species from foods from selective agar and to inoculate it on Lysine Iron Agar and Triple Sugar Iron (M021) together. Using these two media greater discrimination can be made between coliform organisms e.g. *Escherichia coli* and *Shigella* species (3, 5).

Peptone and yeast extract provide essential nutrients. Dextrose is a source of fermentable carbohydrate. Ferric ammonium citrate and sodium thiosulphate are indicators of H₂S formation. Cultures that produce hydrogen sulphide cause blackening of the medium due to ferrous sulphide production. Lysine decarboxylation causes an alkaline reaction (purple colour) to give the amine cadaverine and the organisms which do not decarboxylate lysine, produce acid butt (yellow colour). Organisms that deaminate lysine, form alpha - ketocarboxylic acid, which reacts with iron salt near the surface of the medium under the influence of oxygen to form reddish-brown compound. The medium is stabbed to the base of the butt and streaked on slant.

Type of specimen
Isolated organism

Specimen Collection and Handling
After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions
Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.
Performance and Evaluation
Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control
Appearance
Sterile Lysine Iron Agar Slant in glass tube.

Colour of medium
Purple coloured slant

Quantity of medium
7ml of medium in glass tube

Reaction
6.50-6.90

Sterility Test
Passes release criteria

Cultural Response
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>Butt</th>
<th>Slant</th>
<th>H₂S</th>
</tr>
</thead>
<tbody>
<tr>
<td>Citrobacter freundii ATCC 8090</td>
<td>50-100</td>
<td>luxuriant</td>
<td>acidic reaction, yellowing of the medium</td>
<td>alkaline reaction, purple reaction, or no colour change</td>
<td>positive</td>
</tr>
<tr>
<td>Escherichia coli ATCC 25922 (00013*)</td>
<td>50-100</td>
<td>luxuriant</td>
<td>alkaline reaction, purple reaction, purple reaction</td>
<td>or no colour change</td>
<td>negative</td>
</tr>
<tr>
<td>Proteus mirabilis ATCC 25933</td>
<td>50-100</td>
<td>luxuriant</td>
<td>acidic reaction, yellowing of the medium</td>
<td>deep red, lysine deamination</td>
<td>positive</td>
</tr>
<tr>
<td>Salmonella Arizonae ATCC 13314</td>
<td>50-100</td>
<td>luxuriant</td>
<td>alkaline reaction, purple reaction, purple reaction</td>
<td>or no colour change</td>
<td>positive</td>
</tr>
<tr>
<td>Salmonella Enteritidis ATCC 13076 (00030*)</td>
<td>50-100</td>
<td>luxuriant</td>
<td>alkaline reaction, purple reaction, purple reaction</td>
<td>or no colour change</td>
<td>positive</td>
</tr>
<tr>
<td>Salmonella Typhimurium ATCC 14028 (00031*)</td>
<td>50-100</td>
<td>luxuriant</td>
<td>alkaline reaction, purple reaction, purple reaction</td>
<td>or no colour change</td>
<td>positive</td>
</tr>
<tr>
<td>Shigella flexneri ATCC 12022 (00126*)</td>
<td>50-100</td>
<td>luxuriant</td>
<td>acidic reaction, yellowing of the medium</td>
<td>alkaline reaction, purple reaction</td>
<td>negative</td>
</tr>
</tbody>
</table>

Key : *Corresponding WDCM numbers.

Storage and Shelf Life
On receipt store between 2-8°C Use before expiry date on the label. Product performance is best if used within stated expiry period.

Please refer disclaimer Overleaf.
Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (4,6).

Reference

8. Thatcher F.S. and Clark D.S., 1968, University of Toronto Press, p. 10