Lowenstein Jensen Medium Base, Modified

Intended use
Lowenstein Jensen Medium (L. J. Medium) is used for the isolation of *Mycobacterium* species from mixed flora.

Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / 600 ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>L-Asparagine</td>
<td>3.600</td>
</tr>
<tr>
<td>Monopotassium phosphate</td>
<td>2.500</td>
</tr>
<tr>
<td>Magnesium sulphate</td>
<td>0.240</td>
</tr>
<tr>
<td>Sodium citrate</td>
<td>0.600</td>
</tr>
<tr>
<td>Potato Flour</td>
<td>30.000</td>
</tr>
<tr>
<td>Malachite green</td>
<td>0.400</td>
</tr>
</tbody>
</table>

**Formula adjusted, standardized to suit performance parameters

Directions
Suspend 37.24 grams in 600 ml distilled water containing 12 ml glycerol (for bovine bacteria or other glycerophobic organisms additions of glycerol is not desirable). Heat if necessary to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Meanwhile prepare 1000 ml of whole egg emulsion collected aseptically. Aseptically add and mix egg emulsion base and LCN Supplement (FD338) gently to obtain uniform mixture. Distribute in sterile screw capped tubes. Arrange tubes in a slanted position. Coagulate and inspissate the medium in an inspissator water bath or autoclave at 85°C for 45 minutes.

Principle And Interpretation
Solid media used for isolation and cultivation of Mycobacteria are either egg-based or agar-based. Egg-based media contain whole eggs or egg yolk, potato flour, salts and glycerol and are solidified by inspissation. Of the egg-based media, Lowenstein Jensen Medium is most commonly used (1). L.J. Medium was originally formulated by Lowenstein, containing congo red and malachite green dyes (2). Jensen (3) modified Lowensteins medium by altering the citrate and phosphate contents, eliminating the congo red dye and by increasing the malachite green concentration. This medium supports the growth of a wide variety of Mycobacteria and can also be used for niacin testing (6).

Lincomycin, Cycloheximide and Nalidixic acid along with malachite green prevents growth of the majority of contaminants surviving decontamination of the specimen while encouraging earliest possible growth of Mycobacteria. Do not add glycerol to the medium if bovine or other glycerophobic strains are to be cultured (7). Malachite green serves as an inhibitor and also as pH indicator. Formation of blue zone indicates a decrease in pH by gram-positive contaminants and yellow zones of dye destruction by gram-negative bacilli. Proteolytic contaminants cause localized or complete digestion of medium. LCN Supplement contains cycloheximide, lincomycin and nalidixic acid. Cycloheximide suppresses the growth of saprophytic organisms, Lincomycin inhibits gram positive organisms while nalidixic acid inhibits gram negative organisms in clinical samples. Refer appropriate references for standard test procedures of decontamination and isolation (1, 4,8-9).

Type of specimen
Clinical samples - Sputum sample

Specimen Collection and Handling
For clinical samples follow appropriate techniques for handling specimens as per established guidelines (1, 4,5,8-9)

Warning and Precautions:
In Vitro diagnostic Use only. 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
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Gelling
Free flowing powder

Colour and Clarity of prepared medium
The mixture of sterile basal medium and whole egg emulsion, when inspissated, coagulates to yield pale bluish green coloured, opaque smooth slants

Cultural Response
M2032: Cultural characteristics observed in presence of 5-10% Carbon dioxide, with added egg emulsion base, after an incubation at 35-37°C for 2-4 weeks.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Growth</th>
<th>Growth with Gruft Supplement (FD053)</th>
<th>Colony Characteristic</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mycobacterium avium ATCC25291</td>
<td>luxuriant</td>
<td>good-luxuriant</td>
<td>smooth, non-pigmented colonies</td>
</tr>
<tr>
<td>Mycobacterium gordonae ATCC 14470</td>
<td>luxuriant</td>
<td>good-luxuriant</td>
<td>smooth, yellow, orange colonies</td>
</tr>
<tr>
<td>Mycobacterium kansasii ATCC 12478</td>
<td>luxuriant</td>
<td>good-luxuriant</td>
<td>photochromogenic, smooth to rough</td>
</tr>
<tr>
<td>Mycobacterium smegmatis ATCC 14468</td>
<td>luxuriant</td>
<td>good-luxuriant</td>
<td>wrinkled, creamy white colonies</td>
</tr>
<tr>
<td>M. tuberculosis H37RV ATCC 25618</td>
<td>luxuriant</td>
<td>good-luxuriant</td>
<td>granular, rough, warty, dry friable colonies</td>
</tr>
</tbody>
</table>

Storage and Shelf Life:
Store between 10-30°C in a tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Product performance is best if used within stated expiry period.

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 clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (4,5).

References

Please refer disclaimer Overleaf.

IVD

In vitro diagnostic medical device

CE Marking

Storage temperature

10°C

30°C

Do not use if package is damaged

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