LV Agar Base, Modified (Liver-Veal-Agar Base, Modified)  

**Intended Use:**
Recommended for isolation of *Clostridium botulinum* in accordance with FDA BAM, 1998.

**Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>HL infusion from #</td>
<td>50.000</td>
</tr>
<tr>
<td>HMV infusion from $</td>
<td>500.000</td>
</tr>
<tr>
<td>Proteose peptone</td>
<td>20.000</td>
</tr>
<tr>
<td>Peptone special</td>
<td>1.300</td>
</tr>
<tr>
<td>Tryptone</td>
<td>1.300</td>
</tr>
<tr>
<td>Dextrose (Glucose)</td>
<td>5.000</td>
</tr>
<tr>
<td>Starch soluble</td>
<td>10.000</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>5.000</td>
</tr>
<tr>
<td>M-Protein, purified</td>
<td>2.000</td>
</tr>
<tr>
<td>Sodium nitrate</td>
<td>2.000</td>
</tr>
<tr>
<td>Gelatin</td>
<td>20.000</td>
</tr>
<tr>
<td>Agar</td>
<td>15.000</td>
</tr>
<tr>
<td>Final pH (at 25°C)</td>
<td>7.3 ± 0.2</td>
</tr>
</tbody>
</table>

**Directions**
Suspend 97 grams in 1000 ml warm purified / distilled water. Heat to boiling to dissolve the medium completely, and sterilize at 15 lbs (121°C) for 15 min. Cool to 45-50 °C. Aseptically add 80 ml Egg yolk emulsion, 50% (FD045F). Mix well and pour into sterile Petri dishes.

**Principle And Interpretation**
Anaerobic bacteria live in an oxygen-free environment. Some of the anaerobic bacteria die in presence of oxygen while others fail to grow and multiply (1). Liver Veal Agar Base, Modified (M1872) is a modification of the medium formulated by Spray, 1936 (8). It is recommended by the FDA Bacteriological Analytical Manual (BAM) (4) for the growth of anaerobic organisms especially *Clostridium botulinum*. This may also be used in supplementation with 50% egg yolk (FD045F) (3,4).

*Clostridium botulinum* is an anaerobic, rod-shaped spore forming bacterium that produces a protein with characteristic neurotoxicity. Under certain conditions, these organisms may grow in foods producing highly dangerous botulinum toxin(s). Botulinum toxin has been classified into botulinum A, botulinum B up to G. Among this all except F and G are known to cause animal botulism. Different strains are classified through antigenic characterization using appropriate antitoxins. They are also differentiated into general groups on the basis of cultural, biochemical, and physiological characteristics.

Both the infusions: HL infusion from and HMV infusion from, other peptones and gelatin serve as sources of carbon, nitrogen, amino acids and various vitamins. Dextrose serves as the energy source. Starch enhances growth of anaerobic bacteria. Sodium chloride maintains the osmotic equilibrium of the medium. Agar acts as the solidifying agent.

According to the FDA BAM protocol, suspected samples after preliminary examination is proceeded for enrichment of the organism. 1-2 g or 1-2 ml of samples after removing the dissolved oxygen content are inoculated into Cooked meat medium (M149) and Tryptone Peptone Glucose Yeast Extract Broth Base w/o Trypsin (M969) and incubated at 35°C and 25°C respectively. Upon 5 days of incubation, growth is checked by turbidity, gas production, and digestion of meat particles and different staining procedures of the growth. For isolation of pure cultures, alcohol pretreated samples are inoculated into Liver-Veal-Agar Base, Modified (M1872) and/or Anaerobic Egg Agar Base (M902F). The luster zones of different types vary often including variation in yellow precipitates. Serological, biochemical and invivo assays are performed to confirm the serotypes.
Type of specimen
Food and dairy samples.

Specimen Collection and Handling:
For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (1,7,9). 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.

Limitations:
1. Further biochemical and serological tests must be carried out for further identification.

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
Light yellow to brownish yellow homogeneous free flowing powder
Gelling
Firm, comparable with 1.5% Agar gel
Colour and Clarity of prepared medium
Amber coloured clear to slightly opalescent gel forms in Petri plates, may have slight precipitate.
Reaction
Reaction of 9.7% w/v aqueous solution at 25°C. pH : 7.3±0.2
pH
7.10-7.50
Cultural Response
Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours with addition of Egg yolk emulsion(under the atmospheric requirement of organism).

Organism Growth
Clostridium botulinum luxuriant
ATCC 25763
Clostridium tetani ATCC luxuriant
10709
Neisseria meningitidis ATCC luxuriant
13090
Streptococcus pneumoniae luxuriant
ATCC 6303

Storage and Shelf Life
Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order 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. Use before expiry date on the label.
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 sample must be decontaminated and disposed of in accordance with current laboratory techniques (5,6).
Reference


Disclaimer :

User must ensure suitability of the product(s) in their application prior to use. Products conform solely to the information contained in this and other related HiMedia™ publications. The information contained in this publication is based on our research and development work and is to the best of our knowledge true and accurate. HiMedia™ Laboratories Pvt Ltd reserves the right to make changes to specifications and information related to the products at any time. Products are not intended for human or animal or therapeutic use but for laboratory,diagnostic, research or further manufacturing use only, unless otherwise specified. Statements contained herein should not be considered as a warranty of any kind, expressed or implied, and no liability is accepted for infringement of any patents.