**Clostridium perfringens Agar Base**

**M2070**

**Intended Use**
Recommended for isolation and identification of *Clostridium perfringens* from unheated food material.

**Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>HMH extract</td>
<td>5.000</td>
</tr>
<tr>
<td>Proteose peptone</td>
<td>10.000</td>
</tr>
<tr>
<td>Peptone</td>
<td>10.000</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>5.000</td>
</tr>
<tr>
<td>Lactose</td>
<td>10.000</td>
</tr>
<tr>
<td>Phenol red</td>
<td>0.050</td>
</tr>
<tr>
<td>Agar</td>
<td>20.000</td>
</tr>
<tr>
<td>Final pH (at 25°C)</td>
<td>7.2±0.2</td>
</tr>
</tbody>
</table>

**Directions**
Suspend 60.05 grams in 900 ml distilled water. Heat to boiling to dissolve the medium completely. DO NOT AUTOCLAVE. Cool to 45-50°C and aseptically add 100 ml sterile Egg Yolk Tellurite Emulsion (FD046). Mix well and pour into sterile Petri plates.

**Principle And Interpretation**

*Clostridium perfringens*, ranked behind *Salmonella* species and *Staphylococcus aureus*, has been the third most common etiological agent of food-borne disease (1). *Clostridium* species are spore forming, gram-positive rods occurring naturally in soil (2). *C.perfringens* food poisoning results from eating contaminated food. The major virulence factor of *C.perfringens* is the CPE enterotoxin, which is secreted upon invasion of the host gut, and contributes to food poisoning and other gastrointestinal illnesses (2). *C.perfringens* cells may lose viability if the suspected food samples are refrigerated, thereby making it difficult to incriminate the organisms in food poisoning outbreaks (3). *C.perfringens* Agar Base is recommended for detecting *C.perfringens* from raw foods. If desired, Kanamycin can be added to the medium which restricts the growth of other bacteria. HMH extract, proteose peptone and peptone provides nitrogenous, carbonaceous nutrients, amino acids and other complex nutrients. Lecithinase of *C.perfringens* degrades lecithin of egg yolk, forming an insoluble opaque precipitate (4). Addition of tellurite and Kanamycin aids for selective isolation of *C.perfringens*.

**Type of specimen**
Food samples: meat and meat products

**Specimen Collection and Handling:**
For food samples, follow appropriate techniques for sample collection and processing as per guidelines (4). 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. Due to variable nutritional requirements, some strains show poor growth on this medium.
2. Further biochemical test must be carried out for confirmation.

Please refer disclaimer Overleaf.
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 light pink homogeneous free flowing powder

Gelling
Firm, comparable with 2.0% Agar gel.

Colour and Clarity of prepared medium
Basal medium - Red coloured, clear to very slightly opalescent gel. After addition of Egg Yolk Emulsion - Light red coloured, opaque gel forms in Petri plates

Reaction
Reaction of 6.0% w/v aqueous solution at 25°C. pH : 7.2±0.2

pH
7.00-7.40

Cultural Response
M2070: Cultural characteristics observed with added Egg Yolk Tellurite Emulsion (FD046) when incubated anaerobically, at 35-37°C for 24 - 48 hours.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Inoculum (CFU)</th>
<th>Growth</th>
<th>Recovery</th>
<th>Colour of colony</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clostridium perfringens ATCC 13124 (00007*)</td>
<td>50-100</td>
<td>good-luxuriant</td>
<td>&gt;=50%</td>
<td>black</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>opaque zone</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>around the colony</td>
</tr>
<tr>
<td>Clostridium perfringens ATCC 12916 (00080*)</td>
<td>50-100</td>
<td>good-luxuriant</td>
<td>&gt;=50%</td>
<td>black</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>opaque zone</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>around the colony</td>
</tr>
</tbody>
</table>

Key:- (*) Corresponding WDCM numbers

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. 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.