Yeast Mould Chloramphenicol Agar, Modified

Intended Use
Recommended for enumeration of yeasts and moulds from food using membrane filter technique. Also recommended when ISO-Grid and Neo-Grid protocols of filtration are followed.

Composition**

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Soya peptone</td>
<td>20.000</td>
</tr>
<tr>
<td>Tryptone</td>
<td>20.000</td>
</tr>
<tr>
<td>Dextrose (Glucose)</td>
<td>5.000</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>5.000</td>
</tr>
<tr>
<td>Dipotassium hydrogen phosphate</td>
<td>2.400</td>
</tr>
<tr>
<td>Trypan Blue</td>
<td>0.030</td>
</tr>
<tr>
<td>Chloramphenicol</td>
<td>0.100</td>
</tr>
<tr>
<td>Agar</td>
<td>15.000</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 67.53 grams in 995 ml purified / distilled water. Heat to boiling to dissolve the medium completely by agitating intermittently. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C and aseptically add the rehydrated contents of 1 vial of Chlortetracycline selective supplement (FD120). Mix well and pour into sterile Petri plates.

Principle And Interpretation
Yeast Mould Chloramphenicol Agar, Modified is recommended for enumeration of yeasts and moulds from foods using membrane filter technique. ISO-Grid and Neo-Grid protocols of filtration can be adopted for the same (1,2). To quantify the number of yeasts and moulds, a known amount of sample homogenate of food is prepared. Further 1 ml of homogenate is passed through the pre filter and ISO GRID membrane filter. The membrane filter is then placed on surface of Yeast Mould Chloramphenicol Agar, Modified plate. The surface of plate should be dry to avoid merging of developed colonies. Plates should be incubated in inverted position. Results may be interpreted after 48-52 hours on incubation at 20-25°C. It may be incubated further upto 72 hours for slow growing yeasts. The membrane filter is examined for developed colonies. Yeasts develop as blue coloured colonies while mols as blue-grey in colour.

This medium contains soya peptone and tryptone supplying necessary nitrogenous, carbonaceous substances, long chain amino acids and vitamin sources. Dextrose (Glucose) serves as fermentable carbohydrate. Sodium chloride maintains osmotic balance. Dipotassium hydrogen phosphate is a buffering agent. Trypan blue imparts blue colour to colonies of yeasts and moulds (5). Chlortetracycline and chloramphenicol selectively inhibits bacterial growth.

Type of specimen
Food samples

Specimen Collection and Handling
For food samples, follow appropriate techniques for sample collection and processing as per guidelines (6). 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. It may be incubated further upto 72 hours for slow growing yeasts.

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  
Cream to tan homogeneous free flowing powder  
Gelling  
Firm, comparable with 1.5% Agar gel  
Colour and Clarity of prepared medium  
Light blue to dark blue grey coloured clear to slightly opalescent gel forms in Petri plates  
Reaction  
Reaction of 6.75% w/v aqueous solution at 25°C. pH: 7.0±0.2  
\[ \text{pH} = 6.80-7.20 \]

Cultural Response  
Cultural characteristics observed with added chlortetracycline Selective supplement (FD120) after an incubation at 20-25°C for 48-72 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>Aspergillus brasiliensis ATCC 16404</td>
<td>50-100</td>
<td>good-luxuriant</td>
<td>&gt;=50%</td>
<td>blue</td>
</tr>
<tr>
<td>Candida albicans ATCC 10231</td>
<td>50-100</td>
<td>good-luxuriant</td>
<td>&gt;=50%</td>
<td>blue</td>
</tr>
<tr>
<td>Escherichia coli ATCC 25922</td>
<td>&gt;=10⁴</td>
<td>inhibited</td>
<td>0%</td>
<td>-</td>
</tr>
<tr>
<td>Bacillus subtilis subsp. spizizenii ATCC 6633</td>
<td>&gt;=10⁴</td>
<td>inhibited</td>
<td>0%</td>
<td>-</td>
</tr>
<tr>
<td>Saccharomyces cerevisiae ATCC 9763</td>
<td>50-100</td>
<td>good-luxuriant</td>
<td>&gt;=50%</td>
<td>blue</td>
</tr>
</tbody>
</table>

Key: (*) Corresponding WDCM numbers. (#) Formerly known as Aspergillus niger

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 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 clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (3,4).

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.