MUG Nutrient HiCynth™ Agar

Intended Use:
Recommended for detection of *Escherichia coli* in water and food samples by a fluorogenic procedures.

**Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>HiCynth™ Peptone No.1*</td>
<td>5.000</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>5.000</td>
</tr>
<tr>
<td>HiCynth™ Peptone No.5*</td>
<td>3.000</td>
</tr>
<tr>
<td>4-Methylumbelliferyl β-D-Glucuronide (MUG)</td>
<td>0.100</td>
</tr>
<tr>
<td>Agar</td>
<td>15.000</td>
</tr>
<tr>
<td>Final pH (at 25°C)</td>
<td>7.4±0.2</td>
</tr>
</tbody>
</table>

**Formula adjusted, standardized to suit performance parameters**

* Chemically defined peptones

**Directions**

Suspend 28.1 grams in 1000 ml purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

**Principle And Interpretation**

*Escherichia coli* is the member of faecal coliform group, presence of which in water indicates faecal contamination. These bacteria possess the enzyme β-glucuronidase and are capable of cleaving the fluorogenic substrate 4-Methylumbelliferyl β-D-Glucuronide (MUG) with the release of the corresponding fluorogen, 4-Methylumbelliferone (1). Therefore incorporation of MUG and subsequent fluoroscense is confirmatory for presence of *E. coli* with no further confirmation required (2). MUG Nutrient Agar is recommended for detection of *E. coli* in water and food samples by a fluorogenic method. Presumptive *E. coli* in the samples can be directly inoculated into the medium.

MUG Nutrient HiCynth™ Agar is prepared by replacing animal and vegetable peptones with chemically defined peptones to avoid BSE/TSE risks associated with animal peptones.

HiCynth™ Peptone No.1, HiCynth™ Peptone No.5 provide nitrogenous compounds and vitamin B complex. MUG is cleaved by the enzyme β-glucuronidase of *E. coli* to release 4-methylumbelliferone which produces visible green-blue fluorescence under long wave UV light (1). Some strains of *Salmonella* and *Shigella* species also produce glucuronidase (5). Refer appropriate references for standard procedures (1).

**Type of specimen**
Food samples; Water Sample

**Specimen Collection and Handling**

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (7).
For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (1). 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. This medium is general purpose medium and may not support the growth of fastidious organisms.
2. Further biochemical and serological tests must be carried out for further identification.
3. Some organism may show poor growth due to nutritional variation.

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 yellow homogeneous free flowing powder

Gelling
Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium
Light amber coloured clear to slightly opalescent gel forms in Petri plates

Reaction
Reaction of 2.81% w/v aqueous solution at 25°C. pH : 7.4±0.2

pH
7.20-7.60

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>Recovery</th>
<th>Fluorescence (under UV light at 366 nm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Escherichia coli ATCC 25922</td>
<td>50-100</td>
<td>good-luxuriant &gt;=70%</td>
<td>positive</td>
<td></td>
</tr>
<tr>
<td>Pseudomonas aeruginosa ATCC 27853</td>
<td>50-100</td>
<td>good-luxuriant &gt;=70%</td>
<td>negative</td>
<td></td>
</tr>
<tr>
<td>Staphylococcus aureus subsp. aureus ATCC 25923</td>
<td>50-100</td>
<td>good-luxuriant &gt;=70%</td>
<td>negative</td>
<td></td>
</tr>
<tr>
<td>Streptococcus pyogenes ATCC 19615</td>
<td>50-100</td>
<td>good-luxuriant &gt;=70%</td>
<td>Negative</td>
<td></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 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. 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 (3,4).

Reference

Please refer disclaimer Overleaf.