Pseudomonas Agar (For Fluorescein)

**Intended Use:**
Recommended for detection of fluorescein production by *Pseudomonas* species.

**Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tryptone</td>
<td>10.000</td>
</tr>
<tr>
<td>Proteose peptone</td>
<td>10.000</td>
</tr>
<tr>
<td>Dipotassium hydrogen phosphate</td>
<td>1.500</td>
</tr>
<tr>
<td>Magnesium sulphate</td>
<td>1.500</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 38 grams in 1000 ml purified / distilled water containing 10 ml glycerol. 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**

*Pseudomonas* Agar (For Fluorescein) is based on the formula described by King et al (3) and as modified in the U.S. Pharmacopeia (5) for the detection of fluorescein production a water soluble, chloroform insoluble fluorescent pigment by *Pseudomonas* species (4). The medium enhances the elaboration of fluorescein by *Pseudomonas* and inhibits the pyocyanin formation. The fluorescein pigment diffuses from the colonies of *Pseudomonas* into the agar and shows yellow fluorescent colouration. Some *Pseudomonas* strains produce small amounts of pyocyanin resulting in a yellow-green colouration.

Tryptone and proteose peptone provide the essential nitrogenous nutrients, carbon, sulphur and trace elements for the growth of *Pseudomonas*. Dipotassium hydrogen phosphate buffers the medium while magnesium sulphate provides necessary cations for the activation of fluorescein production. Salt concentration exceeding 2% affects pigment production. UV illumination may be bactericidal, so make sure that there is good growth before placing culture under UV light (4). A pyocyanin-producing *Pseudomonas* strain will usually also produce fluorescein. It must, therefore, be differentiated from other simple fluorescent *Pseudomonads* by other means. Temperature can be a determining factor as most other fluorescent strains will not grow at 35°C. Rather, they grow at 25-30°C (4).

**Type of specimen**

Pharmaceutical samples

**Specimen Collection and Handling:**

For pharmaceutical samples follow appropriate techniques for handling specimens as per established 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. This medium is general purpose medium and may not support the growth of fastidious organisms.

**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.

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Please refer disclaimer Overleaf.
Quality Control

Appearance
Cream to yellow homogeneous free flowing powder

Gelling
Firm, comparable with 1.5% Agar gel

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

Reaction
Reaction of 3.8% w/v aqueous solution (containing 1% v/v glycerol) at 25°C. pH : 7.0±0.2

pH
6.80-7.20

Cultural Response
Cultural characteristics observed with added 1% glycerol after an incubation at 35-37°C for 18-24 hours.

Organism | Inoculum (CFU) | Growth | Recovery | Colour of colony |
----------|----------------|--------|----------|-----------------|
_Pseudomonas aeruginosa_ ATCC 17934 | 50-100 | luxuriant | >=70% | greenish yellow |
_Pseudomonas aeruginosa_ ATCC 27853 (00025*) | 50-100 | luxuriant | >=70% | greenish yellow |
_Pseudomonas aeruginosa_ ATCC 9027 (00026*) | 50-100 | luxuriant | >=70% | greenish yellow |

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. 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 (1,2).

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.