Pseudomonas Agar, Modified (For Fluorescein)

M120F

For detection of fluorescein production by *Pseudomonas* species in accordance with FDA BAM, 1998.

Composition**

Ingredients	Gms / Litre
Tryptone	10.000
Proteose peptone	20.000
Dipotassium phosphate	1.500
Magnesum sulphate	0.730
Agar	15.000
Final pH (at 25°C)	7.0

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 47.23 grams in 1000 ml 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.

Principle And Interpretation

Pseudomonas Agar (for Fluorescein) is based on the formula described by FDA BAM (1) for the detection of fluorescein production , a water soluble, chloroform insoluble fluorescent pigment by *Pseudomonas* species (2). 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 phosphate buffers the medium while magnesium sulphate provides necessary cations for the activation of fluorescein production. Salt concentration exceeding 2% affects pigment production. Fluoroscein production is better visualized under UV light. Since it may be bactericidal, precaution should be taken that there is good growth in the broth before visualising it under UV light (2). A pyocyanin-producing *Pseudomonas* strain will usually also produce fluorescein. In order to differentiate the two species, sensitivity to temperature can be taken as a factor. Most of the simple fluorescent strains will not grow at 35°C. Rather, they grow at 25-30°C (2).

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 4.72% w/v aqueous solution (containing 1% v/v glycerol) at 25°C. pH : 7.0

pH
7.00

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

Cultural Response

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony

Cultural Response

Pseudomonas aeruginosa ATCC 17934	50-100	luxuriant	>=70%	greenish yellow
Pseudomonas aeruginosa ATCC 27853	50-100	luxuriant	>=70%	greenish yellow
Pseudomonas aeruginosa ATCC 9027	50-100	luxuriant	>=70%	greenish yellow

Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label.

Reference

1.FDA, U.S. 1998. Bacteriological Analytical Manual. 8 ed. Gaithersburg, MD: AOAC International.
2.MacFaddin, J. F. 1985. Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria vol. 1. Baltimore: Williams and Wilkins.

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