Technical Information

Medium 21. Pseudomonas Agar Medium for Detection of Pyocyanin

Product Code: DM 1119M

Application: Pseudomonas Agar for detection of Pyocyanin is recommended for the detection of pyocyanin production by *Pseudomonas* species in accordance with Indian Pharmacopoeia, 2007.

Composition**

Ingredients	Gms / Litre
Pancreatic digest of gelatin	20.000
Anhydrous potassium sulphate	10.000
Anhydrous magnesium chloride	1.400
Agar	15.000
pH after sterilization(at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

Pseudomonas Agar formulated by King et al [1] and recommended in Indian Pharmacopoeia [2] is used for detecting pyocyanin, a water soluble pigment by *Pseudomonas* species isolated from pharmaceutical preparation and clinical specimens such as stools, wounds, and urine [3]. *Pseudomonas* species are commonly causative agent of nosocomial, skin and burn infections. Pseudomonas strains are reported to produce phenazine pigments like Pyocyanin- blue green redox-active secondary metabolite pigment, pyorubin-rust brown pigment, -oxyphenzine- a breakdown product of Pyocyanin, pyoverdin-a water soluble yellow green pigments also known as fluorescein. Pyocyanin is readily recovered in large quantities in sputum from patients with cystic fibrosis, an infection caused by *Pseudomonas* [4, 5]. This medium enhances the formation of Pyocyanin and/or pyorubin and reduces that of fluorescein Pancreatic digest of gelatin provides essential nutrients for growth of *Pseudomonas,* while glycerol provides carbon and energy to the cell. The pyocyanin pigment diffuses from the colonies of *Pseudomonas* into the agar and shows blue colouration. Potassium sulphate and magnesium chloride enhances the pyocyanin production and suppresses the fluorescein production. Low content of phosphorous in the medium also aids in inhibiting the production of fluorescein. Some *Pseudomonas* strains produce small amounts of fluorescein resulting in a blue-green colouration.

Strains of *Pseudomonas aeruginosa* that may fail to produce Pyocyanin are not detected in this medium. Production of other pigments may mask the presence of Pyocyanin.

Methodology

Suspend 46.4 grams of powder media in 1000 ml purified / distilled water containing 10 ml glycerin. Shake well & heat to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Quality Control

Physical 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

pH range 7.00-7.40

Cultural Response/Characteristics

DM 1119M: Growth Promotion is carried out in accordance with the harmonized method of IP. Cultural response was observed after an incubation at 33-37°C for not less than 3 days. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar.

Organism	Inoculum (CFU)	Observed Lot value (CFU)	Recovery	Characteristi colonial morphology	Fluorescence in UV light	Growth
Pseudomonas aeruginosa ATCC 9027	50-100	35 -100	>=70 %	Generally greenish	positive	positive
Pseudomonas aeruginosa ATCC 27853	50-100	35 -100	>=70 %	Generally greenish	positive	positive

Storage and Shelf Lifez

Dried Media: Store below 30°C in tightly closed container and use before expiry date as mentioned on the label.
Prepared Media: 2-8^0 in sealable plastic bags for 2-5 days.

Further Reading

1. King, Ward and Raney, 1954, J.Lab. and Clin. Med., 44:301
2. Indian Pharmacopoeia, 2007, Govt. of India, Ministry of Health and Family Welfare, New Delhi, India.
3. MacFaddin J., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. I, Williams and Wilkins, Baltimore.
4. Daly J A, Boshard R, and Matsen J M, 1984, J Clin Microbiol. 19: 742
5. Lau GW, Hassett DJ, Ran H, Kong F., 2004. Trends Mol Med. 10:599

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