

Pseudomonas Isolation HiVeg™ Agar Base

MV406

Intended Use:

Recommended for selective isolation and identification of *Pseudomonas aeruginosa* from various specimens.

Composition**

Ingredients	g / L
HiVeg™ peptone	20.000
Magnesium chloride	1.400
Potassium sulphate	10.000
Triclosan (Irgasan)	0.025
Agar	13.600
Final pH (at 25°C)	7.0±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 45.03 grams in 1000 ml purified/distilled water containing 20 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 aeruginosa is an important human pathogen commonly found in nosocomial infections. It successfully combines adaptability to a variety of moist environments with a collection of potent virulence factors (1). *Pseudomonas* infections usually occur at any site where moisture tends to accumulate e. g. tracheostomies, in-dwelling catheters, burns, the external ear and weeping cutaneous wounds (2). *Pseudomonas Isolation Agar Base*, used for the selective isolation and identification of *P. aeruginosa*, is a modification of Medium A, originally formulated by King, Ward and Raney (3). The medium contains pigment-enhancing components and the selective agents, triclosan (4) which selectively inhibits non-pseudomonads. The pigment-enhancers i.e. potassium sulphate and magnesium chloride enhance the blue or blue-green pigment production by *P. aeruginosa*, thus aiding in its identification. *Pseudomonas Isolation HiVeg™ Agar Base* is prepared by using vegetable peptones in place of animal based peptones which make the media free of BSE/TSE risks.

HiVeg™ peptone provides nitrogenous compounds and other essential growth nutrients. Glycerol is a source of energy and promotes pyocyanin i.e. pigment production which is characteristic of *Pseudomonas* (5,6). Potassium sulphate and magnesium chloride enhance pyocyanin production. Triclosan (7) selectively inhibits gram-positive and gram-negative bacteria but *Pseudomonas* species are resistant to it. Some pyocyanin producing strains may also produce small amounts of fluorescein, resulting in the production of a blue-green to green pigment. Presumptive *Pseudomonas* should be further confirmed by performing biochemical tests, as some strains of *Pseudomonas* do not produce pyocyanin (8).

Type of specimen

Food samples; Water samples

Specimen Collection and Handling:

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (9). For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards.(10) After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. The media should be handled by trained personnel only. 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. Presumptive *Pseudomonas* should be further confirmed by performing biochemical tests, as some strains of *Pseudomonas* do not produce pyocyanin (8).

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 1.36% Agar gel.

Colour and Clarity of prepared medium

Yellow coloured clear to slightly opalescent gel forms in Petri plates.

Reaction

Reaction of 4.5% w/v aqueous solution at 25°C. pH : 7.0±0.2

pH

6.80-7.20

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony
<i>Escherichia coli</i> ATCC 25922 (00013*)	≥10 ⁴	inhibited	0%	-
<i>Proteus mirabilis</i> ATCC 25933	≥10 ⁴	inhibited	0%	-
<i>Pseudomonas aeruginosa</i> ATCC 10145 (00024*)	50-100	luxuriant	≥50%	green
<i>Pseudomonas aeruginosa</i> ATCC 27853 (00025*)	50-100	luxuriant	≥50%	blue to blue-green

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 (11,12).

Reference

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