King B Medium (Pseudomonas F Agar) ISO

Cat. 1532

For the identification and confirmation of Pseudomonas spp based on fluorescein production.

Practical information

Aplications	Categories
Confirmation	Pseudomonas aeruginosa
Detection	Pseudomonas

Industry: Water / Clinical

Regulations: ISO 16266

CE IVD

Principles and uses

King B Medium (Pseudomonas F Agar) is prepared according to the formula described by King et al. for the detection and differentiation of Pseudomonas aeruginosa (among other Pseudomonas), based on pyocyanin production.

The medium is recommended by ISO 16266 for the confirmation of reddish brown colonies in the Pseudomonas CN Agar (Cat. 1153) that have been positive oxidase. This method is recommended for bottled water, or for other types of water that have little background interfering flora, such as pool waters and water intended for human consumption.

Pseudomonas aeruginosa is a free-living bacterium, present in soil and water. It has become more and more known as an emerging opportunistic pathogen of clinical importance. Various different epidemiological studies track its occurrence as a nosocomial pathogen and claim that antibiotic resistance is increasing in clinical isolates.

Peptone mixture provides nitrogen, vitamins, minerals and amino acids essential for growth. It also promotes the production of pyocyanin. Dipotassium phosphate is a phosphorus source, and magnesium sulfate provides cations to activate pyocyanin production. Glycerol is a carbon source. Bacteriological agar is the solidifying agent.

Formula in g/L

Bacteriological agar	15	Magnesium sulfate	1,5
Peptone mixture	20	Dipotassium hydrogen phosphate	1,5

Typical formula g/L * Adjusted and/or supplemented as required to meet performance criteria.

Preparation

Suspend 38 grams of the medium in one liter of distilled water. Add 10 ml of glycerol. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Dispense into appropriate containers and sterilize in autoclave at 121 ºC for 15 minutes.

Instructions for use

» For clinical diagnosis, the type of sample is any clinical sample, and especially those that have a possible contamination with normal flora.
- Inoculate on the surface making parallel striae with the handle or swab.
- Incubate in aerobic conditions at at 36±2 ºC for a maximum of 5 days.
- Reading and interpretation of the results.

» For other uses not covered by the CE marking:

For the confirmation of Pseudomonas aeruginosa according to ISO 16266:
- Subcultivate the reddish brown colonies obtained in the Pseudomonas Agar CN (Cat. 1153) and that have been positive oxidase.
- Incubate at 36±2 ºC for a maximum of 5 days.
- Examine daily growth under UV radiation.
- Note the presence of any fluorescence.
- Bacteria that produce pyocyanin (blue/green) or are positive oxidase, giving fluorescence and synthesizing ammonia from acetamide, are counted as Pseudomonas aeruginosa.

Quality control

Solubility	Appearance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25ºC)
w/o rests	Fine powder	Light beige	Amber, slightly opalescent	7,2 ± 0,2

Microbiological test

Incubation conditions: (36±2 ºC / 18-24 h)

Microorganisms	Specification	Characteristic reaction
Pseudomonas aeruginosa ATCC 10145	Good growth	Presence of fluorescence under UV radiation
Escherichia coli ATCC 25922	Good growth	No fluorescence
Pseudomonas aeruginosa ATCC 27853	Good growth	Presence of fluorescence under UV radiation
Escherichia coli ATCC 8739	Good growth	No fluorescence
Pseudomonas aeruginosa ATCC 9027	Good growth	Presence of fluorescence under UV radiation

Storage

Temp. Min.:2 ºC
Temp. Max.:25 ºC

Bibliography

King E.O. Ward M.K. Raney D.E.-J. Lab. and Clin Med, 1954. 44. 301-307
Bacteriological Analytical Manual, 8th edition. 1995. AOAC International, Gaithersburg, MD.
The United States Pharmacopoeia. 1995. The United States Pharmacopoeia, 23rd ed. United States Pharmacopoeial Convention, Rockville, MD.

ISO 16266. Water quality. Detection and enumeration of Pseudomonas aeruginosa. Method by membrane filtration