





# KING B MEDIUM (PSEUDOMONAS F AGAR) USP

**CAT N°: 1532** For the identification of *Pseudomonas spp* based on fluorescein production

## FORMULA IN g/I

Peptone Mixture	20.00	Magnesium Sulfate	1.50		
Dipotassium Phosphate	1.50	Bacteriological Agar	14.00		
Final pH 7.0 + 0.2 at 25°C					

#### **PREPARATION**

Suspend 37 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°C for 15 minutes. The prepared medium should be stored at 8-15°C. The color is amber, slightly opalescent.

The dehydrated medium should be homogeneous, free-flowing and light beige in color. If there are any physical changes, discard the medium.

#### **USES**

KING B MEDIUM (Pseudomonas F Agar Base) is prepared according to the formula described by King *et al.* for the detection and differentiation of *Pseudomonas aeruginosa* from other *Pseudomonas* based on fluorescein (pyoverdin) production and pyocyanin inhibition.

*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 aids in the production of pyoverdin. Dipotassium phosphate is a phosphorus source, and Magnesium sulfate provides cations to activate pyoverdin production. Glycerol is a carbon source. Bacteriological agar is the solidifying agent.

Inoculate and incubate at  $35 \pm 2^{\circ}$ C for 18 - 24 hours.

This medium promotes the production of fluorescein (pyoverdin), a green-yellow fluorescent pigment that oxidizes to yellow. It is water-soluble and, unlike pyocyanin (blue-green pigment), it is not soluble in chloroform. The pigment diffuses throughout the medium and the fluorescent yellow-green color is observed.

### MICROBIOLOGICAL TEST

The following results were obtained in the performance of the medium from type cultures, with glycerol added, after incubation at a temperature of  $35 \pm 2^{\circ}$ C and observed after 18 - 24 hours.

Microorganisms	Growth	Colony Color
Pseudomonas aeruginosa ATCC 9027	Good	Yellow-green
Pseudomonas aeruginosa ATCC 10145	Good	Yellow-green
Pseudomonas aeruginosa ATCC 27853	Good	Yellow-green
Pseudomonas aeruginosa ATCC 25619	Good	Yellow-green

## **BIBLIOGRAPHY**

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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, 23<sup>rd</sup> ed. United States Pharmacopoeial Convention, Rockville, MD.



### **STORAGE**

Once opened keep powdered medium closed to avoid hydration.





