CETRIMIDE AGAR BASE EUROPEAN PHARMACOPOEIA, USP

CAT Nº: 1102 For the selective isolation and identification of *Pseudomonas aeruginosa*

FORMULA IN g/l

Pancreatic Digest of Gelatin	20.00	Cetrimide	0.30
Potassium Sulfate	10.00	Bacteriological Agar	13.60
Magnesium Chloride	1.40		

Final pH 7.2 ± 0.2 at 25ºC

Pseudomonas aeruginosa
ATCC 27853

PREPARATION

Suspend 45.3 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 colour is white-opaque.

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

USES

CETRIMIDE AGAR BASE is recommended by the European Pharmacopoeia for the selective isolation and identification of *Pseudomonas aeruginosa*. This medium promotes the production of fluorescein (pyoverdin), a green-yellow fluorescent pigment that oxidizes to yellow. Fluorescein is not soluble in chloroform, unlike pyocyanin (blue-green pigment). The pigment diffuses throughout the medium and the fluorescent yellow-green colour is observed.

Strains of *Pseudomonas aeruginosa* are identified from specimens because, in addition to their colonial morphology and the characteristic grape-like odor of aminoacetophenone, they produce pyocyanin, a blue, water-soluble, nonfluorescent, phenazine pigment. *P. aeruginosa* is the only specie of *Pseudomonas* or Gram-negative rod known to excrete pyocyanin.

Gelatin pancreatic digest provides nitrogen, vitamins, minerals and amino acids essential for growth. Glycerol is the carbon source. Magnesium chloride and Potassium Sulfate enhance the production of pyocyanin and pyoverdin. Cetrimide is the selective agent as it inhibits the growth of the accompanying microbial flora.

The European Pharmacopoeia, USP method recommends to inoculate the plates at 30-35ºC for 18-72 hours and to incubate *E. coli* as a negative control at 30-35ºC.

The identification of *P. aeruginosa* is completed by performing the oxidase test. Add a few drops of a freshly prepared N,N-dimethyl-p phenylenediamine monohydrochloride solution to the growth on the nutrient agar slant. Oxidase positive cultures develop a pink colour which successively becomes maroon, dark red, and black in 10 to 30 minutes.

MICROBIOLOGICAL TEST

The following results were obtained in the performance of the medium, with 10 ml of glycerol, from type cultures after incubation at a temperature of 30-35ºC and observed after 18-72 hours.

Microorganisms	Growth	Colony Color	Inoculum (cfu)	Recovery Rate (%)
Escherichia coli ATCC 25922	Inhibited	--------	≥100	≤0.01
Escherichia coli ATCC 8739	Inhibited	--------	≥100	≤0.01
Pseudomonas aeruginosa ATCC 27853	Good	Yellow-green	≤100	≥30
Pseudomonas aeruginosa ATCC 9027	Good	Yellow-green	≤100	≥30
Staphylococcus aureus ATCC 25923	Inhibited	---------	≥100	≤0.01
Staphylococcus aureus ATCC 6538	Inhibited	---------	≥100	≤0.01

BIBLIOGRAPHY

King, Ward and Raney. J. Lab. and Clin. Med. 44:301. 1954. Brown and Lowbury. J. Clin. Path. 18:752. 1965.
Lowbury. J. Clin. Path. 4:66. 1951. Lowbury and Collins. J. Clin. Path. 8:47. 1955.
European Pharmacopoeia; 7.0

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STORAGE

Once opened keep powdered medium closed to avoid hydration.