

## ANTIBIOTIC MEDIUM N° 3 USP

CAT N°: 1534

Standard medium for use in antibiotic assays

### FORMULA IN g/l

Gelatin Peptone	5.00	Beef Extract	1.50
Dipotassium Phosphate	3.68	Monopotassium Phosphate	1.32
Sodium Chloride	3.50	Dextrose	1.00
Yeast Extract	1.50		

Final pH 7.0± 0.2 at 25°C

### PREPARATION

Suspend 17.5 grams of the medium in one liter of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Sterilize at 121°C for 15 minutes. Distribute into appropriate containers. The prepared medium should be stored at 8-15°C. The color is clear amber.

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

### USES

ANTIBIOTIC MEDIUM N° 3 is prepared according to the formula specified by the Food and Drug Administration (FDA) and the United States Pharmacopoeia (USP).

Gelatin peptone, Yeast extract and Beef extract provide nitrogen, vitamins, minerals and amino acids essential for growth. Potassium phosphates act as a buffer system. Dextrose is the fermentable carbohydrate providing carbon and energy. Bacteriological Agar is the solidifying agent.

The potency of an antibiotic can be demonstrated under appropriate conditions by its inhibitory effect on microorganisms. Reduction in antimicrobial activity may reveal changes not demonstrated by chemical methods.

Antibiotic Medium N° 3 can be used with the following microbiological methods for Antibiotic Assays:

1. Cylinder method in plates.
2. Serial dilution method.
3. Turbidimetric method.

In the cylinder method in plates, Antibiotic Medium N° 3 is used to resuspend the inoculum in the potency assay for penicillin, erythromycin, neomycin, chlortetracycline and chloramphenicol.

The serial dilution method is used for penicillin assay.

Lastly, this medium can also be used in the turbidimetric determination of the potency of bacitracin, streptomycin and terramycin. The turbidimetric method is based on the inhibition of growth of a microbial culture in a fluid medium containing a uniform solution of an antibiotic. Use of this method is appropriate only when test samples are clear.

Plates are prepared and incubated following the FDA and the USP guidelines. The use of standardized culture media and strict control of all test conditions are essential requirements in the microbiological assay of antibiotics in order to obtain satisfactory test results.

## MICROBIOLOGICAL TEST

The following results were obtained in the performance of the medium from type cultures after incubation at a temperature of  $35 \pm 2^\circ\text{C}$  and observed after 24 - 48 hours.

Microorganisms	Growth	Inhibition zones
<i>Micrococcus luteus</i> ATCC 9431	Good	Erythromycin
<i>Staphylococcus aureus</i> ATCC 6538	Good	Kanamycin, Neomycin
<i>Klebsiella pneumoniae</i> ATCC 10031	Good	Streptomycin

## BIBLIOGRAPHY

Grove and Randall. Assay Methods of Antibiotics, Medical Encyclopedia Inc. New York 1955. United States Pharmacopoeia Convention. 1955. The United States Pharmacopoeia, 23rd Ed. Biological Tests and Assays, p. 1690-1696. The United States Pharmacopoeia Convention, Rockville, Md



## STORAGE

Once opened keep powdered medium closed to avoid hydration.

