



Antibiotic Assay Medium A

M003B

Intended use

Antibiotic Assay Medium A is used for microbiological diffusion assay of several antibiotics as per British Pharmacopoeia.

Composition**

Ingredients	Gms / Litre
Peptone	6.000
Tryptone\$	4.000
Yeast extract	3.000
HM Peptone B#	1.500
Glucose monohydrate	1.000
Agar	15.000
pH after sterilization (at 25°C)	6.6±0.1

**Formula adjusted, standardized to suit performance parameters

\$- Equivalent to Pancreatic digest of casein

#- Equivalent to Beef extract

Directions

Suspend 30.45 grams (the equivalent weight of dehydrated media) in 1000 ml R-water/purified / distilled water. 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.

Adjust the pH of the medium, using freshly prepared buffer solution as recommended by the British Pharmacopoeia for the antibiotic assayed.

Advice :Recommended for the microbiological assay of Josamycin, Josamycin propionate, Bacitracin zinc

Principle And Interpretation

This medium is also used as inoculum and maintenance medium for different test organisms for antibiotic assays. Composition of this medium is in accordance with British Pharmacopoeia(1).

Essential nutrients, vitamins, mineral, trace elements and growth factors are supplied by peptone, tryptone, yeast and HM peptone B. Glucose monohydrate in the medium serves as the carbon source for stimulating the growth of the test microorganism. Agar provides excellent medium for antibiotic diffusion and gives well defined zones of inhibition. Freshly prepared plates should be preferably used for assaying antibiotics. Test organisms is inoculated in sterile seed agar pre-cooled to 40-45°C and spread evenly over the surface of solidified base agar. All conditions in the microbiological assay must be controlled carefully. One of the critical and important step for obtaining good results is use of appropriate standard culture media.

Type of specimen

Pharmaceutical preparations

Specimen Collection and Handling

For pharmaceutical samples follow appropriate techniques for handling specimens as per established guidelines (1).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

Read the label before opening the container. 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. Freshly prepared plates must be used or it may result in erroneous results.

Please refer disclaimer Overleaf.

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 1.5% Agar gel

Colour and Clarity of prepared medium

Yellow coloured slightly opalescent gel forms in Petri plates.

pH

6.50-6.70

Growth Promotion Test

As per British Pharmacopoeia

Cultural Response

Cultural characteristics observed after an incubation at specified temperature for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed	Incubation Temperature
<i>Bacillus subtilis subsp. spizizenii</i> ATCC 6633 (00003*)	50-100	good-luxuriant	≥70 %	Josamycin, Josamycin propionate	35-37°C
<i>Micrococcus luteus</i> ATCC 10240	50-100	good-luxuriant	≥70 %	Bacitracin zinc (Adjust the pH to 7.0 ± 0.1)	35-39°C
<i>Micrococcus luteus</i> ATCC 9341	50-100	luxuriant	≥70 %	Rifamycin sulphate	35-39°C

*- Corresponding WDCM numbers

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. 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. Use before expiry date on the label.

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 (2,3).

Reference

1. British Pharmacopoeia, 2020, The Stationery office British Pharmacopoeia
2. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
3. Jorgensen, J.H, Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

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Disclaimer :

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