Potato Dextrose Agar

MH096

Intended use

Recommended for the cultivation of yeasts and moulds from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP/IP.

Composition**

Ingredients	Gms / Litre
Infusion from potatoes	200.000
Dextrose (Glucose)	20.000
Agar	15.000
pH after sterilization (at 25°C)	5.6±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 39.0 grams in 1000 ml purified/distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates or tubes as desired. In specific work, when pH 3.5 is required, acidify the medium with sterile 10% tartaric acid. The amount of acid required for 100 ml of sterile, cooled medium is approximately 1 ml. Do not heat the medium after addition of the acid.

Principle And Interpretation

Yeast and moulds constitute a large and divergent group of microorganisms consisting of several thousands species. Yeast and moulds can cause various degrees of food decomposition. Invasion and growth may occur on virtually any type of food if environmental conditions are not limiting. Some foodborne yeasts and moulds are undesirable because of potential hazards to human and animal health (1).

Potato Dextrose Agar, prepared in accordance with the harmonized methodology of USP/EP/BP/JP/IP (2,3,4,5,6) is recommended for microbial limit tests in pharmaceutical testing. It is also used for stimulating sporulation, for maintaining stock cultures of certain dermatophytes and for differentiation of typical varieties of dermatophytes on the basis of pigment production (7).

Potato infusion and dextrose (glucose) promote luxuriant fungal growth. Adjusting the pH of the medium by tartaric acid to 3.5 inhibits the bacterial growth. Heating the medium after acidification should be avoided as it may hydrolyse the agar, which can render the agar unable to solidify.

Type of specimen

Pharmaceutical samples

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (2,3,4,5,6).

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 guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. For heavily contaminated samples, the media must be supplemented with inhibitory agents for inhibiting bacterial growth with lower pH.
2. Some pathogenic fungi may produce infective spores which are easily dispersed in air, so examination should be carried out in safety cabinet.
3. Further biochemical tests should be carried out for confirmation.

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 clear to slightly opalescent gel forms in Petri plates
Reaction
pH of 3.9% w/v aqueous solution at 25°C (after sterilization).
pH
5.40-5.80
Growth Promotion Test
Growth Promotion was carried out in accordance with the harmonized method of USP/EP/BP/JP/IP, and growth was observed at 20-25°C for specified time. Recovery rate is considered as 100% for fungus growth on Sabouraud Dextrose Agar
Growth Promoting Properties
Growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating <= 100 cfu
Cultural Response
Cultural characteristics observed after incubation at 20-25 °C for 2-5 days. Recovery rate is considered as 100% for fungus growth on Sabouraud Dextrose Agar.

Organism	Inoculum (CFU)	Growth	Observed Lot value (CFU)	Recovery	Incubation temperature	Incubation period
Test strain preparation						
# *Aspergillus brasiliensis* ATCC 16404 (00053*)	50 -100	luxuriant	25 -100	>=50 %	20 -25 °C	5 -7 Day
Additional Microbiological Testing						
Aspergillus fumigatus ATCC 9197	50 -100	luxuriant	25 -100	>=50 %	20 -25 °C	5 -7 Day
Candida albicans ATCC 10231 (00054*)	50 -100	luxuriant	35 -100	>=70 %	20 -25 °C	2 -3 Day
Saccharomyces cerevisiae ATCC 9763 (00058*)	50 -100	luxuriant	35 -100	>=70 %	20 -25 °C	2 -5 Day
Rhodotorula mucilaginosa DSM 70403		luxuriant			20 -25 °C	3 -5 Day
Geotrichum candidum DSM 1240		good- luxuriant			25 -30 °C	3 -5 Day
Penicillium communae ATCC 10248		fair -good			25 -30 °C	3 -5 Day
Trichophyton ajelloi ATCC 28454		fair-good			25 -30 °C	3 -7 Day

Key : (#) - Formerly known as *Aspergillus niger,* (*) - corresponding WDCM numbers

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. 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. 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 (8,9).

Reference

1. Salfinger Y., and Tortorello M.L. , 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
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3. European Pharmacopoeia, 2022, 10 th volume, European Directorate for the quality of medicines & Healthcare.
4. The British Pharmacopoeia, 2022, Medicines and Healthcare products Regulatory Agency.
5. The Japanese Pharmacopoeia, 17th edition, 2016, The Ministry of Health, Labour and welfare.
6. Indian Pharmacopoeia, 2022, Indian Pharmacopoeia Commission, Ministry of Health and Family Welfare Government of India.
7. MacFaddin J., 1985, Media for the Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol.1, Williams and Wilkins, Baltimore
8. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
9. 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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