Plate Count Agar w/o Dextrose(Standard Methods Agar w/o Dextrose)

Intended Use:

M2047

Recommended for the determination of plate counts of microorganisms in water samples.

Composition**

Ingredients	Gms / Litre
Tryptone	5.000
Yeast extract	2.500
Agar	15.000
Final pH (at 25°C)	7.0±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 22.5 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. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

Principle And Interpretation

Plate Count Agar is formulated as described by Buchbinder et al (2) which is recommended by APHA (1) and FDA (3). Plate Count Agar w/o Dextrose is recommended for the detection of plate counts in water sample. Tryptone provides nitrogeneous and carbonaceous compounds, long chain amino acids and other essential growth nutrients. Yeast extract supplies Vitamin B complex. APHA recommends the use of pour plate technique. The samples are diluted and appropriate dilutions are added in Petri plates. Sterile molten agar is added to these plates and plates are rotated gently to ensure uniform mixing of the sample with agar. The poured plate count method is preferred to the surface inoculation method, since it gives higher results.

Method : Prepare serial dilution in duplicates of the sample to be tested and place one ml of the sample in Petri plate and then overlay or pour plate with sterile Plate Count Agar w/o Dextrose. Incubate one set of plate at 34-38°C for 40-48 hours and second set of plates at 20-24°C for 64-72 hours. (dilution of the sample in compliance with NF EN ISO 8199) (4)

Type of specimen

Water samples.

Specimen Collection and Handling

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (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. Further biochemical testing is required for identification of microorganism.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within expiry period when stored at recommended the 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
Light yellow coloured clear to slightly opalescent gel forms in Petri plates.
Reaction
Reaction of 2.35% w/v aqueous solution at 25°C. pH : 7.0±0.2

pH
6.80-7.20
Cultural Response
Cultural characteristics observed after an incubation at 34-38°C for 40- 48 hours and 20-24°C for 64-72 hours.

Organism	Inoculum (CFU)	Growth	Recovery
Bacillus subtilis subsp. spizizenii ATCC 6633 (00003*)	50-100	luxuriant	>=70%
Enterococcus faecalis ATCC 29212 (00087*)	50-100	luxuriant	>=70%
Escherichia coli ATCC 25922 (00013*)	50-100	luxuriant	>=70%
Escherichia coli ATCC 8739 (00012*)	50-100	luxuriant	>=70%
Staphylococcus aureus subsp. aureus ATCC 25923 (00034*)	50-100	luxuriant	>=70%
Candida albicans ATCC 10231 (00054*)	50-100	luxuriant	>=70%
# *Aspergillus brasiliensis* ATCC 16404 (00053*)	50-100	luxuriant	>=70%

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 clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (5,6).

Reference

1. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.
2. Buchbinder L., Baris Y., Aldd E., Reynolds E., Dilon E., Pessin V., Pincas L. and Strauss A., 1951, Publ. Hlth. Rep., 66:327.
3. FDA Bacteriological Analytical Manual, 2005, 18th Ed., AOAC, Washington, DC.
4. ISO 8199:2005 Water quality -- General guidance on the enumeration of micro-organisms by culture.
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
6. 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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