



Anaerobic Agar without Dextrose

M230

Intended Use:

Recommended for carbohydrate fermentation studies and for studies of hemolytic activity of *clostridia*, *streptococci* and other organisms.

Composition**

Ingredients	g / L
Tryptone	17.500
Sodium chloride	2.500
Sodium thioglycollate	2.000
Sodium formaldehyde sulfoxylate	1.000
Methylene blue	0.002
Agar	15.000
Final pH (at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 38.0 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. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

Principle And Interpretation

Anaerobic Agar without Dextrose is devoid of dextrose (1) and is used for carbohydrate fermentation studies and for studies of haemolytic activity of *Clostridia*, *Streptococci* and other organisms. Tryptone is the only source of nutrient present. It provides nitrogen and carbon source, long chain amino acids, vitamins and other essential nutrients. Other peptones, rich in carbohydrates like yeast extract or soya peptone are not used in this medium as this medium favours distinct haemolytic reactions. Addition of sodium thioglycollate and sodium formaldehyde sulfoxylate create conditions necessary for cultivation of anaerobic bacteria which is indicated by methylene blue dye present in the medium. Sodium chloride maintains osmotic equilibrium. For haemolytic tests anaerobic blood agar plates may be prepared in one of the following ways;

Sterile blood in about 0.7 ml amount and small inoculum may be mixed with 25-50 ml of cooled agar and mixture is poured into the Petri plate filling it up to 3/4. After solidification the lid is replaced with Brewer Anaerobic Petri plate cover.

An ordinary sterile Blood Agar plate (made from Blood Agar Base or Tryptone Soya Agar) may be streaked with a culture. Melted and cooled Anaerobic Agar without Dextrose is then poured over the Blood Agar to provide the proper depth. After solidification the lid is replaced with anaerobic Petri plate cover.

The anaerobic cover should not rest on the Petri plate bottom: its inner ridge should seal the agar, and the medium within the ridge should not touch the cover at any point. The medium should be cherry red in colour after addition of blood.

Type of specimen

Clinical sample- stool, abscess

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (2,3).

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

Warning and Precautions

In Vitro diagnostic use only. For professional use only. 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 clinical specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. The anaerobic cover should not rest on the Petri plate bottom: its inner ridge should seal the agar, and the medium within the ridge should not touch the cover at any point.
2. The medium should be cherry red in colour after addition of blood.

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

Light amber coloured clear to slightly opalescent gel forms in Petri plates that becomes greenish due to aeration on standing.

Reaction

Reaction of 3.8% w/v aqueous solution at 25°C. pH : 7.2±0.2

pH

7.00-7.40

Cultural Response

Cultural characteristics observed under anaerobic condition after an incubation at 35-37°C for 18-48 hours.

Organism	Inoculum (CFU)	Growth	Recovery
<i>Clostridium butyricum</i> ATCC 13732	50-100	good-luxuriant	≥50%
<i>Clostridium perfringens</i> ATCC 12919	50-100	good-luxuriant	≥50 %
<i>Clostridium sporogenes</i> ATCC 11437	50-100	good-luxuriant	≥50%

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 (2,3).

Reference

1. Vera J., 1942, J. Bact., 44:497.
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.

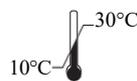
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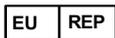
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