



Anaerobic Agar (Brewer)

M491

Composition**

Ingredients	Gms / Litre
Proteose peptone	10.000
Tryptone	5.000
Yeast extract	5.000
Dextrose (Glucose)	10.000
Sodium chloride	5.000
Sodium thioglycollate	2.000
Sodium formaldehyde sulfoxylate	1.000
Resazurin	0.002
Agar	15.000
Final pH (at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 53.0 grams in 1000 ml 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

Brewer (1) devised this medium for use with Brewer anaerobic cover to permit surface growth of anaerobes and microaerophiles on agar without the use of anaerobic jar. This medium is suitable for isolation of facultative and obligate anaerobes and for the study of colonial morphology as colonies can be readily seen on the light coloured agar and are easily accessible (2,3).

Dispense 50-60 ml medium per 95 x 20 mm plate. For best results, use porous tops for the plates during solidification to obtain a dry surface. Inoculation can be done by streaking or smearing. After inoculation of the medium, cover with Brewer Anaerobic Petri dish cover. The sealing ring inside the cover should make a perfect contact with the medium and must not be broken before the end of the incubation period. When standard plates are used, dispense 0.1 to 1.0 ml of inoculum into plates and mix 20-25 ml of sterile medium.

Proteose peptone, tryptone and yeast extract provides nitrogen, carbon, long chain amino acids, vitamins and other essential growth nutrients. Dextrose is a carbohydrate source. This medium contains sodium thioglycollate and sodium formaldehyde sulfoxylate that provide adequate anaerobiosis, which is indicated by resazurin present in the medium. Resazurin imparts pink colour to the medium in presence of oxygen.

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 red due to aeration on standing.

Reaction

Reaction of 5.3% 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 botulinum</i> ATCC 19397	50-100	luxuriant	>=50%
<i>Clostridium perfringens</i> ATCC 12924	50-100	luxuriant	>=50%
<i>Clostridium sporogenes</i> ATCC 11437	50-100	luxuriant	>=50%

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium 20-30 2-8°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 (4,5).

Reference

1. Brewer, 1942, Science, 95, 587.
2. Isenberg (Ed.), 1992, Clinical Microbiology Procedures Handbook, American Society for Microbiology, Washington, D.C.
3. Baron E. J., Peterson and Finegold S. M., Bailey & Scotts Diagnostic Microbiology, 9th Ed., 1994, Mosby-Year Book, Inc., St. Louis, Mo.

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