

Wilkins Chalgren Anaerobic Broth Base Intended Use:

M863

Used for cultivation and susceptibility testing of anaerobic bacteria.

Composition**		
Ingredients	g / L	
Tryptone	10.000	
Peptone	10.000	
Yeast extract	5.000	
Dextrose (Glucose)	1.000	
Sodium chloride	5.000	
L-Arginine	1.000	
Sodium pyruvate	1.000	
Hemin	0.005	
Menadione	0.0005	
Final pH (at 25°C)	7.1±0.2	

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 33.01 grams in 1000 ml distilled water. Heat if necessary to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C before adding antibiotics to be tested. Mix gently and dispense into sterile tubes.

For cultivation of anaerobes, aseptically add the rehydrated contents of 2 vials each of Anaero Supplement (FD001) or G.N. Anaero Supplement (FD002) as required into sterile medium before dispensing into sterile tubes.

Principle And Interpretation

Wilkins Chalgren Anaerobic Broth Base, formulated by Wilkins and Chalgren (1), is the preferred medium for susceptibility testing of anaerobes. This medium is also recommended for testing anaerobic bacteria (2,3,4). Wilkins Chalgren Anaerobic Broth Base is similar to the agar medium, except the agar (3). The broth medium is especially useful in the broth micro-dilution tests (5). Wilkins Chalgren Broth media need to be appropriately supplemented to support the growth of certain anaerobic bacteria.

Hemin and Menadione (Vitamin K3) enhances the growth of *Bacteroides* species and *Prevotella melaninogenica*, respectively and many other species of gram-negative anaerobic rods (3,6). The medium can also be supplemented with defibrinated or lysed blood for the growth of fastidious anaerobic bacteria (7).

Tryptone and Peptone serve as sources of essential nutrients including carbon and nitrogen. Yeast extract provides vitamins and other growth factors like purines and pyrimidines that are essential for the growth of *P. melaninogenica*. Arginine serves as an amino acid source while pyruvate serves as an energy source. The medium can be made selective for non-sporing anaerobic bacteria and gram-negative anaerobic bacteria by addition of Anaero Supplement (FD001) and G.N. Anaero Supplement (FD002) respectively.

Type of specimen

Clinical sample - stool, abscess

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (6,8). 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. Individual organisms differ in their growth requirement and may show variable growth patterns on the medium.

2.Each lot of the medium has been tested for the organisms specified on the COA. It is recommended to users to validate the medium for any specific microorganism other than mentioned in the COA based on the user's unique requirement. 3.Proper anaerobic conditions must be maintained for optimal recovery of organisms.

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

Colour and Clarity of prepared medium

Medium amber coloured clear solution in tubes.

Reaction

Reaction of 3.3% w/v aqueous solution at 25°C. pH : 7.1±0.2

pН

6.90-7.30

Cultural Response

Cultural characteristics observed with added Anaero Supplement (FD001) or G.N. Anaero Supplement (FD002) under anaerobic conditions, after an incubation at 35-37°C of 48 hours.

Organism	Inoculum (CFU)	Growth
<i>Bacteroides fragilis</i> ATCC 25285	50-100	luxuriant
Clostridium perfringens ATCC 12924	50-100	luxuriant
Prevotella melaninogenicus ATCC 15930	50-100	luxuriant
Escherichia coli ATCC 25922 (00013*)	>=10 ⁴	Inhibited

Key: (*) Corresponding WDCM numbers.

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder 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 (6,8).

Reference

1. Wilkins T. D. and Chalgren S., 1976, Antimicrob. Agents Chemother., 10:926

2. Clinical and Laboratory Standards Institute, 2006, Methods for Antimicrobial Susceptibility Testing of Anaerobic Bacteria, Approved standard M11-A3, CLSI, Villanova, Pa.

3. Collee J. G., Fraser A. G., Marmion B. P., Simmons A., (Eds.), Mackie and McCartney Practical Medical Microbiology, 1996, 14th Edition, Churchill Livingstone.

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- 5. Quinto G. and Sebald M., 1964, Am. J. Med. Technol., 30:381.
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In vitro diagnostic

medical device

IVD



-30°C Storage temperature

Do not use if package is damaged

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