TECHNICAL DATA SHEET

WILKINS-CHALGREN AGAR

CULTURE OF ANAEROBIC BACTERIA

1 INTENDED USE

Wilkins Chalgren Agar is used to determine the minimal inhibitory concentrations of antibiotics for anaerobic bacteria by the dilution method. It can also be used for the abundant growth and isolation of anaerobic bacteria.

2 HISTORY

Wilkins and Chalgren established the formula of the medium in 1976. They showed that it had the advantage of not requiring blood to obtain satisfactory growth of anaerobic bacteria of clinical importance. The modalities of utilization are described in the NCCLS Collaborative Study of the Proposed Reference Dilution Method of Antimicrobial Susceptibility Testing of Anaerobic Bacteria.

3 PRINCIPLES

The combination of casein and gelatin peptones, yeast extract and glucose makes the medium very nutritious.

Hemin stimulates the growth of fastidious bacteria.

Vitamin K1 favors the growth of *Bacteroides*.

Sodium chloride maintains osmotic equilibrium.

4 TYPICAL COMPOSITION

The composition can be adjusted in order to obtain optimal performance.

For 1 liter of media :

- Tryptone ..... 10,0 g
- Pancreatic digest of gelatin ..... 10,0 g
- Yeast extract ..... 5,0 g
- L-arginine ..... 1,0 g
- Glucose ..... 1,0 g
- Hemin ..... 5,0 mg
- Vitamin K1 ..... 0,5 mg
- Sodium pyruvate ..... 1,0 g
- Sodium chloride ..... 5,0 g
- Bacteriological agar ..... 15,0 g

pH of the ready-to-use media at 25 °C : 7,1 ± 0,2.

5 PREPARATION

- Dissolve 48,0g of dehydrated media (BK101) in 1 liter of distilled or demineralized water.
- Slowly bring to boiling, stirring with constant agitation until complete dissolution.
- Dispense in tubes or vials.
- Sterilize in an autoclave at 121°C for 15 minutes.
- Cool and maintain the media in a molten state at 44-47 °C.
- If needed, add the antibiotics to test.
- Pour into sterile Petri plates and let solidify on a cold, flat surface.

✓ **Reconstitution:**
48,0 g/L

✓ **Sterilization:**
15 min at 121 °C

NOTE

For the culture of certain fastidious microorganisms, the media can be supplemented with defibrinized blood, 5 mL per 100 mL of molten media held at 44-47°C.

6 INSTRUCTIONS FOR USE

- Dry the plates in an incubator, covers partially removed.
- Inoculate the samples to test.
- Incubate under anaerobic conditions at 37 °C for 24 to 48 hours.

✓ **Inoculation:**
On surface
✓ **Incubation:**
24 h to 48 h at 37 °C

7 RESULTS

Verify the growth of the microorganisms on the plates.

8 QUALITY CONTROL

Dehydrated media : white-cream powder, free-flowing and homogeneous.
Prepared media : amber agar.

Typical culture response after 48 hours of incubation under anaerobic conditions at 37 °C :

Microorganisms		Growth
Clostridium perfringens	WDCM 00007	Good, score 2
Clostridium sporogenes	WDCM 00008	Good, score 2

9 STORAGE / SHELF LIFE

Dehydrated media : 2-30 °C.
The expiration date is indicated on the label.

Prepared media in vials (*) : 180 days at 2-8 °C .
Prepared media in plates (*) : 30 days at 2-8 °C.
(*) Benchmark value determined under standard preparation conditions, following manufacturer's instructions.

10 PACKAGING

Dehydrated media :
500 g bottle BK101HA

11 BIBLIOGRAPHY

Wilkins, T.D., and Chalgren, S. 1976. Medium for use in antibiotic susceptibility testing of anaerobic bacteria. Antimicrob. Agents Chemother., 10 (6): 926.

N.C.C.L.S. 1982. Tentative Standard Reference Agar Dilution Procedure for Susceptibility Testing of Anaerobic Bacteria , 2, n°3.

Lennette, E.H., Balows, A., Hausler, Jr., W.J., and Shadomy, H.J. 1985. Manual of Clinical Microbiology. A.S.M. Washington. 4 th Ed., 988-990.

12 ADDITIONAL INFORMATION

The information provided on the labels take precedence over the formulations or instructions described in this document and are susceptible to modification at any time, without warning.

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