WILKINS-CHALGREN AGAR (7232)

Intended Use
Wilkins-Chalgren Agar is used for the cultivation of anaerobic microorganisms.

Product Summary and Explanation
Wilkins-Chalgren Agar was designed by Wilkins and Chalgren for use in determining minimal inhibitory concentration (MIC's) of antibiotics for anaerobic bacteria by the agar dilution procedure.[1] This medium was selected because it does not require the addition of blood to support satisfactory growth of most anaerobes. Anaerobic bacteria cause a variety of human infections including endocarditis, meningitis, wound infections following bowel surgery or trauma, and bacteremia.[2,3] The survival of anaerobic bacteria is dependent on their sensitivity to oxygen, nutritional requirements, appropriate collection, culture medium, and incubation time and temperature.[4]

Principles of the Procedure
Enzymatic Digest of Casein and Enzymatic Digest of Gelatin provide nitrogen, amino acids, and minerals in Wilkins-Chalgren Agar. Yeast Extract is added to provide vitamins and other growth factors, including purines and pyrimidines. Sodium Chloride maintains the osmotic balance of the medium. Dextrose is a carbon source. L-Arginine contributes to growth of *Eubacterium lentum*. Sodium Pyruvate provides an energy source for *Veillonella* spp. and degrades hydrogen peroxide. Hemin (X Factor) and Vitamin K are essential for growth of a number of species of anaerobes. Agar is the solidifying agent.

Formula / Liter
Enzymatic Digest of Casein	10 g
Enzymatic Digest of Gelatin	10 g
Yeast Extract	5 g
Sodium Chloride	5 g
Dextrose	1 g
L-Arginine	1 g
Sodium Pyruvate	1 g
Hemin	0.005 g
Vitamin K	0.0005 g
Agar	15 g

Final pH: 7.1 ± 0.2 at 25°C
Formula may be adjusted and/or supplemented as required to meet performance specifications.

Precautions
1. For Laboratory Use.
2. IRRITANT. Irritating to eyes, respiratory system, and skin.

Directions
1. Suspend 48 g of the medium in one liter of purified water.
2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
3. Autoclave at 121°C for 15 minutes.

Quality Control Specifications
Dehydrated Appearance: Powder is homogeneous, free flowing, and light beige.

Prepared Appearance: Prepared medium is yellow-beige and trace to slightly hazy.

Expected Cultural Response: Cultural response on Wilkins-Chalgren Agar incubated anaerobically at 35 ± 2°C and examined for growth after 48 - 72 hours.

Microorganism	Approx. Inoculum (CFU)	Expected Results
Bacteroides fragilis ATCC® 25285	10 - 300	Growth
Clostridium novyi ATCC® 7659	10 - 300	Growth
Clostridium perfringens ATCC® 13124	10 - 300	Growth

The organisms listed are the minimum that should be used for quality control testing.

Test Procedure

For a complete discussion of aerobic and anaerobic bacteria from clinical specimens, refer to appropriate procedures outlined in the references.[4-6] Refer to standard methods for the examination of bacteria in food.[7,8]

Results

Refer to appropriate references for results.

Storage

Store sealed bottle containing the dehydrated medium at 2 - 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping container tightly closed.

Expiration

Refer to expiration date stamped on container. The dehydrated medium should be discarded if not free flowing, or if appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitation of the Procedure

Due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.

Packaging

Wilkins-Chalgren Agar	**Code No.**	**7232A**	**500 g**
		7232B	**2 kg**
		7232C	**10 kg**

References

1. **Wilkins, T. D., and S. Chalgren.** 1976. Medium for use in antibiotic susceptibility testing of anaerobic bacteria. Antimicrob. Agents. Chemother. **10:**926.
2. **Balows, A., W. J. Hausler, Jr., K. L. Herrmann, H. D. Isenberg, and H. J. Shadmony (eds.).** 1991. Manual of clinical microbiology, 5th ed. American Society for Microbiology, Washington, D.C.
3. **Smith, L. D. S.** 1975. The pathogenic anaerobic bacteria, 2nd ed. Charles C. Thomas, Springfield, Ill.
4. **Isenberg, H. D. (ed.).** 1992. Clinical microbiology procedures handbook. American Society for Microbiology, Washington, D.C.
5. **Murray, P. R., E. J. Baron, M. A. Pfaller, F. C. Tenover, and R. H. Yolken (eds.).** 1995. Manual of clinical microbiology, 6th ed. American Society for Microbiology, Washington, D.C.
6. **Baron, E. J., L. R. Peterson, and S. M. Finegold.** 1994. Bailey & Scott's diagnostic microbiology, 9th ed. Mosby-Year Book, Inc., St. Louis, MO.
7. **www.fda.gov/Food/ScienceResearch/LaboratoryMethods/BacteriologicalAnalyticalmanualBAM/default.htm.**
8. **Vanderzant, C., and D. F. Splittstoesser (eds.).** 1992. Compendium of methods for the microbiological examination of food, 3rd ed. American Public Health Association, Washington, D.C.

Technical Information

Contact Acumedia Manufacturers, Inc. for Technical Service or questions involving dehydrated culture media preparation or performance at (517)372-9200 or fax us at (517)372-2006.

NEOGEN CORPORATION
620 Lesher Place, Lansing MI 48912
517/372-9200 • 800/783-3212 • fax 800/875-8563
neogen-info@neogen.com • www.neogen.com