



# PRODUCT SPECIFICATION SHEET

## Wilkins Chalgren Anaerobic Broth Base (DM293)

### Intended Use

Wilkins Chalgren Anaerobic Broth Base (DM293) is recommended for cultivation and susceptibility testing of anaerobic bacteria.

### Product Summary and Explanation

Anaerobic bacteria are widespread in soil, marshes, lake and river sediments, oceans, sewage, food and animals. In humans, anaerobic bacteria normally are prevalent in the oral cavity around the teeth, in the gastrointestinal tract, in the orifices of the genitourinary tract and on the skin. Anaerobic infections in humans and various animals can involve virtually any organ under immunocompromised conditions.<sup>(1)</sup> Also, anaerobic infections are often associated with tissue necrosis and abscess formation, leading to impaired delivery of antimicrobial agents in blood to the actual site of infection. This explains why anaerobic infections are often aggressively managed with debridement, aspiration and/or surgical removal of infected tissue. Because of the technical and interpretive difficulties associated with anaerobic susceptibility testing, presentation of definitive recommendations is difficult.<sup>(2)</sup> The survival of anaerobic bacteria is dependent on their sensitivity to oxygen, nutritional requirements, appropriate collection, culture medium, and incubation time and temperature.<sup>(3)</sup>

Wilkins Chalgren Anaerobic Broth Base, formulated by Wilkins and Chalgren,<sup>(4)</sup> is the preferred medium for susceptibility testing of anaerobes. This medium is also recommended for testing anaerobic bacteria.<sup>(5,6,7)</sup> Wilkins Chalgren Anaerobic Broth Base is similar to the agar medium, except the agar.<sup>(8)</sup> The broth medium is especially useful in the broth micro-dilution tests.<sup>(9)</sup> This medium was selected because it does not require the addition of blood to support satisfactory growth of most anaerobes. Wilkins Chalgren Broth media need to be appropriately supplemented to support the growth of certain anaerobic bacteria.

### Principles of the Procedure

Wilkins Chalgren Anaerobic Broth Base contains peptic digest of animal tissues and casein enzymic hydrolysate serve as sources of essential nutrients for microbial metabolism 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. Sodium chloride helps to maintain the osmotic balance of the medium. The medium can be made selective for non-spore anaerobic bacteria and gram-negative anaerobic bacteria by addition of NonSpore Anaerobic Supplement (MS022) and G. N. Spore Anaerobic Supplement (MS017) respectively. Hemin and Menadione (Vitamin K<sub>3</sub>) enhances the growth of *Bacteroides* species and *Prevotella melaninogenica*, respectively and many other species of gram-negative anaerobic rods.

### Formula / Liter

Ingredients	Gms / Liter
Casein enzymic hydrolysate	10.00
Peptic digest of animal tissue	10.00
Yeast extract	5.00
Dextrose	1.00
Sodium chloride	5.00
L-Arginine	1.00
Sodium pyruvate	1.00
Hemin	0.005





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Menadione	0.0005
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 only.
2. IRRITANT. Irritating to eyes, respiratory system, and skin.

### Directions

1. Suspend 33 grams in one liter of distilled water.
2. Heat, if necessary, to dissolve the medium completely.
3. Dispense as desired.
4. Autoclave at 121°C, 15 psi pressure, for 15 minutes / validated cycle.
5. Cool to 50°C before adding antibiotics to be tested.
6. Mix gently and dispense into sterile tubes.
7. For cultivation of anaerobes, aseptically add the rehydrated contents of 2 vials each of Non-Spore Anaerobic Supplement (MS022) or G. N. Spore Anaerobic Supplement (MS017) as desired to the sterile molten medium before pouring into sterile Petri plates.

### Quality Control Specifications

<b>Dehydrated Appearance</b>	Cream to yellow homogeneous free flowing powder
<b>Prepared Medium</b>	Medium amber coloured clear solution in tubes.
<b>Reaction of 3.3% solution</b>	pH 7.1 ± 0.2 at 25°C
<b>Gel Strength</b>	Not Applicable

**Expected Cultural Response:** Cultural characteristics observed with added Non-Spore Anaerobic Supplement (MS022) or G.N.Spore Anaerobic Supplement (MS017) Under anaerobic conditions, after an incubation at 35-37°C of 48 hours.

Sr. No.	Organisms	Results to be achieved	
		Inoculum (CFU)	Growth
1.	<i>Bacteroides fragilis</i> ATCC 25285	50-100	good-luxuriant
2.	<i>Clostridium perfringens</i> ATCC 12924	50-100	good-luxuriant
3.	<i>Prevotella melaninogenicus</i> ATCC 15930	50-100	good-luxuriant
4.	<i>Escherichia coli</i> ATCC 25922	>=10 <sup>3</sup>	inhibited

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

### Test Procedure

Refer to appropriate references for standard test procedures for a complete discussion on susceptibility testing of anaerobic bacteria.

### Results

Refer to appropriate references and standard test procedures for interpretation of results.





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### Storage

Store the sealed bottle containing the dehydrated medium at 10 - 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light.

### Expiration

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

### Limitations of the Procedure

1. Anaerobe Broth MIC is supplemented to a final concentration of 0.5 µg per mL of vitamin K<sub>3</sub> and 5.0 µg of hemin per mL.
2. CLSI changed their recommendations to include use of broth with a final concentration of 1 µg of vitamin K<sub>1</sub> per mL.
3. To follow CLSI recommendations, the concentration of vitamin K<sub>3</sub> should be increased accordingly. A final concentration of 0.5 µg of vitamin K<sub>3</sub> per mL is sufficient, but some fastidious anaerobes may need a higher concentration of vitamin K<sub>3</sub>.
4. Consult appropriate texts for detailed information and recommended procedures.

### Packaging

**Product Name : Wilkins Chalgren Anaerobic Broth Base**

**Product Code : DM293**

**Available Pack sizes : 500gm**

### References

1. Koneman E. W., Allen S. D., Janda W. M., Schreckenberger P. C., Winn W. C. Jr., 1992, *Colour Atlas and Textbook of Diagnostic Microbiology*, 4th Ed., J. B. Lippincott Company.
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3. Isenberg, H. D. (ed.). 1992. *Clinical microbiology procedures handbook*. American Society for Microbiology, Washington, D.C.
4. Wilkins T. D. and Chalgren S., 1976, *Antimicrob. Agents Chemother.*, 10 : 926.
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6. Clinical and Laboratory Standards Institute, 2006, *Methods for Antimicrobial Susceptibility Testing of Anaerobic Bacteria*, Approved standard M11-A3, CLSI, Villanova, Pa.
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9. Quinto G. and Sebald M., 1964, *Am. J. Med. Technol.*, 30:381.

### Further Information

For further information please contact your local MICROMASTER Representative.





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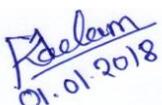
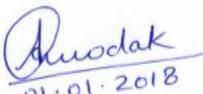


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DM293PSS,QAD/FR/024,Rev.00/01.01.2018

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