

MAST® Culture Media and Supplements

Technical Information Sheet

Product Code DM 235



Wilkins Chalgren Agar

A medium recommended for the culture and antibiotic susceptibility testing of anaerobes.

1. Description

In 1976 Wilkins and Chalgren¹ described a new medium for use in the antibiotic susceptibility testing of anaerobes. The medium was designed to support the growth of most clinically isolated anaerobes without the addition of blood.

Casein and gelatin peptones were used in the formulation, in preference to the less consistent meat peptones, and various growth factors were included. Yeast extract was added to supply vitamins, purines and pyrimidines to enhance the growth of *Bacteroides melaninogenicus* and pyruvate to *Peptostreptococcus aerobius*. Arginine was added to improve the growth of *Eubacterium lentum* and pyruvate to provide an energy source for assacharolytic organisms such as *Veillonella*. Also included were haemin for growth of *B.fragilis* and other *Bacteroides* spp., vitamin K1 for some strains of *B.melaninogenicus* and sodium chloride to make the medium isotonic so that blood could be added when haemolytic reactions need to be detected. Wilkins and Chalgren concluded that the new medium was comparable to or better than media such as Brucella Agar or Schaedler Blood

Agar for the growth of anaerobes. Later Sutter *et al*² recommended the use of Wilkins-Chalgren Agar as the standard medium in a reference method for anaerobic susceptibility testing. Traditionally, antibiotic therapy for anaerobic infections is based on predictable susceptibility patterns and rapid susceptibility testing is not required. However, with the emergence of resistance, susceptibility patterns are changing and it has become desirable to monitor these changes. Susceptibility testing therefore should be carried out as a routine procedure, for reference purposes, retrospectively (Hamilton-Miller³). Numerous techniques have been proposed for anaerobic susceptibility testing but despite practical difficulties the agar dilution test was considered by Brown and Waatti,⁴ to be the most precise method.

The development of MAST ADATAB™ for the preparation of antibiotic containing agar plates has overcome the practical difficulties of the agar dilution method. ADATAB™ is available for all antibiotics currently used in the treatment of anaerobic infections.

2. Technical Formula*

Formula	grams per litre
Enzymic digest of casein	10.0
Pancreatic digest of gelatin	10.0
Yeast extract	5.0
Glucose	1.0
Sodium chloride	5.0
L-arginine hydrochloride	1.0
Sodium pyruvate	1.0
Haemin	0.005
Menadione	0.0005
Agar	12.0

pH approx.7.1

4. In Use

1. After autoclaving allow the medium to cool to 55°C and hold in a waterbath at this temperature.

2a. Preparation of recommended break-point concentrations

Using sterile forceps add one ADATAB™ for each 100ml of medium. Allow the tablets to break up and dissolve completely. Mix well to complete dispersal.

2b. Preparation of alternative concentrations

Different break-point concentration from those recommended can be prepared by varying the volume of medium to which the ADATAB™ is added or by using more than one tablet per volume of medium.

The ADATAB™ can also be placed in a suitable diluent and serial dilutions made from this concentration. In this way a range of concentrations for M.I.C. determinations can be easily prepared.

3. Directions

1. Suspend by swirling 45g of powder in 1 litre or the contents of the sachet in the stated volume of distilled or deionised water.
2. Bring to the boil to dissolve completely.
3. Autoclave at 121°C (15 p.s.i.) for 15 minutes
4. Mix well before pouring.

3. Label Petri dishes with the concentrations to be prepared using the self-adhesive labels provided.

4. Pour culture plates of normal thickness (15-20ml) and allow to set. Dry the plates before use.

5. Prepare a heavy suspension of each organism under test and inoculate onto the plate. The use of the MAST SCAN 400 Multipoint Inoculator or INO1 Hand Inoculator will ensure maximum economy.

5. References

1. Wilkins TD, Chalgren S. *Antimicrobial Agents Chemother.* 1976; **10**: 926-928.
2. Sutter VL, Barry AL, Wilkins TD, Zabransky R. *Antimicrobial Agents Chemother.* 1979; **16**: 495-502
3. Hamilton-Miller JMT. *J Antimicrobial Chemother.* 1975; **1**: 278-289.
4. Brown WJ, Waatti PE. *Antimicrobial Agents Chemother.* 1980; **17**: 629-635.

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