PRODUCT SPECIFICATION SHEET

Schaedler Anerobic Agar (DM238)

Intended Use
Schaedler Anerobic Agar (DM238) is recommended for enumeration of aerobes and anaerobes present in gastrointestinal tract.

Product Summary and Explanation
In 1965, Schaedler, Dubos and Costello[1] reported on the bacterial flora of the gastrointestinal tract of mice. During these studies, several new media formulations were introduced. The majority of these contained inhibitors of specific bacterial species or groups since the authors indicated the need for selective media when processing specimens which contain large numbers of a heterogeneous bacterial population. The basal medium, without inhibitors, is the original version of the medium designated as Schaedler Agar. It was formulated to support the growth of fastidious anaerobic microorganisms such as lactobacilli, streptococci, clostridia and *Bacteroides*. Mata and coworkers,[2] studying the fecal microflora in healthy persons in Central America, further modified Schaedler Agar to produce a number of new formulations. Schaedler Agar serves as an excellent basal media to which blood or other enrichments can be added to enhance the recovery of fastidious anaerobic organisms.
The modifications in the basal medium of Schaedler included adjustments in the peptone content and an increase in the sodium chloride content. Additionally, the dextrose concentration was reduced to avoid interference with haemolytic reactions and the yeast extract level lowered to avoid darkening of the medium.[3]
Schaedler Agar supplemented with Vitamin K_1 and 5% sheep blood is used for the recovery of fastidious anaerobic bacteria such as *Bacteroides*. Inclusion of Colistin and Nalidixic acid in the formulation (Schaedler CNA Agar) along with 5% sheep blood is used for the selective isolation of the anaerobic gram-positive cocci,[4] *Peptococcus* and *Peptostreptococcus* species. Inclusion of Kanamycin and Vancomycin in the formulation (Schaedler KV Agar) along with 5% sheep blood is used for selective isolation of gram-negative anaerobes. Addition of Sodium Polyanethol Sulphonate (SPS) is recommended when using this medium for blood culture.[5] It inhibits phagocytosis and neutralizes the antibacterial activity of fresh blood components.[6,7] Vitamin K_1 enables the cultivation of *Bacteroides melaninogenicus*[8] and stimulates growth of other *Bacteroides* species and gram-positive spore formers.[9]

Principles of the Procedure
Schaedler Anerobic Agar contains combination of casein enzymic hydrolysate, proteose peptone and papaic digest of soyabean meal, yeast extract and Lcystine provide nitrogenous growth factors, vitamins and other essential growth nutrients. Dextrose serves as energy and carbon source. Hemin and sheep blood stimulates the growth of fastidious microorganisms and stimulates growth of other *Bacteroides* species and gram-positive spore formers.

Formula / Liter

Ingredients	Gms / Liter
Casein enzymic hydrolysate	5.67
Proteose peptone	5.00
Papaic digest of soyabean meal	1.00
Yeast extract	5.00
Dextrose	5.83
Sodium chloride	1.67
Dipotassium hydrogen phosphate	0.83
Tris hydroxymethyl aminomethane	3.00
L-Cystine	0.40
Hemin	0.01
Agar	15.00
Final pH: 7.6 ± 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 43.41 grams in 950 ml distilled water.
2. Heat to boiling with frequent agitation to dissolve the medium completely.
3. Autoclave at 121°C, 15 psi pressure, for 15 minutes / validated cycle.
4. Cool to 45-50°C and add 5% sterile defibrinated blood if desired.
5. Mix well before dispensing.
6. Avoid overheating and photooxidation of the medium, as it will retard the growth of bacteria.
7. If desired, add rehydrated contents of 1 vial each of Vitamin K_1 Supplement (MS019) and CNA Supplement (MS021) to prepare Schaedler CNA Agar or to prepare Schaedler KV Agar, aseptically add rehydrated contents of 1 vial each of Vitamin K_1 Supplement (MS019) and KV Supplement (MS014) respectively to 1000 ml of Schaedler Agar.

Quality Control Specifications

Dehydrated Appearance	Cream to yellow homogeneous free flowing powder
Prepared Medium	Light amber coloured clear to slightly opalescent gel forms in Petri plates
Reaction of 4.34% Solution	pH : 7.6 ± 0.2 at 25°C
Gel Strength	Firm, comparable with 1.5% Agar gel

Expected Cultural Response: Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours under anaerobic condition.

Sr. No.	Organisms	Results to be achieved		
		Inoculum (CFU)	Growth	Recovery
1.	*Bacteroides fragilis ATCC 25285*	50 - 100	good-luxuriant	>=50%
2.	*Clostridium butyricum ATCC 13732*	50 - 100	good-luxuriant	>=50%
3.	*Clostridium perfringens ATCC 12924*	50 - 100	good-luxuriant	>=50%
4.	*Clostridium sporogenes ATCC 11437*	50 - 100	good-luxuriant	>=50%
5.	*Escherichia coli ATCC 25922*	>=10^3	inhibited	0%
6.	*Streptococcus pyogenes ATCC 19615*	50 - 100	good-luxuriant	>=50%

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

Test Procedure

1. These media should be reduced immediately prior to inoculation by placing them under anaerobic conditions for 18-24 hours.
2. Use standard procedures to obtain isolated colonies from specimens.
3. Inoculate an enrichment broth, such as Enriched Thioglycollate Medium, at the same time as the primary plates to detect small numbers of anaerobes.
4. Incubate plates and tubes immediately after inoculation, with plates in an inverted position (agar side up) under anaerobic conditions at 35°C, or place the media in a holding jar flushed with oxygen-free gas(es) until a sufficient number of plates and tubes is accumulated (no longer than 3 hours).
5. Incubate for at least 48 hours and, if no growth occurs, continue incubation for up to 7 days. It is recommended that an indicator of anaerobiosis be used.
6. Examine the selective medium for growth after 48 hours of incubation. Cultures should not be regarded as negative until after 7 days incubation.
7. Since some anaerobes may be inhibited due to the selective nature of the medium, it is advisable to include a nonselective medium such as Schaedler Agar with Vitamin K1 and 5% Sheep Blood.

Results

1. In order to determine the relationship to oxygen of each colony type present on the medium, follow established procedures.

2. The colony types that prove to contain obligate anaerobes can be further studied.

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. For identification, organisms must be in pure culture. Morphological, biochemical and/or serological tests should be performed for final identification.
2. Consult appropriate texts for detailed information and recommended procedures.

Packaging
Product Name : Schaedler Anerobic Agar
Product Code : DM238
Available Pack sizes : 500gm

References

1. Schaedler R.W., Dubos R. and Castello R., 1965, J. Exp. Med., 122:59.
2. Mata L.J., Carrillo C. and Villatoro E., 1969, Appl. Microbiol., 17:596.
3. MacFaddin J., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol.I, Williams and Wilkins, Baltimore.
4. Estevez, 1984, Lab Med., 15:258.
5. Rosner, 1968, Am. J. Clin. Pathol., 49:216.
6. Garrod, 1966, J. Pathol. Bacteriol., 91:621.
7. Lowrence and Traub, 1969, Appl. Microbiol., 17:839.
8. Gibbons R. J. and MacDonald J. B., 1960, J. Bacteriol., 80:164.
9. Finegold et al, 1974, Manual of Clinical Microbiology, 2nd ed., Lennette and others (Eds.), ASM, Washington, D.C.

Further Information
For further information please contact your local MICROMASTER Representative.

MICROMASTER LABORATORIES PRIVATE LIMITED
Unit 38/39, Kalpataru Industrial Estate,
Off G.B. Road, Near 'R-Mall', Thane (W) – 400607. M.S. INDIA.
Ph: +91-9320126789/9833630009/9819991103
Email: sales@micromasterlab.com

DM238PSS, QAD/FR/024, Rev.00