

PRODUCT SPECIFICATION SHEET

(DM012)

Intended Use

Anaerobic Agar (Brewer) (DM012) is recommended for study of colonial morphology, isolation and sensitivity testing of anaerobic and microaerophilic organisms.

Product Summary and Explanation

Among the vast number of organisms that belong to the Kingdom Fungi, pathogenic fungi constitute a very small group. Fungi that belong to the genera *Aspergillus*, *Candida*, *Cryptococcus*, *Histoplasma* and *Pneumocystis* have the potential to cause human diseases. Members of pathogenic fungi group are scattered throughout four taxonomic classes based on their methods of reproduction viz. *Zygomycetes*, *Basidiomycetes*, *Ascomycetes* and *Deuteromycetes* (Fungi Imperfecti).⁽¹⁾ To confirm the existence and nature of infection by fungi and yeasts, direct methods are more important than indirect methods; identification of the organisms is much more useful than demonstrating the humoral and cellular responses of the host.⁽²⁾

Inhibitory Mold Agar, containing Chloramphenicol, is a moderately selective medium formulated by Ulrich.⁽³⁾ This medium can be used as a general cultivation medium for various strains of pathogenic fungi, especially *Histoplasma capsulatum* and dermatophytes. Chloramphenicol selectively inhibits saprophytic fungi and bacteria while allowing pathogenic fungi to grow. Adding antimicrobial agents to media for the isolation of pathogenic fungi is documented.⁽⁴⁻⁶⁾ Selective fungal media are recommended for the isolation of dermatophytes because these pathogens are not sensitive to Chloramphenicol.⁽⁷⁾

Principles of the Procedure

Mold Inhibitory Agar, Ulrich casein enzymic hydrolysate and peptic digest of animal tissue which provides nitrogen, carbon, and amino acids essential growth nutrients. Yeast extract is a rich source of vitamin B complex. Dextrose, starch and dextrin are energy sources for the metabolism of fungi. Sodium chloride and metallic salts provide essential ions and minerals thereby helps to maintain the osmotic balance of the medium. Chloramphenicol is a broad-spectrum antibiotic inhibits a wide variety of gram-positive and gram-negative bacteria. Potential contaminants of cosmetics and toiletries like *Pseudomonas aeruginosa* and *Serratia marcescens* are effectively inhibited by chloramphenicol. Sodium phosphates buffer the medium.

Formula / Liter

Ingredients	Gms / Liter
Proteose peptone	10.00
Casein enzymic hydrolysate	5.00
Yeast extract	5.00
Dextrose	10.00
Sodium chloride	5.00
Sodium thioglycollate	2.00
Sodium formaldehyde sulfoxylate	1.00
Resazurin	0.002
Agar	15.00
Final pH: 7.2 ± 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 53 grams of the medium in one liter of distilled water.
2. Mix thoroughly and heat to boiling to dissolve the medium completely.
3. Autoclave at 121°C, 15 psi pressure, for 15 minutes / validated cycle.
4. Mix well and pour into sterile petri plates.

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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 that becomes red due to aeration on standing.
Reaction of 5.3% solution	pH 7.2 \pm 0.2 at 25°C
Gel Strength	Firm, comparable with 1.5% Agar gel

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

Sr. No.	Organisms	Results to be achieved		
		Inoculum (CFU)	Growth	Recovery
1.	<i>Clostridium botulinum</i> ATCC 19397	50-100	good-luxuriant	\geq 50%
2.	<i>Clostridium perfringens</i> ATCC 12924	50-100	good-luxuriant	\geq 50%
3.	<i>Clostridium sporogenes</i> ATCC 11437	50-100	good-luxuriant	\geq 50%

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

Test Procedure

1. Refer to appropriate references for information about the processing and inoculation of specimens.
2. For isolation of fungi from potentially contaminated specimens, a nonselective medium should be inoculated along with the selective medium.
3. Incubate the plates at 25-30°C in an inverted position (agar side up) with increased humidity.
4. The tubed slants also should be incubated at 25-30°C.
5. For isolation of fungi causing systemic mycoses, two sets of media should be inoculated, with one set incubated at 25-30°C and a duplicate set at 35 \pm 2°C.
6. All cultures should be examined at least weekly for fungal growth and should be held for 4-6 weeks before being reported as negative.

Results

Examine plates for fungal colonies exhibiting typical color and morphology. Biochemical tests and serological procedures should be performed to confirm findings.

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. Antimicrobial agents incorporated into a medium to inhibit bacteria may also inhibit certain pathogenic fungi. Primary isolation should include use of both non-selective and selective media.
2. For identification, organisms must be in pure culture. Morphological, biochemical and/or serological tests should be performed for final identification.
3. Consult appropriate texts for detailed information and recommended procedures.

Packaging

Product Name : Anaerobic Agar (Brewer)

Product Code : DMO12

Available Pack sizes : 500gm

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References

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3. Ulrich, J. A. 1956. Media and methods for the isolation and identification of pathogenic fungi. Bacteriol. Proc. SAB, M75, p. 87.
4. Georg, L. K., L. Ajello, and C. Papageorge. 1954. Use of cycloheximide in the selective isolation of fungi pathogenic to man. J. Lab Clin. Med. 44:422-428.
5. Murray, P. R., E. J. Baron, M. A. Pfaller, F. C. Tenover, and R. H. Tenover (eds.). 1995. Manual of clinical microbiology, 6th ed. American Society for Microbiology, Washington, D.C.
6. MacFaddin, J. F. 1985. Media for isolation-cultivation-identification-maintenance of medical bacteria, vol. 1. Williams & Wilkins, Baltimore, MD.
7. Georg, L. K., L. Ajello, E. S. McDonough, and S. Brinkman. 1960. In vitro effects of antibiotics on yeast phase of *Blastomyces dermatitidis* and other fungi. J. Lab & Clin. Med. 55:116-119.

Further Information

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