



PRODUCT SPECIFICATION SHEET

Phenol Red Mannitol Agar (DM198)

Intended Use

Phenol Red Mannitol Agar (DM198) is recommended for mannitol fermentation studies of microorganisms.

Product Summary and Explanation

Phenol Red Agar media are recommended⁽¹⁻⁴⁾ for studying the fermentation of various carbohydrates individually by the pure cultures of microorganisms. Liquid media are generally employed in studying fermentation reactions; but many bacteriologists prefer a solid medium for this purpose. One advantage of employing solid fermentation medium is that it permits observation of fermentation reactions under both aerobic and anaerobic conditions. Deep tubes are used which can provide sufficient anaerobic conditions for the growth of obligate anaerobic bacilli. Formation of any gas that occurs during a reaction is indicated by splitting of the agar or accumulation of gas bubbles at the base.

Principles of the Procedure

Phenol Red Agar Base contains proteose peptone and beef extract which provides the carbon and nitrogen required for good growth of a wide variety of organisms. Sodium chloride maintains the osmotic balance of the medium. Mannitol is the fermentable carbohydrate. Phenol red serves as a pH indicator. A positive mannitol fermentation reaction is indicated by the production of a yellow colour in agar due to the effect of acid production. If the carbohydrate is not fermented, the medium remains red or becomes alkaline (darker red).

Formula / Liter

Ingredients	Gms / Liter
Proteose peptone	10.00
Beef extract	1.00
Sodium chloride	5.00
Mannitol	10.00
Phenol red	0.025
Agar	15.00
Final pH: 7.4 ± 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 41.02 grams of the medium in one liter of distilled water.
2. Heat to boiling, to dissolve the medium completely, with frequent agitation.
3. Dispense in tubes or flasks as desired.
4. Autoclave at 121°C, 15 psi pressure, for 15 minutes / validated cycle.
5. Allow the tubed media to cool in slanted position to form slants with deep butts.

Quality Control Specifications

Dehydrated Appearance	Light yellow to pink homogeneous free flowing powder
Prepared Medium	Red coloured clear to slightly opalescent gel forms in tubes as slants
Reaction of 4.1% solution	pH 7.4 ± 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 18-24 hours.

Sr. No.	Organisms	Results to be achieved			
		Inoculum (CFU)	Growth	Acid	Gas



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1.	<i>Alcaligenes faecalis</i> ATCC8750	50-100	good-luxuriant	negative reaction, no colour change	negative reaction
2.	<i>Escherichia coli</i> ATCC 25922	50-100	good-luxuriant	positive reaction, yellow colour	positive reaction
3.	<i>Klebsiella pneumonia</i> ATCC 13883	50-100	good-luxuriant	positive reaction, yellow colour	positive reaction
4.	<i>Proteus vulgaris</i> ATCC 13315	50-100	good-luxuriant	negative reaction, no colour change	negative reaction
5.	<i>Salmonella Typhimurium</i> ATCC 14028	50-100	good-luxuriant	positive reaction, yellow colour	positive reaction
6.	<i>Shigella flexneri</i> ATCC 12022	50-100	good-luxuriant	positive reaction, yellow colour	negative reaction

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

Test Procedure

1. Using a straight wire inoculate the medium by stabbing into the butt and streaking the surface of the slant.
2. If desired, inoculate obligate anaerobic bacteria into melted medium that has been cooled to 45°C. Prior to incubation allow the agar to solidify.
3. Incubate at 35 ± 2°C for 18-24 hours (or anaerobically for 24-72 hours).
4. Examine periodically for growth, acid production and gas formation.

Results

1. Fermentation of the carbohydrate is indicated by a change in the color of the medium from red to yellow.
2. Gas formation is indicated by the collection of gas bubbles in the base or by splitting of the agar.

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. When inoculating tubes, stab gently and do not use a loop. Rough stabbing or using a loop to stab may give the false appearance of gas production when mechanical splitting of the medium is what actually occurred.
2. Consult appropriate texts for detailed information and recommended procedures.

Packaging

Product Name : Phenol Red Mannitol Agar

Product Code : DM198

Available Pack sizes : 100gm / 500gm

References

1. Forbes, Sahmand Weissfeld. 2007. Bailey & Scott's diagnostic microbiology, 12th edition. Mosby, Inc., St. Louis, Mo.
2. Murray, Baron, Jorgensen, Landry and Pfaller (ed.). 2007. Manual of clinical microbiology, 9th edition. American Society for Microbiology, Washington, D.C.
3. Holt, Krieg, Sneath, Staley and Williams. 1994. Bergey's Manual of determinative bacteriology, 9th edition. Williams & Wilkins, Baltimore, Md.™
4. Ewing. 1986. Edwards and Ewing's identification of *Enterobacteriaceae*, 4th edition. Elsevier Science Publishing Co., Inc., New York, N.Y.





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Further Information

For further information please contact your local MICROMASTER Representative.



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