



PRODUCT SPECIFICATION SHEET

MacConkey Agar w/CV, NaCl and 0.15% Bile Salts (DM147)

Intended Use

MacConkey Agar w/ CV, NaCl and 0.15% Bile salts (DM147) is recommended for identification of *Enterobacteriaceae* in the presence of coliforms and lactose non-fermenters from water, sewage, and food products.

Product Summary and Explanation

MacConkey Agar is based on the bile salt-neutral red-lactose agar of MacConkey.⁽¹⁾ MacConkey Agar is the earliest selective and differential medium for cultivation of enteric microorganisms from a variety of clinical specimens.^(1,2) Subsequently MacConkey Agar and Broth have been recommended for use in microbiological examination of foodstuffs⁽³⁾ and for direct plating / inoculation of water samples for coliform counts.⁽⁴⁾ These media are also accepted by the Standard Methods for the Examination of Milk and Dairy Products^(5,6) and pharmaceutical preparations. The original MacConkey medium was used to differentiate strains of *Salmonella typhosa* from members of the coliform group. Modifications made in the original formula improved the growth of *Shigella* and *Salmonella* strains. These modifications included the addition of 0.5% sodium chloride, decreased agar content, and altered bile salts and neutral red concentrations. The formula improvements gave improved differential reactions between these enteric pathogens and the coliform group. MacConkey Agar is listed as one of the recommended media for the isolation of *E. coli* from non-sterile pharmaceutical products.⁽⁷⁾

MacConkey Agar contains crystal violet and bile salts which attributes selective action to this medium, that inhibit gram-positive organisms and allow gram-negative organisms to grow. Gram-negative bacteria usually grow well on the medium and are differentiated by their ability to ferment lactose. Lactose fermenting strains grow as red or pink and may be surrounded by a zone of acid precipitated bile. The red colour is due to production of acid from lactose, absorption of neutral red and a subsequent colour change of the dye when the pH of medium falls below 6.8. Lactose non-fermenting strains, such as *Shigella* and *Salmonella* are colourless and transparent and typically do not alter appearance of the medium. When lactose non-fermenters grow in proximity to coliform colonies, the surrounding medium appears as cleared areas.

Principles of the Procedure

Peptic digest of animal tissue serves as the main source of nitrogen and vitamins. Lactose serves as the carbon source by being the fermentable carbohydrate. Bile salts and crystal violet serve to make the medium selective by inhibiting accompanying gram-positive bacteria and allowing gram-negative organisms to grow. Neutral red is the pH indicator dye while sodium chloride maintains the osmotic equilibrium of the medium.

Formula / Liter

Ingredients	Gms / Liter
Peptic digest of animal tissue	20.00
Lactose	10.00
Sodium chloride	5.00
Bile salt	1.50
Neutral red	0.05
Crystal violet	0.001
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 51.55 grams of the medium in one liter of distilled water.
2. Heat to boiling with gentle swirling to dissolve the agar completely.
3. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.
4. Avoid overheating. Cool to 45 - 50°C and pour into sterile Petri plates.
5. The surface of the medium should be dry when inoculated.



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Quality Control Specifications

Dehydrated Appearance	Light yellow to pink homogeneous free flowing powder
Prepared Medium	Red with purplish tinge coloured, clear to slightly opalescent gel forms in Petri plates
Reaction of 5.15% Solution	pH: 7.2 ± 0.2 at 25°C
Gel Strength	Firm, comparable with 1.5% Agar gel

Growth Promotion Test

Growth Promotion is carried out in accordance with the harmonized method of IP.

Expected Cultural Response: Cultural characteristics observed after an incubation at 30-35 °C for 18-72 hours. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar (DM247).

Sr. No.	Organisms	Results to be achieved				
		Inoculum (CFU)	Growth	Lot value (CFU)	Recovery	Colour of colony
1.	<i>Escherichia coli</i> ATCC 25922	50 -100	good-luxuriant	25 -100	≥50 %	pink to red with bile precipitate
2.	<i>Escherichia coli</i> NCTC 9002	50 -100	good-luxuriant	25 -100	≥50 %	pink to red with bile precipitate
3.	<i>Enterobacter aerogenes</i> ATCC 13048	50-100	good-luxuriant	25 -100	≥50 %	pink to red
4.	<i>Enterococcus faecalis</i> ATCC 29212	50-100	fair to good	15-40	30-40%	colourless to pink
5.	<i>Proteus vulgaris</i> ATCC 13315	50 -100	good-luxuriant	25 -100	≥50 %	colourless
6.	<i>Salmonella Paratyphi A</i> ATCC 9150	50 -100	good-luxuriant	25 -100	≥50 %	colourless
7.	<i>Shigella flexneri</i> ATCC 12022	50 -100	fair to good	15-40	30-40%	colourless
8.	<i>Salmonella Paratyphi B</i> ATCC 8759	50 -100	good-luxuriant	25 -100	≥50 %	colourless
9.	<i>Salmonella Enteritidis</i> ATCC 13076	50 -100	good-luxuriant	25 -100	≥50 %	colourless
10.	<i>Salmonella Typhi</i> ATCC 6539	50 -100	good-luxuriant	25 -100	≥50 %	colourless
11.	<i>Staphylococcus aureus</i> ATCC 25923	≥10 ³	inhibited	0	0%	--
12.	<i>Staphylococcus aureus</i> ATCC 6538	≥10 ³	inhibited	0	0%	--
13.	<i>Salmonella Typhimurium</i> ATCC 14028	50 -100	good-luxuriant	25 -100	≥50 %	colourless
14.	<i>Staphylococcus epidermidis</i> ATCC 12228	≥10 ³	inhibited	0	0%	--
15.	<i>Corynebacterium diphtheriae</i> type gravis	≥10 ³	inhibited	0	0%	--

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

Test Procedure

Refer to appropriate references using MacConkey Agar for the isolation and identification of enteric organisms.

Results

1. Lactose-fermenting organisms grow as pink to red colonies with or without a zone of precipitated bile.
2. Non-lactose fermenting organisms grow as colorless or clear colonies.



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MacConkey Agar (DM143)

Lactose fermenting colonies of *Escherichia coli* ATCC 25922 & Non-lactose fermenting colonies of *Salmonella typhi* ATCC 6539

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. Although MacConkey Agar w/ CV, NaCl and 0.15% Bile salt is a selective medium primarily for Gram-negative enteric bacilli, biochemical and serological testing using pure cultures are recommended for complete identification.
2. Consult appropriate texts for detailed information and recommended procedures.
3. Incubation of MacConkey Agar w/ CV, NaCl and 0.15% Bile salt plates under increased CO₂ has been reported to reduce the growth and recovery of a number of strains of gram-negative bacilli.

Packaging

Product Name : MacConkey Agar w/ CV, NaCl and 0.15% Bile salt

Product Code : DM147

Available Pack sizes : 100gm / 500gm

References

1. MacConkey, 1905, J. Hyg., 5:333.
2. MacConkey, 1900, The Lancet, ii:20.
3. Downes F. P. and Ito K. (Ed.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th ed., APHA, Washington, D.C.
4. Greenberg A. E., Clesceri L. S. and Eaton A. D., (Eds.), 2005, Standard Methods for the Examination of Water and Wastewater, 21st ed., APHA, Washington, D.C.
5. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
6. The United States Pharmacopoeia XXI and the National Formulary, 16th ed., 1985, United States Pharmacopoeial Convention, Inc., Washington, D.C.
7. United States Pharmacopeial Convention, Inc. 2008. The United States pharmacopeia 31/The national formulary 26, Supp. 1, 8-1-08, online. United States Pharmacopeial Convention, Inc., Rockville, Md.





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Further Information

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