

MacConkey Agar, Granulated

GMH081

MacConkey Agar, granulated is recommended for selective isolation and differentiation of *E.coli* and other enteric bacteria from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP.

Composition**

Ingredients	Gms / Litre
Pancreatic digest of gelatin	17.000
Peptones (meat and casein)	3.000
Lactose monohydrate	10.000
Sodium chloride	5.000
Bile salts	1.500
Neutral red	0.030
Crystal violet	0.001
Agar	13.500
pH after sterilization (at 25°C)	7.1±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 49.53 grams (the equivalent weight of dehydrated medium per litre) in 1000 ml purified/distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well before pouring into sterile Petri plates. The surface of the medium should be dry when inoculated.

Principle And Interpretation

MacConkey Agar is the earliest selective and differential medium for cultivation of coliform organisms (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). This medium is also accepted by the Standard Methods for the Examination of Milk and Dairy Products (5). It is recommended in pharmaceutical preparations and is in accordance with the harmonized method of USP/EP/BP/JP (6,7,8,9).

Pancreatic digest of gelatin and peptones (meat and casein) provide the essential nutrients, vitamins and nitrogenous factors required for growth of microorganisms. Lactose monohydrate is the fermentable source of carbohydrate. The selective action of this medium is attributed to crystal violet and bile salts, which are inhibitory to most species of gram-positive bacteria. Sodium chloride maintains the osmotic balance in the medium.

After enrichment of *Escherichia coli* in MacConkey Broth (GMH083/ MH083), it is then subcultured on MacConkey Agar. 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. *Yersinia enterocolitica* may appear as small, non-lactose fermenting colonies after incubation at room temperature.

Quality Control

Appearance

Light yellow to pink coloured granular medium

Gelling

Firm comparable with 1.35% Agar gel.

Colour and Clarity of prepared medium

Red with purplish tinge coloured clear to slightly opalescent gel forms in Petri plates.

pH

6.90-7.30

Cultural Response

Growth Promotion is carried out in accordance with the harmonized method of ICH (USP/EP/BP/JP). Cultural response was observed after an incubation at 30-35°C for 18-72 hours. Recovery rate is considered as 100% for bacteria growth on Soybean Casein Digest Agar.

Growth promoting properties

Growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating 100 cfu (at 30-35°C for ≤18 hours).

Indicative properties

Colonies are comparable in appearance and indication reaction to those previously obtained with previously tested and approved lot of medium occurs for the specified temperature for a period of time within the range specified inoculating ≤100 cfu (at 30-35°C for 18-72 hours).

Cultural Response

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony	Incubation period
Growth Promoting + Indicative					
<i>Escherichia coli</i> ATCC 8739	50 -100	luxuriant	≥50 %	pink-red with bile precipitate	18 -72 hrs
Additional Microbiological testing					
<i>Escherichia coli</i> ATCC 25922	50 -100	luxuriant	≥50 %	pink to red with bile precipitate	18 -24 hrs
<i>Escherichia coli</i> NCTC 9002	50 -100	luxuriant	≥50 %	pink to red with bile precipitate	18 -24 hrs
<i>Enterobacter aerogenes</i> ATCC 13048	50 -100	luxuriant	≥50 %	pink to red	18 -24 hrs
<i>Enterococcus faecalis</i> ATCC 29212	50 -100	fair to good	30 -40 %	colourless to pale pink	18 -24 hrs
<i>Salmonella</i> Typhimurium ATCC 14028	50 -100	luxuriant	≥50 %	colourless	18 -24 hrs
<i>Staphylococcus aureus</i> ATCC 6538	≥10 ³	inhibited	0 %		≥24 hrs
<i>Staphylococcus aureus</i> ATCC 25923	≥10 ³	inhibited	0 %		≥24 hrs
<i>Salmonella</i> Enteritidis ATCC 13076	50 -100	luxuriant	≥50 %	colourless	18 -24 hrs
<i>Salmonella</i> Paratyphi A ATCC 9150	50 -100	luxuriant	≥50 %	colourless	18 -24 hrs
<i>Salmonella</i> Paratyphi B ATCC 8759	50 -100	luxuriant	≥50 %	colourless	18 -24 hrs
<i>Salmonella</i> Typhi ATCC 6539	50 -100	luxuriant	≥50 %	colourless	18 -24 hrs
<i>Salmonella</i> Abony NCTC 6017	50 -100	luxuriant	≥50 %	colourless	18 -24 hrs
<i>Proteus vulgaris</i> ATCC 13315	50 -100	luxuriant	≥50 %	colourless	18 -24 hrs
<i>Shigella flexneri</i> ATCC 12022	50 -100	fair to good	30 -40 %	colourless	18 -24 hrs
<i>Staphylococcus epidermidis</i> ATCC 12228	≥10 ³	inhibited	0 %		≥24 hrs
<i>Corynebacterium diphtheriae</i> type gravis	≥10 ³	inhibited	0 %		≥24 hrs

Storage and Shelf Life

Please refer disclaimer Overleaf.

Store below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label.

Reference

1. MacConkey, 1900, The Lancet, ii:20.
2. MacConkey, 1905, J. Hyg., 5:333.
3. Downes F P and Ito K (Eds.), 2001, Compendium of Methods For The Microbiological Examination of Foods, 4th ed., APHA, Washington, D.C
4. Eaton A. D., Clesceri L. S. and Greenberg A W., (Eds.), 2005, Standard Methods for the Examination of Water and Wastewater, 21st ed., APHA, Washington, D.C.5. ,
5. Wehr H M and Frank J H., 2004, Standard Methods for the Examination of Dairy Products,, 17th ed., APHA Inc., Washington, D.C.
6. The United States Pharmacopoeia, 2014, The United States Pharmacopoeial Convention, Rockville, MD.
7. British Pharmacopoeia, 2014, The Stationery Office British Pharmacopoeia.
8. European Pharmacopoeia, 2014, European Department for the Quality of Medicines of Council of Europe.
9. Japanese Pharmacopoeia, 2008, Published by Society of Japanese Pharmacopoeia, Tokyo, Japan.

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