Technical Information

MacConkey Agar

Product Code: DM 1081H

Application: MacConkey Agar is recommended for selective isolation of *Escherichia coli* from pharmaceutical products and is in accordance with methodology of BP. It is also recommended for selective isolation and differentiation of lactose fermenting and lactose non fermenting enteric bacteria.

Composition**

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

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

MacConkey Agar is one of the earliest selective and differential medium for cultivation of coliform organisms [1, 2]. Subsequently MacConkey Agar and Broth has also been recommended for microbiological examination of foodstuffs [3] direct plating / inoculation of water samples [4] and examination of Milk and Dairy Products using Standard Methods [5]. British pharmacopoeia [6] has recommended this medium for the subculture and identification of *Escherichia coli.* It has also been named as Agar Medium H. and is also recommended by USP/BP/EP/JP [6-9] for the name.

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 (DM1083B), 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 bacteria 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 bacteria 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.

Methodology

Suspend 49.53 grams of dehydrated medium in 1000 ml purified/distilled water. Shake well & heat to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes i.e. validated cycle. AVOID OVERHEATING. Cool to 45-50°C. Mix well before pouring into sterile Petri plates. The surface of the medium should be dry when inoculated.

Quality Control

Physical Appearance

Light yellow to pink homogeneous free flowing powder

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 Range:- 6.90-7.30

Cultural Response/Characteristics

DM 1081H: Growth Promotion is carried out in accordance with the harmonized method of BP. 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 <=18-72 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).

Organism	Inoculum(CFU)	Growth	Observed Lot value (CFU)	Recovery	Colour of colony	Incubation temperature	Incubation period
Escherichia coli ATCC 8739	50 -100	luxuriant	25 -100	>=50 %	pink-red with hrs bile precipitate	30 -35 °C	18 -24 hrs
Escherichia coli ATCC 25922	50 -100	luxuriant	25 -100	>=50 %	pink to red with hrs bile precipitate	30 -35 °C	18 -24 hrs
Enterobacter aerogenes ATCC 13048	50 -100	luxuriant	25 -100	>=50 %	pink to red with bile precipitate	30 -35 °C	18 -24 hrs
Enterococcus faecalis ATCC 29212	50 -100	luxuriant	25 -100	>=50 %	pink to red colourless to	30 -35 °C	18 -24 hrs
Salmonella Typhimurium ATCC 14028	50 -100	luxuriant	0	30 -40 %	colourless	30 -35 °C	18 -24 hrs
Staphylococcus aureus ATCC 6538	>=10^3	luxuriant	0	>=50 %	colourless	30 -35 °C	18 -24 hrs
Staphylococcus aureus ATCC 25923	>=10^3	luxuriant	25 -100	0 %	colourless	30 -35 °C	>=24 hrs
Salmonella Enteritidis ATCC 13076	50 -100	luxuriant	25 -100	0 %	colourless	30 -35 °C	>=24 hrs
Salmonella Paratyphi A ATCC 9150	50 -100	luxuriant	25 -100	>=50 %	colourless	30 -35 °C	18 -24 hrs
Salmonella Paratyphi B ATCC 8759	50 -100	luxuriant	25 -100	>=50 %	colourless	30 -35 °C	18 -24 hrs
Salmonella Typhi ATCC 6539	50 -100	luxuriant	25 -100	>=50 %	colourless	30 -35 °C	18 -24 hrs
Salmonella Abony NCTC 6017	50 -100	luxuriant	25 -100	>=50 %	colourless	30 -35 °C	18 -24 hrs
Proteus vulgaris ATCC 13315	50 -100	luxuriant	25 -100	>=50 %	colourless	30 -35 °C	18 -24 hrs
Shigella flexneri ATCC 12022	50 -100	fair-good	25 -100	>=50 %	colourless	30 -35 °C	18 -24 hrs
Staphylococcus epidermidis ATCC 12228	>=10^3	inhibited	25 -100	>=50 %	-	30 -35 °C	>=24 hrs
Corynebacterium diphtheriae type gravis	>=10^3	inhibited	15 -40	30 -40 %	-	30 -35 °C	>=24 hrs

Storage and Shelf Life-

Dried Media: Store below 30°C in tightly closed container and use before expiry date as mentioned on the label.
Prepared Media: 2-8^0 in sealable plastic bags for 2-5 days.

Further Reading

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, Washingt on, 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. British Pharmacopoeia 2011, The Stationery office British Pharmacopoeia
7. The United States Pharmacopoeia 2011, The United States Pharmacopoeial Convention. Rockville, MD.
8. European Pharmacopoeia 2011, European Dept. for the quality of Medicines
9. Japanese Pharmacopoeia, 2008.

Disclaimer :

- User must ensure suitability of the product(s) in their application prior to use.
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