



## MacConkey Agar w/ CV w/o NaCl

M1582

MacConkey Agar w/ CV w/o NaCl is a differential medium recommended for the selection and recovery of the *Enterobacteriaceae* and related enteric gram-negative bacilli.

### Composition\*\*

Ingredients	Gms / Litre
Peptic digest of animal tissue	1.500
Casein enzymic hydrolysate	1.500
Pancreatic digest of gelatin	17.000
Lactose	10.000
Bile salts	1.500
Crystal violet	0.001
Neutral red	0.030
Agar	15.000
Final pH ( at 25°C)	7.1±0.2

\*\*Formula adjusted, standardized to suit performance parameters

### Directions

Suspend 46.53 grams in 1000 ml distilled water. Heat to boiling with gentle swirling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Avoid overheating. Cool to 45-50°C and pour into sterile Petri plates. The surface of the medium should be dry when inoculated.

### Principle And Interpretation

The MacConkey media are well known and popular enrichment system for coliform bacteria. MacConkey Agar is the earliest selective and differential medium for cultivation of enteric microorganisms from a variety of clinical specimens (1, 2). Original formulation of MacConkey included ox bile as inhibitor of gram-positive bacteria and litmus as the indicator of the acid production from lactose. Later litmus was substituted by phenol red indicator to make interpretation easier and more precise. The most significant modification to the original formulation is the substitution of ox bile by purified bile salts that improve the selectivity and avoid the inherent turbidity which is due to the fat material of the bile. Another modification was the inclusion of supplementary inhibitors such as crystal violet and pH indicator neutral red.

MacConkey Agar has been recommended for use in microbiological examination of foodstuffs (3) and for direct plating 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).

Original medium contains protein, bile salts, sodium chloride and two dyes. Omission of sodium chloride from the medium prevents the spreading of *Proteus* colonies (6). The selective action of this medium is attributed to crystal violet and bile salts, which are inhibitory to most species of gram-positive bacteria. 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 coloured colonies 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.

### Quality Control

#### Appearance

Light yellow to light pink homogeneous free flowing powder

#### Gelling

Firm comparable with 1.5% Agar gel.

**Colour and Clarity of prepared medium**

Orange red coloured clear to slightly opalescent gel forms in Petri plates.

**Reaction**

Reaction of 4.65% w/v aqueous solution at 25°C. pH : 7.1±0.2

**pH**

6.90-7.30

**Cultural Response**

M1582: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony
<i>Escherichia coli</i> ATCC 25922	50-100	luxuriant	≥50%	pink to red with bile precipitate
<i>Enterobacter aerogenes</i> ATCC 13048	50-100	luxuriant	≥50%	pink to red
<i>Enterococcus faecalis</i> ATCC 29212	50-100	fair to good	30-40%	colourless to pink
<i>Proteus vulgaris</i> ATCC 13315	50-100	luxuriant	≥50%	colourless
<i>Salmonella Paratyphi A</i> ATCC 9150	50-100	luxuriant	≥50%	colourless
<i>Shigella flexneri</i> ATCC 12022	50-100	fair to good	30-40%	colourless
<i>Salmonella Paratyphi B</i> ATCC 8759	50-100	luxuriant	≥50%	colourless
<i>Salmonella Enteritidis</i> ATCC 13076	50-100	luxuriant	≥50%	colourless
<i>Salmonella Typhi</i> ATCC 6539	50-100	luxuriant	≥50%	colourless
<i>Staphylococcus aureus</i> ATCC 25923	≥10 <sup>3</sup>	inhibited	0%	

**Storage and Shelf Life**

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 A., 1900, The Lancet, II:20.
2. MacConkey A., 1905, J. Hyg., 5:333.
3. Speck M. L., (Ed.), 1985, Compendium of Methods for the Microbiological Examination of Foods, 2nd Ed., APHA, Washington, D.C.
4. Greenberg A. E., Clesceri L. S. and Eaton A. D., (Eds.), 1992, Standard Methods for the Examination of Water and Wastewater, 18th ed., APHA, Washington, D.C.
5. Marshall R., (Ed.), 1992, Standard Methods for the Examination of Dairy Products, 16th Ed., APHA, Washington, D.C.
6. Cruickshank R. Duguid J. P., Marmion B. P., Swain R. H. A., (Eds.), 1975, Medical Microbiology, 12th Ed., Vol. II, Churchill Livingstone, Edinburgh, London.

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