

MacConkey Agar w/o CV, NaCl w/ 0.5% Bile Salts, Granulated

GM082A

MacConkey Agar w/o CV, NaCl w/ 0.5% Bile Salts, granulated is recommended for the cultivation and differentiation of enteric bacteria, restricting swarming of *Proteus* species from specimens such as urine, which may contain large number of *Proteus* species as well as potentially pathogenic gram-positive organisms.

Composition**

Ingredients	Gms / Litre
Peptic digest of animal tissue	20.000
Lactose	10.000
Bile salts	5.000
Neutral red	0.075
Agar	12.000
Final pH (at 25°C)	7.4±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 47.07 grams of medium in 1000 ml distilled water. Heat to boiling with gentle swirling to dissolve the agar 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.

Note: For the cultivation of *Vibrio* species, add 5 grams per litre of Sodium chloride before sterilization.

Principle And Interpretation

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 was 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) and pharmaceutical preparations (6). The original medium contains protein, bile salts, sodium chloride and two dyes. MacConkey Agar w/o CV, NaCl W/ 0.5% Bile Salts is a modification of the original formulation with the exception of crystal violet and sodium chloride. This medium prevents the swarming of *Proteus* species that are generally encountered in pathological specimens. Also potentially pathogenic gram-positive organisms can be isolated using this medium.

The selective action of this medium is attributed to 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 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 1.2% Agar gel.

Colour and Clarity of prepared medium

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

Reaction

Reaction of 4.7% w/v aqueous solution at 25°C. pH : 7.4±0.2

pH

7.20-7.60

Cultural Response

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

Organism	Inoculum (CFU)	Growth	Recovery	Colour of Colony
Cultural Response				
<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%	pale pink to red
<i>Proteus vulgaris</i> ATCC 13315	50-100	Luxuriant	≥50%	colourless
<i>Salmonella</i> Paratyphi A ATCC 9150	50-100	Luxuriant	≥50%	colourless
<i>Shigella flexneri</i> ATCC 12022	50-100	Fair to good	30-40%	colourless
<i>Salmonella</i> Paratyphi B ATCC 8759	50-100	Luxuriant	≥50%	colourless
<i>Salmonella</i> Enteritidis ATCC 13076	50-100	Luxuriant	≥50%	colourless
<i>Salmonella</i> Typhi ATCC 6539	50-100	Luxuriant	≥50%	colourless
<i>Staphylococcus aureus</i> ATCC 25923	50-100	fair-good	30-40%	pale pink-red

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, 1900, The Lancet, ii:20.
2. MacConkey, 1905, J. Hyg., 5:333.
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. Rice E.W., Baird R.B., Eaton A. D., and Clesceri L. S. (Eds.), 2012, Standard Methods for the Examination of Water and Wastewater, 22nd 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, 2014, The United States Pharmacopoeial Convention, Rockville, MD.

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