



Technical Data

MacConkey Agar w/ CV, NaCl and 0.15% Bile salts

M081A

Intended Use:

To identify *Enterobacteriaceae* in the presence of coliforms and lactose non fermenters from water, sewage, food products and clinical samples.

Composition**

Ingredients	g / L
Peptone	20.000
Lactose	10.000
Sodium chloride	5.000
Bile salts	1.500
Neutral red	0.050
Crystal violet	0.001
Agar	15.000
Final pH (at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 51.55 grams 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. Avoid overheating. Cool to 45-50°C. Mix well and pour 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 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). Original medium contains protein, bile salts, sodium chloride and two dyes.

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 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.

Peptone serves as a source of carbon, nitrogen, long chain amino acids, vitamins and other essential growth nutrients. Lactose serves as the carbon source by being the fermentable carbohydrate. Bile salts and crystal violet serves to make the medium selective by inhibiting accompanying gram-positive bacteria. Neutral red is the pH indicator dye while sodium chloride maintains the osmotic equilibrium of the medium.

Type of specimen

Clinical samples: faeces, urine and other pathological material; Food and dairy samples; water samples.

Specimen Collection and Handling

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (3,4,5).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (6).

After use, contaminated materials must be sterilized by autoclaving before discarding.

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (7,8).

Warning and Precautions

In Vitro diagnostic use. For professional use only. Read the label before opening the container. Wear protective gloves/ protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling clinical specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Individual organisms differ in their growth requirement and may show variable growth patterns on the medium.
2. Each lot of the medium has been tested for the organisms specified on the COA. It is recommended to users to validate the medium for any specific microorganism other than mentioned in the COA based on the user's unique requirement.
3. Though the medium is recommended for selective isolation, further biochemical and serological testing must be carried out for complete identification.
4. The surface of the medium should be dry when inoculated.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Light yellow to pink homogeneous free flowing powder

Gelling

Firm comparable with 1.5% Agar gel.

Colour and Clarity of prepared medium

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

pH

pH of 5.15% w/v aqueous solution at 25°C pH 7.00-7.40

Cultural Response

Cultural response was observed after an incubation at 35-37°C for 18-24 hours. Recovery rate is considered as 100% for bacteria growth on Soybean Casein Digest Agar.

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony
<i>Escherichia coli</i> ATCC 8739 (00012*)	50 -100	luxuriant	≥50 %	pink-red with bile precipitate
<i>Escherichia coli</i> ATCC 25922 (00013*)	50 -100	luxuriant	≥50 %	pink-red with bile precipitate
# <i>Klebsiella aerogenes</i> ATCC 13048 (00175*)	50 -100	luxuriant	≥50 %	pink-red with bile precipitate
<i>Enterococcus faecalis</i> ATCC 29212 (00087*)	50 -100	none-poor	0 -10 %	colourless to pale pink
<i>Salmonella</i> Typhimurium ATCC 14028 (00031*)	50 -100	luxuriant	≥50 %	colourless
<i>Staphylococcus aureus</i> subsp. <i>aureus</i> ATCC 6538 (00032*)	≥10 ⁴	inhibited	0 %	
<i>Staphylococcus aureus</i> subsp. <i>aureus</i> ATCC 25923 (00034*)	≥10 ⁴	inhibited	0 %	
<i>Salmonella</i> Enteritidis ATCC 3076 (00030*)	50 -100	luxuriant	≥50 %	colourless
<i>Salmonella</i> Paratyphi A ATCC 9150	50 -100	luxuriant	≥50 %	colourless
<i>Salmonella</i> Paratyphi B ATCC 8759	50 -100	luxuriant	≥50 %	colourless

<i>Salmonella</i> Typhi ATCC 6539	50 -100	luxuriant	≥50 %	colourless
<i>Salmonella</i> Abony NCTC 6017 (00029*)	50 -100	luxuriant	≥50 %	colourless
## <i>Proteus</i> <i>hauseri</i> ATCC 13315	50 -100	luxuriant	≥50 %	colourless
<i>Shigella</i> <i>flexneri</i> ATCC 12022 (00126*)	50 -100	fair to good	30 -40 %	colourless
<i>Staphylococcus</i> <i>epidermidis</i> ATCC 12228 (00036*)	≥10 ⁴	inhibited	0 %	
<i>Corynebacterium</i> <i>diphtheriae</i> type <i>gravis</i>	≥10 ⁴	inhibited	0 %	

Key : (*) Corresponding WDCM numbers

(##) Formerly known as *Proteus vulgaris* (#) Formerly known as *Enterobacter aerogenes*

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (7,8).

Reference

1. MacConkey, 1900, The Lancet, ii:20.
2. MacConkey, 1905, J. Hyg., 5:333.
3. Salfinger Y., and Tortorello M.L., 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
4. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C. 7.
5. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D.C.
6. Lipps WC, Braun-Howland EB, Baxter TE, eds. Standard methods for the Examination of Water and Wastewater, 24th ed. Washington DC:APHA Press; 2023.
7. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
8. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

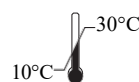
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