



PRODUCT SPECIFICATION

MACCONKEY AGAR

Without Crystal Violet and with 0.15% Bile Salts

CMB0166S, CMB0166M

Food/Dairy/Clinical/Water Analysis

Dehydration Culture Medium

INTENDED USE

Recommended media for the isolation and differentiation of lactose-fermenting and lactose-non-fermenting Gram-negative enteric bacilli from pharmaceutical, food, dairy, water samples, and Clinical and non-clinical samples.

COMPONENT DESCRIPTION

MacConkey agar comprises several key ingredients, including peptone, proteose peptone, lactose, bile salts, sodium chloride, neutral red, and agar. Peptone and protease peptone serve as nitrogen and other essential nutrients that promote microbial growth. Lactose acts as a carbon source and is crucial for the selective identification of lactose-fermenting microorganisms. Bile salts function as selective agents that inhibit the growth of gram-positive organisms. Neutral red serves as a pH indicator dye. Agar functions as a solidifying agent. Upon fermentation of lactose, acid is produced, leading to bile salt precipitation. Colonies of lactose-fermenting coliform bacteria exhibit a pink coloration, often accompanied by a zone of precipitation around them, whereas non-lactose fermenting bacteria remain colourless.

PRINCIPLE

The key reference for MacConkey Agar development was first published in 1900 by A.T. MacConkey¹. The MacConkey agar is used for the isolation of gram-negative enteric bacteria and the differentiation of lactose fermenting from lactose non-fermenting gram-negative bacteria². This media is for the isolation and differentiation of lactose-fermenting and lactose-non-fermenting Gram-negative enteric bacilli from, clinical⁵, and non-clinical samples, as well as from food, dairy^{3,6}, and water samples^{4,7,8}. MacConkey Agar without Crystal Violet (w/o CV) is a selective and differential culture medium designed for the isolation and differentiation of Gram-negative bacteria, specifically Enterobacteriaceae, from clinical, environmental, and food samples. The absence of crystal violet reduces inhibition of Gram-positive bacteria, supporting broader bacterial growth, while 0.15% bile salts and NaCl enhance selectivity for Gram-negative organisms.

TYPICAL FORMULA*

Components	Gram/ Litre (g/l)
Peptone	17.0 g
Proteose Peptone	3.0 g
Bile Salt	1.5 g
Sodium Chloride	5.0g
Neutral Red	0.03g
Agar	15.0 g
pH	7.1 ± 0.2 at 25°C

*Formula adjusted to meet formulation parameters

PREPARATION

Add 51.53g of media to the 1000ml of purified/distilled water. To dissolve the media in purified water, heat the media and boil it for a minute with frequent mixing. Sterilize at 121°C for 15 minutes by autoclaving (for Sterilization use validated cycles for autoclaving). Ensure the media should be poured into a sterile Petri dish at a temperature between 45°C to 50°C.

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Page 1 | 3

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QUALITY CONTROL

Physical Appearance

Dehydrated Culture Media (DCM) Appearance: Light pink coloured, free-flowing powder, hygroscopic in nature, and free from any extraneous material.

Solubility: Soluble in Purified water or distilled or de-ionized water upon boiling, orange-red coloured, opalescent solution

Gel Strength: Firm comparable with 1.5% agar gel

Prepared Media Appearance: Orange-red coloured, opalescent

pH: 7.1 ± 0.2 at 25°C

Microbiological Performance

Growth Promotion Test (GPT)

MacConkey Agar (CMB0166M) was utilized as the test medium, while Soybean Casein Digest Agar (SCDA) served as the reference (control) medium. Both media were prepared according to the product specifications. The recovery criteria for the test medium have been established at greater than 70% compared to the reference (control) medium.

QC Organisms	Control Inoculum (in CFU)	Incubation Temp. (°C)	Incubation Duration (in hours)	Growth Observed
<i>Escherichia coli</i> ATCC 25922	≤100CFU	35°C to 37°C	24 hrs. to 48 hrs.	Compliant
<i>Salmonella typhimurium</i> ATCC 14028	≤100CFU	35°C to 37°C	24 hrs. to 48 hrs.	Compliant
<i>Salmonella enteritidis</i> ATCC 13076	≤100CFU	35°C to 37°C	24 hrs. to 48 hrs.	Compliant
<i>Shigella flexneri</i> ATCC 12022	≤100CFU	35°C to 37°C	24 hrs. to 48 hrs.	Compliant
<i>Staphylococcus aureus</i> ATCC 13048	≤100CFU	35°C to 37°C	24 hrs. to 48 hrs.	Compliant
<i>Klebsiella aerogenes</i> ATCC 27853	≤100CFU	35°C to 37°C	24 hrs. to 48 hrs.	Compliant

ATCC is a registered trademark for the American Type Culture Collection.

STORAGE

Keep the container in a cool and dry place away from direct sunlight exposure, and store the product between 10°C to 30°C in its original packaging (for dehydrated Culture Media) and for prepared media store at 20°C to 25°C. Unopened containers should be stored at the recommended temperature for the entire shelf life. For product shelf-life, please refer product label. Once the seal of the container is opened, ensure the container cap is firmly tight before storing it in a cool and dry place as per the product label. Also, prevent long exposure to air contact as this may lead to lump formation and compromise product quality.

REQUIREMENTS

Material required and provided: Dehydrated Culture Media, MacConkey Agar (CMB0166M)

Material required and NOT Provided: Biosafety cabinet, vortex, incubator, Sterile IPA, Lint-free Mops, Micropipettes, Sterile Tip box, Petri plates, Reference Culture Standard/Quality Control organisms, Loops, Marker for labeling.

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PRECAUTIONS & WARNINGS

- Use limited to in-vitro diagnostics only
- For trained professional use only
- Please do not use expired products
- Do not use the product if visible signs of deterioration or contamination
- Always perform Growth Promotion Test (GPT) for each new lot received to stay compliant as per harmonized pharmacopeia requirement

DISPOSAL

Dispose of the used or unused product in accordance with local regulations.

REFERENCES

1. MacConkey, A.T. (1900), *A note on a new medium for the growth and differentiation of the bacillus coli communis and the bacillus typhi abdominalis*. The Lancet Journal, Vol. ii: p-20
2. MacConkey, A.T. (1905), *Lactose-fermenting bacteria in faeces*. Journal of Hygiene, Vol. 5, No. 3; p-333-378.
3. Wehr H. M. and Frank J. H. (2004), *Standard Methods for the Microbiological Examination of Dairy Products*, 17th Ed., APHA Inc., Washington, D.C.
4. ISO 9308-1:2014. (2014), *Water quality: Enumeration of Escherichia coli and Coliform bacteria- Part-1: Membrane Filtration Method*.
5. Jorgensen JH, Pfaller MA, Carroll KC, Funke G, Landry ML, Richter SS, Warnock DW (ed), (2015), *Manual of clinical microbiology*, 11th ed, vol 1 ASM Press, Washington, DC.
6. FDA. (2005), *Bacteriological Analytical Manual*, 18th Ed., AOAC, Washington, DC.
7. US Food and Drug Administration (FDA). (2020), *Enumeration of E. coli and the Coliform bacteria*, Chapter 4; Bacteriological Analytical Manual (BAM).
8. American Public Health Association (APHA). (2023), *Standard Methods for the Examination of water and wastewater*, 24th Ed.

DISCLAIMER

Users are encouraged to assess the product's suitability for their specific applications. While we strive for accuracy in the information provided, please note that it is subject to change. Unless specified otherwise, these products are intended solely for laboratory, diagnostic, research, or manufacturing use and are not designed for human, animal, or therapeutic use. This document is not to be interpreted as a warranty, and we kindly disclaim any liability related to potential patent infringements.

Consult MicroBio holds the capability to adjust specifications and information regarding our products when necessary, allowing us to continuously enhance the quality and relevance of what we offer.

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Page 3 | 3

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