MacConkey HiVeg® Agar w/o CV w/ 0.003% NR and 1.5% Agar

MV008

Intended use:

Recommended for selective isolation and differentiation of lactose fermenting and lactose non fermenting enteric bacteria.

Composition**

Ingredients	g / L
HiVeg® peptone	17.000
HiVeg® peptone No. 3	3.000
Lactose	10.000
Synthetic detergent	1.500
Sodium chloride	5.000
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 51.53 gms of medium in 1000 ml purified/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. 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 (4). MacConkey HiVeg® Agar w/o CV w/ 0.003% NR and 1.5% Agar is prepared by completely replacing animal based peptone with vegetable peptones to avoid BSE/TSE risks associate with animal peptones.

HiVeg® peptone and HiVeg® peptone No. 3 serves as a source of carbon, nitrogen, long chain amino acids and other essential growth nutrients. The selective action of this medium is attributed to synthetic detergents, which is 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 colonies. 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.

Type of specimen

Food and dairy sample; Water samples

Specimen Collection and Handling:

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

Warning and Precautions

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 specimens. Safety guidelines may be referred in individual safety data sheets

Limitations

1. Although this medium is selective for gram negative organisms, biochemical identification and serological testing using pure cultures is recommended for complete identification.
2. It is advised to incubate for recommended period and temperature to avoid misintepretation of results.
3. It is advised to read the results immediately after icubation, as overgrowth of *Proteus* species may mask other colonies.

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 temmperature.

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

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

Reaction

Reaction of 5.15% w/v aqueous solution at 25°C. pH : 7.1±0.2

pH

6.90-7.30

Cultural Response

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

Organism	Inoculum (CFU)	Growth	Recovery	Colour of Colony
Escherichia coli ATCC 25922 (00013*)	50-100	luxuriant	>=50%	pink to red with bile precipitate
# *Klebsiella aerogenes* ATCC 13048 (00175*)	50-100	luxuriant	>=50%	pink to red
Enterococcus faecalis ATCC 29212 (00087*)	50-100	none-poor	<=10%	pale pink to red
## *Proteus hauseri* ATCC 13315	50-100	luxuriant	>=50%	colourless
Salmonella Paratyphi A ATCC 9150	50-100	luxuriant	>=50%	colourless
Shigella flexneri ATCC 12022 (00126*)	50-100	fair to good	30-40%	colourless
Salmonella Paratyphi B ATCC 8759	50-100	luxuriant	>=50%	colourless
Salmonella Enteritidis ATCC 13076 (00030*)	50-100	luxuriant	>=50%	colourless
Salmonella Typhi ATCC 6539	50-100	luxuriant	>=50%	colourless
Staphylococcus aureus subsp. *aureus* ATCC 25923 (00034*)	$>=10^4$	inhibited	0%	
Corynebacterium diphtheriae type gravis	$>=10^4$	inhibited	0%	
Staphylococcus epidermidis ATCC 12228 (00036*)	$>=10^4$	inhibited	0%	

Key : * Corresponding WDCM numbers.

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

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder 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 sample must be decontaminated and disposed of in accordance with current laboratory techniques (3,7).

Reference

1. MacConkey, 1900, The Lancet, ii:20.
2. MacConkey, 1905, J. Hyg., 5:333.
3. 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.
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.
5. 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.
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.

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