MacConkey Medium

MF012/E*

Intended Use:

Recommended for detection and enumeration of coliforms.

Directions

The test sample should be filtered through a sterile membrane filter having pore size of 0.22µ / 0.45µ. Rehydrate the nutrient pad with 2.0-2.5 ml sterile distilled / purified water. After filtration, remove the membrane filter aseptically using sterile forceps. Place the membrane filter on rehydrated nutrient pad. Incubate the inoculated nutrient. Interpret the results qualitatively by observing the presence or absence of growth and quantitatively by counting the number of colonies on the surface of the membrane filter and calculating CFU/ml.

Principle And Interpretation

DriFilter Membrane Nutrient Pad Medium is ready to use sterile culture media in the form of a 50 mm biological inert absorbent pads impregnated with Plate Count medium, then dried and sterilized in 55 mm Petri plate. They eliminate the need of laborious media preparation and autoclaving procedures. The nutrient pads are to be just rewetted with sterile distilled water and are ready to use. Use of nutrient pads allows larger sample volumes to be tested at a time. Interpretation of results is directly by counting the CFUs and also quantifies the microbial load present in the sample.

MacConkey agars are slightly selective and differential plating media mainly used for the detection and isolation of gram-negative organisms from clinical (10), dairy (14), food (5,11), water (1), pharmaceutical (3,12) and industrial sources (15). It is also recommended for the selection and recovery of the *Enterobacteriaceae* and related enteric gram-negative bacilli. USP recommends this medium for use in the performance of Microbial Limit Tests (12). These agar media are selective since the concentration of bile salts, which inhibit gram-positive microorganisms, is low in comparison with other enteric plating media. The medium M081, which corresponds with, that recommended by APHA can be used for the direct plating of water samples for coliform bacilli, for the examination of food samples for food poisoning organisms (11) and for the isolation of *Salmonella* and *Shigella* species in cheese (14). Other than that this medium is also used for count of coli-aerogenes bacteria in cattle and sheep faeces (9), the count of coli-aerogenes and non-lactose fermenters in poultry carcasses (9), bacterial counts on irradiated canned minced chicken (13) and the recognition of coli-aerogenes bacteria during investigations on the genus Aeromonas (4).

MacConkey Agar is the earliest selective and differential medium for cultivation of enteric microorganisms from a variety of clinical specimens (7,8). The 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 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, transparent and typically do not alter appearance of the medium.

Peptone, Tryptone and gelatin peptone are sources of nitrogen, carbon, long chain amino acids and other nutrients. Lactose is a fermentable carbohydrate,Sodium chloride maintains the osmotic equilibrium. Bile salts and crystal violet are selective agents that inhibit growth of gram-positive organisms. Neutral red is the pH indicator dye.

Type of specimen

Foodstuffs and dairy samples, water samples, pharmaceutical samples.

Specimen Collection and Handling

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (5,11,14.).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards.(1)

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per guidelines.(3,12)

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. Though the medium is recommended for selective isolation, further biochemical and serological testing must be carried out for further confirmation.
2.The surface of the medium should be dry when inoculated.

Performance and Evaluation

Performace 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
Dry filter membrane pad of 50 mm diamete

Colour
Light pink coloured nutrient pad

Sterility test
Passes release criteria

Cultural Response
Cultural characteristics observed after incubation at 35-37ºC for 18-24 hours

Organism	Inoculum (CFU)	Growth	Colour of colony
Escherichia coli ATCC 25922 (00013*)	50 -100	luxuriant	Pink
S. choleraesuis ATCC 12011	50 -100	luxuriant	Colourless

Key :- * Corresponding WDCM numbers

Storage and Shelf Life

On receipt store between 10-30°C. Use before expiry date on the label.
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 (6,10).

Reference

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3. British Pharmacopoeia, 2016, The Stationery office British Pharmacopoeia.
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6. Isenberg, H.D. Clinical Microbiology Procedures Handb0ook. 2nd Edition
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13. Thornley Margaret J., 1957, J. Appl. Bacteriol., 20(2), 273-285.
14. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
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Note:

MF000 - Sterile pad packed individually in sterile Petri plate without Membrane Filter

MF000E* - Sterile pad packed individually in sterile disposable plastic bag without Membrane Filter