Mannitol Salt Broth

M383

Intended Use

For selective isolation of presumptive pathogenic Staphylococci.

Composition**

Ingredients	g / L
Proteose peptone	10.000
HM peptone B #	1.000
Sodium chloride	75.000
D-Mannitol	10.000
Phenol red	0.025
Final pH (at 25°C)	7.4±0.2

**Formula adjusted, standardized to suit performance parameters
Equivalent to Beef extract

Directions

Suspend 96.02 grams in 1000 ml purified/distilled water. Heat if necessary to dissolve the medium completely. Dispense in tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. ***Note:****This product contains 7.5% sodium chloride as one of its ingredients. On repeated exposure to air and absorption of moisture sodium chloride has tendency to form lumps, therefore we strongly recommend storage in tightly closed containers in dry place away from bright light.*

Principle And Interpretation

Mannitol Salt Broth is prepared as suggested by Chapman (1) and is used for the selective isolation of pathogenic Staphylococci. This medium is recommended for the detection and enumeration of coagulase-positive Staphylococci in milk (2) food (3) and other specimens. Mannitol Salt Broth is used for the isolation of presumptive pathogenic staphylococci. Pathogenic staphylococci ferment mannitol and produce a yellow coloured medium.

The medium contains HM peptone B and proteose peptone which makes it very nutritious as they provide essential growth factors and trace nutrients. Many other bacteria except Staphylococci are inhibited by 7.5% sodium chloride. Mannitol is the fermentable carbohydrate source. The differential action of the medium is attributed to D-Mannitol. *Staphylococcus aureus* ferments mannitol to produce yellow coloured medium. Most coagulase-negative species of Staphylococci and Micrococci do not ferment mannitol and therefore the medium remains red in colour. The colour of the medium is due to the reactivity of phenol red to the pH of the medium; phenol red is red at pH 8.4 and yellow at 6.8. Presumptive *Staphylococcus* showing yellow coloured medium should be further tested for production of coagulase.

A possible *S. aureus* must be confirmed by the coagulase test. Also the organism should be subcultured to a less inhibitory medium not containing excess salt to avoid the possible interference of salt with coagulase testing or other diagnostic tests (e.g. Nutrient Broth) (M002) (4). Few strains of *S. aureus* may exhibit delayed mannitol fermentation. Negative results should therefore be re-incubated for an additional 24 hours before being discarded (4).

Type of specimen

Clinical samples: pus, urine; Food and dairy samples, water samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (5,6).

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (7,8,9). After use, contaminated materials must be sterilized by autoclaving before discarding.

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. A possible *S.aureus* must be confirmed by the coagulase test.

2.The organism should be subcultured to a less inhibitory medium not containing excess salt to avoid the possible interference of salt with coagulase testing or other diagnostic tests (e.g. Nutrient Broth) (M002) (4).

3.Few strains of *S.aureus* may exhibit delayed mannitol fermentation. Negative results should therefore be re-incubated for an additional 24 hours before being discarded (4).

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

Colour and Clarity of prepared medium

Red coloured clear solution in tubes

Reaction

Reaction of 9.6% 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 18-48 hours.

Organism	Inoculum (CFU)	Growth	Colour of medium
Escherichia coli ATCC 25922 (00013*)	>=10^4	Inhibited	
Staphylococcus aureus subsp. aureus ATCC 25923 (00034*)	50-100	good-luxuriant	yellow
Staphylococcus Epidermidis ATCC 12228 (00036*)	50-100	fair-good	red

Key : (*) Corresponding WDCM numbers.

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-30°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 clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (5,6).

Reference

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2. Marshall R. (Ed.), 1992, Standard Methods for the Examination of Dairy Products, 16th ed., APHA, Washington, D.C.
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4. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore
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6. 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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IVD ***In vitro* diagnostic medical device**

30°C
10°C **Storage temperature**

EC REP

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CE Marking

Do not use if package is damaged

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