Coagulase Mannitol Broth Base

M277

Intended use

Coagulase Mannitol Broth Base is recommended for the simultaneous detection of mannitol fermentation and coagulase production during differentiation of Staphylococci from clinical and non-clinical samples.

Composition**

Ingredients	Gms / Litre
HMH infusion from #	375.000
Peptone	10.000
D-Mannitol	10.000
Sodium chloride	5.000
Phenol red	0.025
Final pH (at 25°C)	7.4±0.2

**Formula adjusted, standardized to suit performance parameters

Equivalent to Heart muscle infusion from

Directions

Suspend 35.0 grams in 1000 ml distilled water. Heat, if necessary, to dissolve the medium completely. Sterilize by autoclaving at 118-121°C for 15 minutes. Cool to 45-50°C. Just before use, add 7-15% v/v sterile, pre-tested, rabbit plasma (coagulase plasma) to the basal medium. Mix well and dispense into sterile tubes

Principle And Interpretation

Coagulase Mannitol Broth Base is used for the isolation of *Staphylococcus aureus* from clinical specimens and for differentiation of *Staphylococcus aureus* from other species on the basis of coagulase production and mannitol fermentation. Chapman for the first time introduced medium for selective isolation and differentiation of Staphylococci (1). Tellurite-glycine media were designed by Zebovitz et al (2) and Marwin (3) for selectively isolating coagulase positive Staphylococcal species. Present medium is based on Esber and Faulconer formulation (4).

Coagulase Mannitol Broth Base simultaneously detects mannitol fermentation and coagulase production by Staphylococci (5). Coagulase Mannitol Broth Base is a good substrate for Staphylococci as well as other fastidious bacteria. Coagulase production and mannitol fermentation observed in Coagulase Mannitol Broth Base is presumptive identification of pathogenic Staphylococci (6). Coagulase production is dependent on the presence of a fermentable sugar like mannitol in this case. It is also dependent on the presence of a protein factor in the brain heart infusion and blood plasma (4). When mannitol is fermented, the pH of the medium drops. This drop in pH is indicated by the change in colour of the phenol red, which turns yellow, and exhibit yellow medium. An opaque broth due to coagulated plasma forms due to growth of coagulase positive organisms.

Staphylococcus epidermidis a coagulase negative and mannitol nonfermenting species, does not change the colour of the medium. Coagulase negative species may ferment mannitol and produce a yellow colour but opacity will not be formed. Production of gas can be determined by placing a small inverted Durhams tube in the medium tube. Mutant or old cultures of Staphylococcus aureus may be weak coagulase producers. They should be freshly subcultured and rechecked. Escherichia coli ferments mannitol and may be weakly coagulase positive.

For the test, use this medium in 2 to 5 ml amounts after adding about 12-15% plasma. Inoculate by adding about 2 drops of test organism and incubate at 37°C and examine after 2-3 hours and also after 4-5 hours of incubation. Un-inoculated control tubes should also be run in parallel with the fermentation tests.

Type of specimen

Clinical samples - Blood, pus ; Food and dairy samples

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (4,5).
For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (1,2,8).

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

Warning and Precautions :

In Vitro diagnostic 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. Saftey guidelines may be referred in individual safety data sheets

Limitations :

This medium is general purpose medium and may not support the growth of fastidious organisms.

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 light pink homogeneous free flowing powder

Colour and Clarity of prepared medium

Red coloured, clear to slightly opalescent solution in tubes

Reaction

Reaction of 3.5% w/v aqueous solution at 25°C. pH : 7.4±0.2

pH

7.20-7.60

Cultural Response

M277: Cultural characteristics observed with added 7-15% v/v sterile pretested coagulase plasma after an incubation at 35-37°C for 18-48 hours.

Organism	Inoculum (CFU)	Growth	Acid from Mannitol	Coagulase activity
Cultural Response				
Staphylococcus aureus subsp. *aureus ATCC 25923 (00034*)*	50-100	luxuriant	positive reaction, yellow colour	positive reaction,clot formation
Staphylococcus epidermidis ATCC 12228	50-100	luxuriant	negative reaction, no colour change	negative reaction,no clot formation

Key : *Corresponding WDCM numbers.

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. Use before expiry date on the label.
Product performance is best if used within stated expiry period.

Disposal

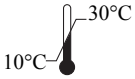
User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance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

Reference

1. Chapman,1946, J. Bact., 51:409.
2. Zebovitz, Evans and Nivens, 1955, J. Bact., 70:686.
3. Marwin, 1958, Am. J. Clin. Pathol., 30:470.
4.Esber and Faulconer, 1959, Am. J. Clin. Pathol., 32:192.

5.Schaub and Merrit, 1960, Bull. Johns Hopkins Hosp., 106:25.
6.Mincheu and Cluff, 1961, J. Chron. Dis., 13:354.
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.

Revision : 02 / 2018

IVD	In vitro diagnostic medical device
CE	CE Marking
	Storage temperature
	Do not use if package is damaged
	HiMedia Laboratories Pvt. Limited, 23 Vadhani Industrial Estate, LBS Marg,Mumbai-86,MS,India
EC REP	CE Partner 4U ,Esdoornlaan 13, 3951 DB Maarn The Netherlands, www.cepartner 4u.eu