

Technical Information

Coagulase Mannitol Broth Base

Product Code: DM 1277

Application: - Coagulase Mannitol Broth Base is recommended for the simultaneous detection of mannitol fermentation and coagulase production during differentiation of Staphylococci.

Composition**

Ingredients	Gms / Litre
Heart muscle, infusion from	10.00
Peptic digest of animal tissue	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

Principle & 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 formulated by Zebovitz et al (2) and Marwin (3) for the selective isolation of coagulase positive Staphylococcal species. Present medium is based on Esber and Faulconer formulation (4).

Coagulase Mannitol Broth Base simultaneously indicates 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.

Methodology

Suspend 35 grams of dehydrated media powder in 1000 ml distilled water. Mix thoroughly & heat, if necessary, to dissolve the medium completely. Sterilize by autoclaving at 118-121°C for 15 minutes. Cool to 45-50°C. Aspectically add 7-15% v/v sterile, pre-tested, rabbit plasma (coagulase plasma) to the basal medium. Mix well before dispensing into sterile tubes.



Dehydrated Culture Media
Bases / Media Supplements

Quality Control

Appearance

Light yellow to light pink homogeneous free flowing powder

Colour and Clarity

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 Range

7.20-7.60

Cultural Response

DM 1277: 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 <i>Staphylococcus aureus</i> ATCC 25923	50-100	luxuriant	positive reaction, Yellow colour	positive reaction,clot formation
<i>Staphylococcus epidermidis</i> ATCC 12228	50-100	luxuriant	negative reaction, no colour change	negative reaction,clot formation

Storage and Shelf Life

Dried Media: Store below 30°C in tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label.

Prepared Media: 2-8° in sealable plastic bags for 2-5 days.

Further Reading

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.

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