

TM 070 – COAGULASE MANNITOL BROTH BASE

INTENDED USE

For detection of coagulase production and mannitol fermentation in differentiation of Staphylococci.

PRODUCT SUMMARY AND EXPLANATION

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. Tellurite-glycine media were designed by Zebovitz et al and Marwin for selectively isolating coagulase positive Staphylococcal species. Present medium is based on Esber and Faulconer formulation.

Coagulase Mannitol Broth Base simultaneously detects mannitol fermentation and coagulase production by Staphylococci. 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. 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 heart muscle infusion and blood plasma. 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 non-fermenting 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 Durham's 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.

COMPOSITION

Ingredients	Gms / Ltr
Heart muscle infusion from	375.000
Peptone	10.000
D-Mannitol	10.000
Sodium chloride	5.000
Phenol red	0.025

PRINCIPLE

Beef heart infusion and peptone provides nutrients to the media. Mannitol acts as a fermentable sugar. Sodium chloride helps in maintaining equilibrium. Agar acts as a solidifying agent.

INSTRUCTION FOR USE

- Dissolve 35.0 grams in 1000 ml purified/ 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













QUALITY CONTROL SPECIFICATIONS

Appearance of Powder: Light yellow to light pink homogeneous free flowing powder.Appearance of prepared medium: Red coloured, clear to slightly opalescent solution in tubes.

pH (at 25°C) : 7.4±0.2

INTERPRETATION

Cultural characteristics observed after incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Acid from Mannitol	Coagulase activity	Incubation Temperature	Incubation Period
Staphylococcus aureus subsp. aureus	25923	50-100	Luxuriant	Positive reaction, yellow colour	Positive reaction, clot formation	35-37°C	18-48 Hours
Staphylococcus epidermidis	12228	50-100	Luxuriant	Negative reaction, no colour change	Negative reaction, no clot formation	35-37°C	18-48 Hours

PACKAGING:

In pack size of 100 m and 500 gm bottles.

STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

REFERENCES

- 1. Chapman, 1946, J. Bact., 51:409.
- 2. Esber and Faulconer, 1959, Am. J. Clin. Pathol., 32:192.
- 3. Marwin, 1958, Am. J. Clin. Pathol., 30:470
- 4. Mincheu and Cluff, 1961, J. Chron. Dis., 13:354.
- 5. Schaub and Merrit, 1960, Bull. Johns Hopkins Hosp., 106:25.
- 6. Zebovitz, Evans and Nivens, 1955, J. Bact., 70:686.





































NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices. *For Lab Use Only Revision: 08 Nov., 2019







