M-Bismuth Sulphite Broth M1101

Intended Use:

Recommended for selective detection of Salmonellae by the membrane filtration technique.

Composition**

Ingredients	Gms / Litre
Peptone	20.000
HM peptone B #	10.000
Dextrose (Glucose)	10.000
Disodium hydrogen phosphate	8.000
Ferrous sulphate	0.600
Bismuth sulphite indicator	16.000
Brilliant green	0.050
Final pH (at 25°C)	7.7±0.2

**Formula adjusted, standardized to suit performance parameters

- Equivalent to Beef extract

Directions

Suspend 64.65 grams in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely. Excessive heating destroys the selective properties of the medium. DO NOT AUTOCLAVE. The medium usually contains flocculent precipitate, which should be dispersed evenly by swirling the flask just before use. Cool to 35°C and saturate sterile absorbent cotton pad with 2 ml of the broth. The medium should be used within 24 hours of rehydration.

Principle And Interpretation

Salmonella is a gram-negative, facultatively anaerobic, non-sporulating, non-motile rod in the family *Enterobacteriaceae.* They are widely distributed in animals affecting mainly the stomach and the intestines. These organisms are difficult to differentiate biochemically from *Escherichia coli*. M-Bismuth Sulphite Broth was formulated by Clark et al (3) and is particularly recommended for detection of *Salmonella* Typhi from water and various clinical specimens by the membrane filtration technique. Preliminary enrichment on a non-selective medium is not necessary. M-Bismuth Sulphite Broth has a composition similar to Bismuth Sulphite Agar (M027), except Agar. Also in the broth medium, all the constituents are in double concentration.

Peptone, HM peptone B and dextrose provide nitrogen and carbon compounds, long chain amino acids and essential growth nutrients. Ferrous sulphate and bismuth sulphite indicator together act as H_2S indicators. Brilliant green acts as selective agent. Luxuriant growth of *Salmonella* Typhi is obtained after 30 hours incubation at 35°C but metallic sheen and brown-black halo is not developed before 40 hours. The importance of this medium has been repeatedly mentioned for detection of *Salmonella* Typhi by membrane filter technique (4,5,6,9).

Type of specimen

Food and dairy samples; Water samples

Specimen Collection and Handling

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (1,10,11). For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (2). 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. DO NOT AUTOCLAVE OR OVERHEAT THE MEDIUM, as it destroys the selectivity of the medium.

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 greenish yellow homogeneous free flowing powder

Colour and Clarity of prepared medium
Greenish yellow coloured opalescent solution with flocculent precipitate

Reaction
Reaction of 6.47% w/v aqueous solution at 25°C. pH : 7.7±0.2

pH
7.50-7.90

Cultural Response
Cultural characteristics observed in humid atmosphere, after an incubation at 35-37°C for 40-48 hours.

Organism	Inoculum (CFU)	Growth	Colour of colony (on membrane filter)
Escherichia coli ATCC 25922 (00013*)	50-100	none-poor	brown-green, if any
Salmonella Typhi ATCC 6539	50-100	luxuriant	black with metallic sheen
Salmonella Typhimurium ATCC 14028 (00031*)	50-100	luxuriant	black with metallic sheen
Staphylococcus aureus subsp. *aureus* ATCC 25923 (00034*)	>=10^4	inhibited	

Key : (*) - Corresponding WDCM numbers.

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C,but not for more than 2 days as after which dye oxidizes to give green medium that could be inhibitory to some Salmonellae. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order 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 sample must be decontaminated and disposed of in accordance with current laboratory techniques (7,8).

Reference

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