Bismuth Sulphite Agar Modified

M1004

Bismuth Sulphite Agar Modified is recommended for the selective isolation and preliminary identification of *Salmonella* Typhi and other Salmonellae from pathological materials, sewage, water supplies, food etc.

Composition**

Ingredients	Gms / Litre
Peptic digest of animal tissue	5.000
Beef extract	5.000
Dextrose	5.000
Disodium phosphate	4.000
Ferrous sulphate	0.300
Bismuth sulphite indicator	8.000
Brilliant green	0.016
Agar	12.700
Final pH (at 25°C)	7.6±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 40 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. DO NOT STERILIZE IN AUTOCLAVE or by fractional sterilization since overheating may destroy the selectivity of the medium.

The sensitivity of the medium depends largely upon uniform dispersion of precipitated bismuth sulphite in the final gel, which should be dispersed before pouring into the sterile Petri plates.

Principle And Interpretation

The Salmonellae constitute the most taxonomically complex group of bacteria among *Enterobacteriaceae* (1). Human *Salmonella* infections are most commonly caused by ingestion of food, water or milk contaminated by human or animal excreta. Humans are the only reservoirs of *S*. Typhi (2). Four clinical types of *Salmonella* infections may be distinguished (3) namely gastroenteritis, bacteremia or septicemia, enteric fever and a carrier state. Of the various media employed for the isolation and preliminary identification of Salmonellae, particularly *Salmonella* Typhi; Bismuth Sulphite Agar is the most productive (4).

Bismuth Sulphite Agar, Modified is a modification of the original formulation of Wilson and Blair Medium (5). It is also recommended for the isolation of *Salmonella* Typhi and other Salmonella (6, 7).

S. Typhi, *S*. Enteritidis and *S*. Typhimurium typically grow as black colonies with a surrounding metallic sheen resulting from hydrogen sulphide production and reduction of sulphite to black ferric sulphide. *Salmonella* Paratyphi A grows as light green colonies. Bismuth Sulphite Agar may be inhibitory to some strains of *Salmonella* species and therefore should not be used as the sole selective medium for these organisms. *Shigella* species are mostly inhibited on this medium; exceptions being *S*. flexneri and *S*. sonnei (8) and also some *Salmonella* like *S*. Sendai, *S*. Berta, *S*. Gallinarum, *S*. Abortus-equi are inhibited (8). Also this medium favors use of larger inoculum as compared to other selective media, as it has unique inhibitory action toward gram-positive organisms and coliforms.

Peptic digest of animal tissue and beef extract serve as sources as carbon, nitrogen, vitamins and essential growth factors. Dextrose is the carbon source. Disodium phosphate maintains the osmotic equilibrium. Bismuth sulphite indicator along with brilliant green inhibits the intestinal gram-positive and gram-negative bacteria. Ferrous sulphate aids in detection of hydrogen sulphide production.

Clinical samples can be directly used to inoculate Bismuth Sulphite Agar. In case of food samples, pre enrichment of the sample is done prior to inoculation.

Quality Control

Appearance
Light yellow to greenish yellow homogeneous free flowing powder

Gelling
Firm, comparable with 1.27% Agar gel.

Colour and Clarity of prepared medium
Greenish yellow coloured opalescent with flocculent precipitate forms in Petri plates.

Reaction
Reaction of 4.0% w/v aqueous solution at 25°C. pH : 7.6±0.2

pH
7.40-7.80

Cultural Response
M1004: Cultural characteristics observed after an incubation at 35-37°C for 40-48 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Colour of Colony
Preparation of test strain				
Cultural Response				
Enterobacter aerogenes ATCC 13048	50-100	none-poor	<=10%	brown-green(depends on inoculum density)
Enterococcus faecalis ATCC 29212	>=10^3	inhibited	0%	
Escherichia coli ATCC 25922	50-100	none-poor	<=10%	brown-green(depends on inoculum density)
Salmonella Typhi ATCC 19430	50-100	good-luxuriant	>=50%	black with metallic sheen
Salmonella Paratyphi B ATCC 8759	50-100	good-luxuriant	>=50%	black with metallic sheen
Salmonella Enteritidis ATCC 13076	50-100	good-luxuriant	>=50%	black with metallic sheen
Shigella flexneri ATCC 12022	50-100	none-poor	<=10%	brown
Salmonella Typhimurium ATCC 14028	50-100	good-luxuriant	>=50%	black with metallic sheen

Storage and Shelf Life

Store below 30°C and prepared medium at 2-8°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.

Reference

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