PRODUCT SPECIFICATION SHEET

Bismuth Sulphite Agar, Modified (DM556)

Intended Use

Bismuth Sulphite Agar, Modified (DM556) is recommended for isolation and preliminary identification of *Salmonella* serotype *Typhi* and other *Salmonellae* from pathological specimen, sewage, water supplies, food, etc.

Product Summary and Explanation

Bismuth Sulphite Agar, Modified is a modification of the original formulation of Wilson and Blair Medium.[5] This medium is recommended for the isolation of *Salmonella typhi* and other *Salmonella*.[6, 7] *Salmonellae* constitute the major part of taxonomically complex group of bacteria among *Enterobacteriaceae*.[1] Ingestion of food, water or milk contaminated by human or animal excreta are the most common causes of human *Salmonella* infections. 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. Among the various media employed for the isolation and preliminary identification of *Salmonellae*, particularly *Salmonella typhi*, this medium is considered the most productive.[4] Hydrogen sulphide production and reduction of sulphite to black ferric sulphide results in the formation of metallic sheen surrounding the black colonies of *S. typhi, S. enteritidis* and *S. typhimurium* typically grow as black colonies. *Salmonella paratyphi A* grows as light green colonies. Bismuth Sulphite Agar should not be used as the sole selective medium for these organisms, as it may be inhibitory to some strains of *Salmonella* species. *Shigella* species are mostly inhibited on this medium; *S. flexneri* and *S. sonnei* being exceptions[8] and also some *Salmonella* like *S. sendai, S. berta, S. gallinarum, S. abortus-equi* are inhibited.[8] This medium also, favors use of larger inoculum as compared to other selective media, as it has unique inhibitory action toward gram-positive organisms and coliforms.

Principles of the Procedure

Bismuth Sulphite Agar, Modified, contains 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 helps in maintaining the osmotic equilibrium. Bismuth sulphite indicator and brilliant green act as inhibitors and inhibits the intestinal gram-positive and gram-negative bacteria. Ferrous sulphate aids in detection of hydrogen sulphide production.

Formula / Liter

Ingredients	Gms / Liter
Peptic digest of animal tissue	5.00
Beef extract	5.00
Dextrose	5.00
Disodium phosphate	4.00
Ferrous sulphate	0.30
Bismuth sulphite indicator	8.00
Brilliant green	0.016
Agar	12.70
Final pH: 7.6 ± 0.2 at 25°C	
Formula may be adjusted and/or supplemented as required to meet performance specifications	

Precautions

1. For Laboratory Use only.
2. IRRITANT. Irritating to eyes, respiratory system, and skin.

3. 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.

Directions

1. Suspend 40 grams of the medium in one litre of distilled water.
2. Heat to boiling, to dissolve the medium completely.
3. DO NOT AUTOCLAVE or by fractional sterilization since overheating may destroy the selectivity of the medium.
4. Mix well and pour into sterile petri plates.

Quality Control Specifications

Dehydrated Appearance	Light yellow to greenish yellow homogeneous free flowing powder
Prepared Medium	Greenish yellow coloured opalescent with flocculent precipitate forms in Petri plates
Reaction of 4.0% Solution	pH : 7.6 ± 0.2 at 25°C
Gel Strength	Firm, comparable with 1.27% Agar gel

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

Sr. No.	Organisms	Results to be achieved			
		Inoculum (CFU)	Growth	Recovery	Color of Colony
1.	*Enterobacter aerogenes ATCC 13048*	50-100	none-poor	<=10%	brown-green (depends on inoculum density)
2.	*Enterococcus faecalis ATCC 29212*	>=10^3	inhibited	0%	-
3.	*Escherichia coli ATCC 25922*	50-100	none-poor	<=10%	brown-green (depends on inoculum density)
4.	*Salmonella typhi ATCC 19430*	50-100	good-luxuriant	>=50%	black with metallic sheen
5.	*Salmonella Paratyphi B ATCC 8759*	50-100	good-luxuriant	>=50%	black with metallic sheen
6.	*Salmonella enteritidis ATCC 13076*	50-100	good-luxuriant	>=50%	black with metallic sheen
7.	*Shigella flexneri ATCC 12022*	50-100	none-poor	<=10%	brown
8.	*Salmonella typhimurium ATCC 14028*	50-100	good-luxuriant	>=50%	black with metallic sheen

The organisms listed are the minimum that should be used for quality control testing.

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Salmonella Typhi ATCC 19430

Test Procedure

1. Clinical samples can directly be used to inoculate on Bismuth Sulphite Agar, Modified.
2. In case of food samples, pre enrichment of the sample is carried out prior to inoculation.
3. Refer to appropriate references for procedures for isolation and preliminary identification of *Salmonella* serotype *Typhi* and other *Salmonellae*.**Results**

Refer to appropriate references and test procedures for results.

Storage

Store the sealed bottle containing the dehydrated medium at 10 - 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light.

Expiration

Refer to the expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if the appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitations of the Procedure

1. Prepared medium should be stored at 2-8°C but not for more than 2 days as after which the dye oxidizes to give green medium that could be inhibitory to some Salmonellae.
2. For identification, organisms must be in pure culture. Morphological, biochemical and/or serological tests should be performed for final identification.
3. Consult appropriate texts for detailed information and recommended procedures.

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Packaging
Product Name : Bismuth Sulphite Agar, Modified (DM556)
Product Code : DM556
Available Pack sizes : 500gm

References

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7. Gunter and Tuft, 1939, J. Lab. Clin. Med., 24:461.
8. MacFaddin J. F., 2000, (Ed.), Biochemical Tests for Identification of Medical Bacteria, 3rd Edition, Lippincott, Williamss & Wilkins, New York.

Further Information

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