

## Technical Information

### Bismuth Sulphite Agar

**Product Code: DM 1027**

**Application:** Bismuth Sulphite Agar 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	10.000
Beef extract	5.000
Dextrose	5.000
Disodium phosphate	4.000
Ferrous sulphate	0.300
Bismuth sulphite indicator	8.000
Brilliant green	0.025
Agar	20.000
Final pH ( at 25°C)	7.7±0.2

\*\*Formula adjusted, standardized to suit performance parameters

#### Principle & Interpretation

The Salmonellae represent the most taxonomically complex group of bacteria among *Enterobacteriaceae* <sup>(1)</sup>. Human Salmonella infections are most commonly transmitted by ingestion of food, water or milk contaminated by human or animal excreta. Humans are the only reservoirs of *S. Typhi* <sup>(2)</sup>. Four Clinical types of salmonella infections have been described <sup>(3)</sup>. They are gastroenteritis, bacteremia or septicemia, enteric fever and a carrier state <sup>(3)</sup>. Different types of media have been employed for the isolation and preliminary identification of Salmonellae, particularly *Salmonella* Typhi; Bismuth Sulphite Agar is found tube more suitable media for this purpose <sup>(4)</sup>.

Bismuth Sulphite Agar is a modification of original Wilson and Blair Medium <sup>(5-7)</sup> which has been also recommended by various Associations <sup>(8-13)</sup> for the isolation and preliminary identification of *Salmonella* Typhi and other Salmonellae from pathological materials, sewage, water, food and other products.

*S. Typhi*, *S. Enteritidis* and *S. Typhimurium* typically grow as black colonies with a surrounding metallic sheen due to 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* like *S. Sendai*, *S. Berta*, *S. Gallinarum*, *S. Abortus-equi* and therefore should not be used as the sole selective medium for these organisms. Most *Shigella* species are inhibited on this medium; except *S. flexneri* and *S. sonnei* <sup>(14)</sup>. This medium favors use of larger inoculum as compared to other selective media, as it has unique inhibitory action towards 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.

#### Methodology

Suspend 52.33 grams of powder media in 1000 ml distilled water. Shake well & heat 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 sterile Petri plates.

## Quality Control

### Physical Appearance

Light yellow to greenish yellow homogeneous free flowing powder

### Gelling

Firm, comparable with 2.0% agar gel.

### Colour and Clarity of prepared medium

Greenish yellow coloured, opalescent with flocculent precipitate forms in Petri plates.

### Reaction

Reaction of 5.23% w/v aqueous solution at 25°C. pH : 7.7±0.2

### pH range

7.50-7.90

### Cultural Response/Characteristics

**DM1027:** Cultural characteristics observed after an incubation at 35-37°C for 40-48 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Colour of Colony
Enterobacter aerogenes ATCC 13048	50-100	None-poor	<=10%	Brown-green(depends on the inoculum density)
Enterococcus faecalis ATCC 29212	>10 <sup>3</sup>	Inhibited	0%	
Escherichia coli ATCC 25922	50-100	None-poor	<=10%	Brown-green(depends on the inoculum density)
Salmonella Enteritidis ATCC 13076	50-100	Good- luxuriant	>=50%	Black with metallic sheen
Salmonella Typhi ATCC 6539	50-100	Good-luxuriant	>=50%	Black with metallic sheen
Salmonella Typhimurium ATCC 14028	50-100	Good-luxuriant	>=50%	Black with metallic sheen
Shigella flexneri ATCC 12022	50-100	None-poor	<=10%	brown
Escherichia coli ATCC 8739	50-100	None-poor	<=10%	brown to green, depends on inoculum density
Escherichia coli NCTC 9002	50-100	None-poor	<=10%	brown to green (depends on inoculum density)
Salmonella Abony NCTC 6017	50-100	Good-luxuriant	>=50%	black with metallic sheen

## Storage and Shelf Life

**Dried Media:** Store below 30°C in tightly closed container and use before expiry date as mentioned on the label.

**Prepared Media:** 2-8<sup>o</sup> in sealable plastic bags for 2-5 days.

## Further Reading

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4. Gunter and Tuft, 1939, J. Lab. Clin. Med., 24:461.
5. Wilson and Blair, 1926, J. Pathol. Bateriaol., 29:310.
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7. Wilson and Blair, 1931, J. Hyg., 31:138



Dehydrated Culture Media  
Bases / Media Supplements

8. Washington J. A., 1981, Laboratory Procedures in Clinical Microbiology, Springer-Verlag, New York.
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10. FDA Bacteriological Analytical Manual, 2005, 18th Ed., AOAC, Washington, D.C.
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12. Downes F. P. and Ito K., (Eds.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., APHA, Washington, D.C.
13. Indian Pharmacopoeia, 1996, Ministry of Health and Family Welfare, Govt. of India, Volume 2.
14. MacFaddin J. F., 2000, (Ed.), Biochemical Tests for Identification of Medical Bacteria, 3rd Edition, Lippincott, Williams & Wilkins, New York.

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