



PRODUCT SPECIFICATION SHEET

Bismuth Sulphite Agar (DM039)

Intended Use

Bismuth Sulphite Agar (DM039) is used for the selective isolation and preliminary identification of *Salmonella* Typhi and other *Salmonellae* from pathological materials, sewage, water supplies, food etc.

Product Summary and Explanation

Salmonellosis continues to be an important public health problem worldwide. The *Salmonellae* constitute the most taxonomically complex group of bacteria among *Enterobacteriaceae* ⁽¹⁾. Infection with non-typhi *Salmonella* often causes a mild, self-limiting illness. Typhoid fever, caused by *Salmonella typhi*, is characterized by fever, headache, diarrhea, abdominal pain, and can result in fatal respiratory, hepatic, and or neurological damage ⁽²⁾. This infection can result from the consumption of raw, undercooked, or improperly processed foods contaminated with *Salmonella* spp. 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⁾

Principles of the Procedure

Bismuth Sulphite Agar is a modification of original Wilson and Blair Medium ⁽⁵⁻⁷⁾. It is also recommended by various Associations ⁽⁸⁻¹³⁾ for the isolation and preliminary identification of *Salmonella typhi* and other *Salmonellae* from pathological materials, sewage, water, food and other products. The typhoid organism grows luxuriantly on the medium, forming characteristic black colonies. Gram-positive bacteria and coliforms are inhibited on Bismuth Sulfite Agar. The inhibitory action of Bismuth Sulfite Agar permits the use of a large inoculum, increasing the possibility of recovering pathogens that may be present in small numbers. Bismuth Sulfite Agar is generally accepted for routine detection of most *Salmonella* spp. Bismuth Sulfite Agar is used for the isolation of *S. typhi* and other *Salmonella* spp. from food, feces, urine, sewage, and other infectious materials. Bismuth Sulfite Agar is a standard methods medium for industrial applications and the clinical environment.

Peptic Digest of Animal Tissue and Beef Extract provide sources of nitrogen, carbon, and vitamins required for organism growth. Dextrose is the carbohydrate present in Bismuth Sulfite Agar. Disodium Phosphate is the buffering agent. Bismuth Sulfite Indicator and Brilliant Green are complementary, inhibiting gram-positive bacteria and coliforms, allowing *Salmonella* spp. to grow. Ferrous Sulfate is used for H₂S production. When H₂S is present, the iron in the formula is precipitated, and positive cultures produce the characteristic brown to black color with metallic sheen. Agar is the solidifying agent.

Formula / Liter

Ingredients	Gms / Litre
Peptic digest of animal tissue	10.00
Beef extract	5.00
Dextrose	5.00
Disodium phosphate	4.00
Ferrous sulphate	0.30
Bismuth sulphite indicator	8.00
Brilliant green	0.025
Agar	20.00
Final pH: 7.7 ± 0.2 at 25°C	
Formula may be adjusted and/or supplemented as required to meet performance specifications	





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Precautions

1. For Laboratory Use only.
2. HARMFUL. Harmful if swallowed, inhaled, or absorbed through the skin. May cause allergic reaction and breathing difficulties. Irritating to eyes, skin, and respiratory system.

Directions

1. Suspend 52.33 grams of the medium in one liter of deionised water.
2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
3. DO NOT STERILIZE IN AUTOCLAVE or by fractional sterilization since overheating may destroy the selectivity of the medium.
4. Cool to 50-55°C, mix well to disperse suspension and pour thick plates (25ml medium per plate).
5. 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.
6. Dry the plates before use but take care to avoid overdrying.
7. Correctly prepared plates should have a smooth, greenish yellow colour with uniformly dispersed flocculent precipitate. There should be no sedimentation of the indicator.

Quality Control Specifications

Dehydrated Appearance	Light yellow to greenish yellow colored, homogeneous, free flowing powder
Prepared Medium	Greenish yellow coloured, opalescent gel with flocculent precipitate forms in Petri plates.
Reaction of 5.23% Solution	pH 7.7 ± 0.2 at 25°C
Gel Strength	Firm, compared to 2.0% Agar Gel.

Expected Cultural Response: Cultural response on Bismuth Sulphite Agar observed after incubation at 35-37°C for 40-48 hours.

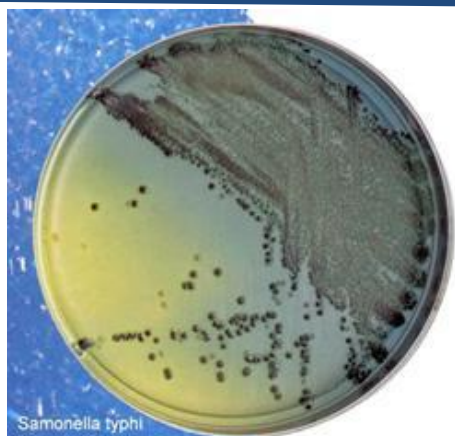
Sr. No.	Organisms	Inoculum (CFU)	Growth	Recovery	Colour of Colony
1.	<i>Enterobacter aerogenes</i> ATCC 13048	50-100	None-poor	≤10%	Brown-green
2.	<i>Enterococcus faecalis</i> ATCC 29212	50-100	Inhibited	0%	--
3.	<i>Escherichia coli</i> ATCC 25922	50-100	None-poor	≤10%	Brown-green
4.	<i>Salmonella enteritidis</i> ATCC 13076	50-100	Good-luxuriant	≥50%	Black with Metallic sheen
5.	<i>Salmonella typhi</i> ATCC 6539	50-100	Good-luxuriant	≥50%	Black with Metallic sheen
6.	<i>Salmonella typhimurium</i> ATCC 14028	50-100	Good-luxuriant	≥50%	Black with Metallic sheen
7.	<i>Shigella flexneri</i> ATCC 12022	50-100	None-poor	≤10%	Brown
8.	<i>Escherichia coli</i> ATCC 8739	50-100	None-poor	≤10%	Brown-green
9.	<i>Escherichia coli</i> NCTC 9002	50-100	None-poor	≤10%	Brown-green
10.	<i>Salmonella abony</i> NCTC 6017	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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Test Procedure

For isolation of *Salmonella typhi* and other *Salmonella* spp. consult appropriate references.

Results

Typical *S. typhi* surface colonies are black, surrounded by black or brown-black zone. This zone may be several times the size of the colony. Other strains of *Salmonella* produce black to green colonies with little or no darkening of surrounding medium. *Shigella* spp. other than *S. flexneri* and *S. sonnei* are inhibited. *S. flexneri* and *S. sonnei* strains that do grow on this medium produce brown to green, raised colonies with depressed centers and exhibit a crater-like appearance.

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 plates of medium should not be stored for longer than two days at 2-8°C; after which time the dye oxidises to give a green medium that can be inhibitory to some salmonellae.
2. Streak for well isolated colonies. In heavy growth areas, *S. typhi* appears light green and may be misinterpreted as negative for *S. typhi*.
3. *S. typhi* and *S. arizonae* are the only enteric organisms to exhibit typical brown zones on the medium. However, *S. arizonae* is usually inhibited. Typical *S. typhi* colonies usually develop within 24 hours; however, all plates should be incubated for a total of 48 hours to allow growth of all typhoid strains. When in doubt, almost any growth on the medium should be subject to further tests.
4. Do not autoclave medium. Heating medium for a long period may destroy selectivity properties.

Packaging

Product Name: Bismuth Sulfite Agar

Product Code : DM039

Available Pack sizes : 100gm / 500gm





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References

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Further Information

For further information please contact your local MICROMASTER Representative.



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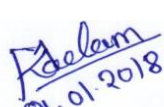
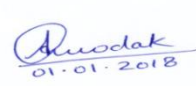
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