BISMUTH SULFITE AGAR (7113)

Intended Use

Bismuth Sulfite Agar is used for the selective isolation of *Salmonella* spp.

Product Summary and Explanation

Salmonellosis continues to be an important public health problem worldwide. Infection with non-typhi *Salmonella* often causes mild, self-limiting illness. Typhoid fever, caused by *S. typhi*, is characterized by fever, headache, diarrhea, and abdominal pain, and can result in fatal respiratory, hepatic, and/or neurological damage.[1] Salmonellosis can result from consumption of raw, undercooked, or improperly processed foods contaminated with *Salmonella*. U. S. federal guidelines require various poultry products to be routinely monitored before distribution for human consumption.

Bismuth Sulfite Agar is a modification of Wilson and Blair formula.[2-4] 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.[5-9]

Principles of the Procedure

Enzymatic Digest of Casein, Enzymatic 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 H_2S production. When H_2S 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

Enzymatic Digest of Casein	5 g
Enzymatic Digest of Animal Tissue	5 g
Beef Extract	5 g
Dextrose	5 g
Disodium Phosphate	4 g
Ferrous Sulfate	0.3 g
Bismuth Sulfite Indicator	8 g
Brilliant Green	0.025 g
Agar	20 g

Final pH: 7.5 ± 0.2 at 25°C

Formula may be adjusted and/or supplemented as required to meet performance specifications.

Precautions

1. For Laboratory Use.
2. HARMFUL. Harmful by inhalation, ingestion, and skin contact.

Directions

1. Suspend 52 g of the medium in one liter of purified water.
2. Heat with frequent agitation and boil for one minute.
3. Mix thoroughly to obtain a uniform suspension prior to dispensing.
4. Prepared plates may be used the same day as prepared.
5. For increased selectivity, current references suggest that prepared BSA plates be stored overnight in the dark at room temperature.[8]

Quality Control Specifications

Dehydrated Appearance: Powder is homogeneous, free flowing, and light green-beige.

Prepared Appearance: Prepared medium at 45 - 50°C is green-yellow and opaque.

Expected Cultural Response: Cultural response on Bismuth Sulfite Agar at 35°C after 40 - 48 hours incubation.

Microorganism	Response	Reactions
Enterococcus faecalis ATCC® 29212	inhibited	---
Escherichia coli ATCC® 25922	partially inhibited	brown-green colonies
Salmonella typhimurium ATCC® 19430	growth	black colonies with metallic sheen
Shigella flexneri ATCC® 12022	growth	brown colonies

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

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 sealed bottle containing the dehydrated medium at 2 - 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping container tightly closed.

Expiration

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

Limitations of the Procedure

1. Streak for well isolated colonies. In heavy growth areas, *S. typhi* appears light green and may be misinterpreted as negative for *S. typhi*.[10]
2. *S. typhi* and *S. arizonae* are the only enteric organisms to exhibit typical brown zones on the medium. However, *S. arizonae* is usually inhibited.[10] 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.[10]
3. Do not autoclave medium. Heating Bismuth Sulfite Agar for a period longer than necessary may destroy selectivity properties.

Packaging

Bismuth Sulfite Agar	**Code No.**	**7113A**	**500 g**
		7113B	**2 kg**
		7113C	**10 kg**

References

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7. **United States Pharmacopeial Convention.** 1995. The United States pharmacopeia, 23rd ed. The United States Pharmacopeial Convention, Rockville, MD.
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9. **Cunniff, P. (ed.).** Official methods of analysis of AOAC International, 16th ed. AOAC International, Arlington, VA.
10. **MacFaddin, J. F.** 1985. Media for isolation-cultivation-identification-maintenance of medical bacteria, Vol. 1. Williams & Wilkins, Baltimore, MD.

Technical Information

Contact Acumedia Manufacturers, Inc. for Technical Service or questions involving dehydrated culture media preparation or performance at (410)780-5120 or fax us at (410)780-5470.