

Bismuth Sulphite HiCynth™ Agar

MCD027

Bismuth Sulphite HiCynth™ Agar is recommended for the selective isolation of Salmonellae from faeces, urine, sewage, and other materials.

Composition**

Ingredients	Gms / Litre
HiCynth™ Peptone No.2*	10.000
HiCynth™ Peptone No.6*	5.000
Dextrose	5.000
Disodium phosphate	4.000
Bismuth ammonium citrate	8.000
Ferrous sulphate	0.300
Brilliant green	0.025
Agar	20.000
Final pH (at 25°C)	7.7±0.2

**Formula adjusted, standardized to suit performance parameters

*Chemically defined peptones

Directions

Suspend 52.33 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 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.

Bismuth Sulphite HiCynth™ Agar is a modification of Bismuth Sulphite Agar wherein peptones are replaced by chemically defined peptones to avoid BSE/TSE risks associated with animal peptones. Bismuth Sulphite Agar is a modification of the original Wilson and Blair Medium (4-6). It is also recommended by various Associations (7-12) 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 resulting from hydrogen sulphide production and reduction of sulphite to black ferric sulphide. *Salmonella Paratyphi A* grows as light green colonies. Bismuth Sulphite HiCynth™ 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* (13) and also some *Salmonella* like *S. Sendai*, *S. Berta*, *S. Gallinarum*, *S. Abortus-equi* are inhibited (13). Also 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.

HiCynth™ Peptone No.2 and HiCynth™ Peptone No.6 serve as sources as carbon, nitrogen, long chain amino acids, 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 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

7.50-7.90

Cultural Response

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

Cultural Response

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

Storage and Shelf Life

Store below 30°C in a tightly closed container 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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