Technical Information

Bismuth Sulphite Agar Medium

Product Code: DM 1027U

Application: Bismuth Sulphite Agar Medium is recommended for the selective isolation of Salmonellae from faeces, urine, sewage and other materials in accordance with United States Pharmacopoeia.

Composition**

Ingredients	Gms / Litre
Pancreatic digest of casein	5.000
Beef extract	5.000
Peptic digest of animal tissue	5.000
Dextrose	5.000
Sodium phosphate	4.000
Ferrous sulphate	0.300
Bismuth sulphite indicator	8.000
Brilliant green	0.025
Agar	20.000
Final pH (at 25°C)	7.6±0.2

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

Bismuth Sulphite Agar Medium is prepared in accordance with USP [1] and is used for the isolation and preliminary identification of *Salmonella* Typhi and other Salmonellae from pathological materials, namely sewage, water, food and other products. This medium is recommended by various Associations for the isolation and preliminary identification of *Salmonella* Typhi and other Salmonellae from different pathological materials, and other products [2-6]. It is a modification of Wilson and Blair medium.

Brilliant green and bismuth sulphite & added into the medium that inhibit the growth of intestinal gram-negative and gram-positive bacteria, Peptic digest of animal tissue, pancreatic digest of casein and beef extract are rich source for supplying essential nutrients for growth of the organism. The fermentable source of carbohydrate in this medium is dextrose, which provides energy for enhanced microbial growth. Phosphates incorporated in the medium act as a good buffering agent. The bismuth ions are reduced to metallic bismuth, which impart the metallic sheen around the colonies. Sulphite is reduced to black ferric sulphide giving the black colour with release of H_2S.

Salmonella Enteritidis and *Salmonella* Typhimurium typically grow as black colonies (rabbit eye colonies) with a surrounding metallic sheen. *Salmonella* Paratyphi A grows as light green colonies. This medium requires use of larger inoculum and heavily contaminated samples as compared to other selective media, as it has unique inhibitory action towards gram-positive and coliform organisms. The medium may be inhibitory to some strains of *Salmonella* species like *S.* Sendai, *S.* Berta, *S.* Gallinarum, *S.* Abortus-equi and therefore should not be used as the sole selective medium for these organisms. Same is hold good for *Shigella* species. Proteus species inhibited

Methodology

Suspend 52.32 grams of powder media in 1000 ml purified/ distilled water. Shake well & heat to boiling to dissolve the medium completely. DO NOT OVERHEAT OR STERILIZE IN AUTOCLAVE or by fractional sterilization since overheating may destroy the selectivity of the medium. Transfer to a water bath maintained at about 50°C .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.

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
Yellow to greenish yellow opalescent with flocculant precipitate
Reaction
Reaction of 5.23% w/v aqueous solution. pH : 7.6±0.2
pH range 7.40-7.80
Cultural Response
Growth Promotion is carried out in accordance with the harmonized method of USP. Cultural response was observed after an incubation at 30-35°C for 24-48 hours. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar.
CulturalResponse/Characteristics
DM1027U: Cultural characteristics observed after incubation at 30-35 °C for 24-48 hours. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar.

Organism	Inoculum (CFU)	Growth	Lot value (CFU)	Recovery	Colour of Colony
Salmonella Typhimurium ATCC 4028	50-100	luxuriant	25-100	>=50%	black or greenish-grey may have sheen
Salmonella A bony NCTC 6017	50-100	Good-luxuriant	25-100	>=50%	Black with metallic sheen
Additional Microbiological Testing					
Enterobacter aerogenes ATCC 13048	50-100	None-poor	0-10	0-10%	brown-green (depends on the inoculums density)
Enterococcus faecalis ATCC 29212	>=10^3	inhibited	0	0%	
Salmonella Enteritidis ATCC 13076	50-100	luxuriant	25-100	>=50%	black with metallic sheen
Salmonella Typhi ATCC 6539	50-100	luxuriant	50-100	>=50%	Black with metallic
Shigella flexneri ATCC 12022	50-100	None-poor	0-10	<=10%	brown
Escherichia coli ATCC 8739	50-100	None-poor	0-10	<=10%	Brown to green, depends on inoculum density

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: 2-8^0 in sealable plastic bags for 2-5 days

Further Reading

1. United States Pharmacopoeia, 2009, U. S. Pharmacopoeial Convention, Inc., Rockville, MD.
2. Washington J.A.,1981,Laboratory Procedures in Clinical Microbiology, Springer-Verlag, New York.
3. Eaton A. D., Clesceri L. S. and Greenberg A W.,(Eds.), 2005, Standard Methods for the Examination of Water and Wastewater, 21st ed., APHA, Washington, D.C.
4. Bacteriological Analytical Manual, 8th Edition, Revision A, 1998. AOAC, Washington D.C.
5. Murray PR, Baren EJ, Jorgensen JH, Pfaller MA, Yolken RH (editors) 2003, Manual of clinical Microbiology, 8th ed., ASM, Washington, D.C..
6. Downes F P and Ito K. (Eds.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th ed., APHA, Washington, D.C

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