Technical Information

Brilliant Green Agar Medium

Product Code: DM 1016U

Application: - Brilliant Green Agar Medium is used for selective isolation of Salmonellae other than *Salmonella* Typhi from faeces, foods, dairy products etc. in accordance with United States Pharmacopoeia.

Composition**

Ingredients	Gms / Litre
Peptic digest of animal tissue	5.000
Pancreatic digest of casein	5.000
Yeast extract	3.000
Lactose	10.000
Sucrose	10.000
Sodium chloride	5.000
Phenol red	0.080
Brilliant green	0.0125
Agar	20.000
Final pH (25°C)	6.9±0.2

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

Brilliant Green Agar medium was first described by Kristensen et al is recommended as a primary plating medium for isolation of Salmonella species and for differentiation of paratyphoid B from other Gram negative enteric bacteria [1]. Kauffmann further modified it for isolation of Salmonella from stool samples [2]. Brilliant green agar is also recommended by APHA [3, 4] FDA [5] and is United States Pharmacopoeia [6]. Heavy inocula and heavily contaminated samples can be analyzed due to the not worthy selectivity of this medium. Brilliant Green Agar is used in the microbial limits test and with novobiocin for testing food and pharmaceutical products. This medium is employed in testing of clinical specimens.

Combination of peptic digest of animal tissue, pancreatic digest of casein and yeast extract makes the medium highly nutritious and supplies amino acids and long chains of peptides. Sodium chloride maintains the osmotic equilibrium. Lactose and sucrose are the fermentable carbohydrate sources. Phenol red serves as an acid base indicator giving yellow colour to lactose and or sucrose fermenting bacteria. This medium also contains brilliant green, which inhibits growth of majority of Gram-negative and Gram-positive, bacteria. Salmonella Typhi, Shigella species, Escherichia coli, Proteus species, Pseudomonas species, Staphylococcus aureus are mostly inhibited.

However, being highly selective, it is recommended that this medium should be used along with a less inhibitory medium to increase the chances of recovery. Often cultures enriched in Selenite or Tetrathionate Broth are plated on Brilliant Green Agar along with Bismuth Sulphite Agar, SS Agar, MacConkey Agar. Non-lactose fermenting bacteria develop white to pinkish red colonies within 18-24 hours of incubation. Salmonella typhi and Shigella species may not grow on this medium; moreover Proteus, Pseudomonas and Citrobacter species may mimic enteric pathogens by producing small red colonies.

Methodology

Suspend 58.09 grams of powder media in 1000 ml purified /distilled water. Shake well & heat to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. AVOID OVERHEATING. Cool to 50°C. Mix well before pouring into sterile Petri plates.

Quality Control

Physical Appearance

Light yellow to light pink homogeneous free flowing powder

Gelling

Firm, comparable with 2.0% agar gel.

Colour and Clarity of prepared medium

Greenish brown clear to slightly opalescent gel forms in Petri plates

Reaction
Reaction of 5.8^{0}C% w/v aqueous solution at 25°C. pH : 6.9±0.2

pH Range 6.70-7.10
Growth Promotion Test
Growth Promotion was observed in accordance with USP.
Cultural Response/ characteristics
DM 1016U: Cultural characteristics observed after an incubation at 35-37°C for 24-48 hours. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar.

Organism	Inoculum (CFU)	Growth	Observed Lot value (CFU)	Recovery	Colour of colony	Incubation temperature
Salmonella Typhimurium ATCC 14028	50-100	good-luxuriant	25-100	>=50 %	pinkish white	24 -48 hrs
Salmonella Abony NCTC 6017	50-100	good-luxuriant	25-100	>=50 %	pinkish white	24 -48 hrs
Additional Microbiological testing						
Salmonella Enteritidis ATCC 13076	50-100	luxuriant	25-100	>=50 %	pinkish white	24 -48 hrs
Salmonella Typhi ATCC 6539	50-100	poor-good	15-40	30-40%	reddish pink	24 -48 hrs
Escherichia coli ATCC 25922	50-100	none-poor	0-10	0-10%	yellowish green	24 -48 hrs
Escherichia coli ATCC 8739	50-100	none-poor	0-10	0-10%	yellowish green	24 -48 hrs
Escherichia coli NCTC 9002	50-100	none-poor	0-10	0-10%	yellowish green	24 -48 hrs
Staphylococcus aureus ATCC 25923	>=10^3	inhibited	0	0%		24 -48 hrs
Staphylococcus aureus ATCC 6538	>=10^3	inhibited	0	0%		24 -48 hrs

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^o in sealable plastic bags for 2-5 days.

Further Reading

1. Kristensen M., Lester V, and Jurgens A., 1925, Brit.J.Exp.Pathol.,6:291.
2. Kauffman F., 1935, Seit F. Hyg. 177:26
3. Downes F P and Ito K. (Eds.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th ed., APHA, Washington, D.C.
4. Standard Methods for the Microbiological Examination of Dairy Products, 1995, 19th Ed, APHA, Washington, D.C.
5. Bacteriological Analytical Manual, 8th Edition, Revision A, 1998. AOAC, Washington D.C.
6. The United States Pharmacopoeia, 2009. USP Conv. Rockville, MD.

Disclaimer :

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