



BRILLIANT GREEN AGAR BASE W/ 1.2% AGAR

TM 047

For selective isolation of *Salmonellae* other than *Salmonellae typhi* from faeces, food and dairy products

Composition

Ingredients	Gms/Ltr.
Agar	12.00
Proteose peptone	10.00
Lactose	10.00
Sucrose	10.00
Sodium chloride	5.00
Yeast extract	3.00
Phenol red	0.080
Brilliant green	0.0125

* Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers below 25°C and protect from direct Sunlight.

Instructions for Use

Dissolve 25.00gms in 500ml distilled water. Gently heat to boiling with gentle swirling and dissolve the medium completely. Sterilize by autoclaving at 15 psi (121 °C) for 15 minutes. AVOID OVERHEATING. Aseptically add rehydrated contents of 1 vial of SULPHA SUPPLEMENT (TS 013). Mix well and dispense into sterile Petri plates.

Appearance: Greenish brown colour, clear to slightly opalescent gel

pH (at 25°C): 6.9 ± 0.2

Principle

BRILLIANT GREEN AGAR BASE W/ 1.2% AGAR, is used for the selective isolation of *Salmonellae* other than *Salmonella typhi* from faeces and foods etc. Brilliant Green Agar Base, Modified, as a primary plating medium for isolation of *Salmonella* species was first described by “**Kristensen**” et. al. and further modified by “**Kauffmann**”. The medium contains Proteose peptone and Yeast extract as sources of nitrogen, vitamins, amino acids and essential nutrients. The two sugars namely Lactose and Sucrose serve as energy sources. Fermentation of Lactose and Sucrose in the medium results in the formation of acidic medium which is detected by Phenol red indicator. Brilliant green dye inhibits gram-positive bacteria and a majority of gram-negative *Bacilli*. Phenol red serves as a pH indicator and yields a yellow color as a result of acid production. Sodium chloride maintains the osmotic equilibrium. Agar is a solidifying agent. Clinical specimens can be directly plated on this medium. 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. The medium can further supplemented with sulphha supplements to inhibit contaminating microorganisms when the sample is suspected to contain large number of competing organisms along with *Salmonella* species. 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



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medium. Moreover *Proteus*, *Pseudomonas* and *Citrobacter* species may mimic enteric pathogens by producing small red colonies.

Interpretation

Cultural characteristics observed after inoculating (10^3 - 10^5 CFU/ml), on incubation at 30 - 35°C for 24 - 48 hours.

Microorganisms	ATCC	Inoculum (cfu/ml)	Growth	Colour of colonies
<i>Salmonella typhimurium</i>	14028	10^3	Luxuriant	Pinkish white
<i>Salmonella enteritidis</i>	13076	10^3	Good	Pinkish white
<i>Escherichia coli</i>	25922	10^3	None to poor	Yellowish green
<i>Salmonella typhi</i>	6539	10^3	Fair to good	Reddish pink
<i>Staphylococcus aureus</i>	25923	10^5	Inhibited	-----

References

1. Downes F. P. and Ito K. (Ed), Compendium of Methods for Microbiological Examination of Foods, 4th Ed. APHA, Washington D.C. (2001).
2. Kauffman F., Seit F. Hyg. 177: 26. (1935).
3. Kristensen M., Lester V, and Jurgens A., Brit.J.Exp. Pathol., 6:291. (1925,).
4. U.S. Food and Drug Administration. Bacteriological analytical manual, 8th ed. AOAC International, Gaithersburg, Md. (1995).