



Brilliant Green Sulpha Agar

M492

Brilliant Green Sulpha Agar is used for the selective isolation and detection of *Salmonella* species in foods especially from eggs and egg products

Composition**

Ingredients	Gms / Litre
Yeast extract	3.000
Proteose peptone	10.000
Lactose	10.000
Sucrose	10.000
Sodium sulphapyridine	1.000
Sodium chloride	5.000
Brilliant green	0.0125
Phenol red	0.080
Agar	20.000
Final pH (at 25°C)	6.9±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 59.09 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. To maintain selectivity of the medium, DO NOT OVER STERILIZE OR OVERHEAT the medium.

Principle And Interpretation

Salmonella species are ubiquitous in the environment. These enter the gastrointestinal tract of animals due to consumption of contaminated feed. Stringent animal husbandry practices are used in the meat (food) industry and inedible raw materials are recycled and discarded. Thus the organisms are further returned to the environment and stay in the global food chain (1, 2). Eggshell and its contents are usually sterile at the time of oviposition. Subsequently it gets contaminated on contact with the nest, the floor and litter of other birds (5-7).

Salmonella species are usually the causative agents of a self-limiting gastroenteritis. In some cases they may also cause typhoid fever. *Salmonella* contamination is most frequently encountered in the poultry industry. Brilliant Green Sulpha Agar is used for the selective isolation and detection of *Salmonella* species in foods especially from eggs and egg products. Brilliant Green Agar was first formulated by Kristensen, Lester and Jargens (3). This was further modified by Osborne and Stokes (4) by the addition of 0.1% sodium sulphapyridine to the original formulation. This addition helped to increase the selective properties of the medium. Colonies of *Salmonella* may sometimes vary from red to pink to white depending upon the time and length of incubation and the strain of *Salmonella*. Do not autoclave the medium for more than 15 minutes as it decreases the selectivity of the medium (8).

Yeast extract and proteose peptone provide essential growth nutrients, amino acids and vitamins. Brilliant green used in the medium is inhibitory to gram-positive and most gram-negative lactose/sucrose fermenting bacilli. Sulphapyridine enhances the selectivity of the medium. The medium does not support luxuriant growth of *Salmonella* Typhi. *Shigella* species also fail to grow on Brilliant Green Sulpha Agar (8). Since Brilliant Green Sulpha Agar is highly selective, a less inhibitory medium should be simultaneously used to recover organisms from the pre-enriched culture (Selenite Cystine Medium).

Quality Control

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 coloured, clear to slightly opalescent gel forms in Petri plates.

Reaction

Reaction of 5.9% w/v aqueous solution at 25°C. pH : 6.9±0.2

pH

6.70-7.10

Cultural Response

M492: Cultural characteristics observed after an incubation at 35-37°C for 24-48 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony
<i>Enterococcus faecalis</i> ATCC 29212	≥10 ³	inhibited	0%	
<i>Escherichia coli</i> ATCC 25922	50-100	none-poor	≤10%	yellow green surrounded by intense yellow-green zone
<i>Proteus vulgaris</i> ATCC 13315	≥10 ³	inhibited	0%	
<i>Salmonella Enteritidis</i> ATCC 13076	50-100	good	≥50%	pink-white, surrounded by a brilliant red-zone
<i>Salmonella Typhimurium</i> ATCC 14028	50-100	good	≥50%	pink - white
<i>Staphylococcus aureus</i> ATCC 25923	≥10 ³	inhibited	0%	

Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label.

Reference

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4. Osborne W. W. and Stokes J. L., 1955, Ottawa; Food and Drug Laboratories, 1962.
5. Brooks and Taylor, 1955, Rep. Rd. Invest. Bd. 60, H. M. S. O., London, England.
6. Forsythe, Ayres and Radlo, 1953, Food Technol., 7:49.
7. Stadelman, Ikeme, Roop and Simmons, 1982, Poultry Sci., 61:388.
8. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. I, Williams & Wilkins, Baltimore, MD.

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