Brilliant Green Agar (NCM0283)

Revision: 0 Effective Date: 11/19/2021

Intended Use

Brilliant Green Agar is used for the selective isolation of *Salmonella* spp. in a laboratory setting. Brilliant Green Agar is not intended for use in the diagnosis of disease or other conditions in humans.

Description

Kristensen, Lester, and Jurgens first described the use of Brilliant Green Agar as a primary plating medium for the isolation of *Salmonella*. The report described the medium as useful for the differentiation of "paratyphoid B" from other intestinal gram-negative bacilli. Kaufmann modified their formula and used Brilliant Green Agar in addition to Tetrathionate Broth for the isolation of *Salmonella* from stool specimens. The outstanding selectivity of this medium permits use of moderately heavy inocula, which should be evenly distributed over the surface. Brilliant Green Agar is used in the microbial limits test. Brilliant Green Agar supplemented with novobiocin is used in food testing.

Typical Formulation

Yeast Extract	3.0 g/L
Enzymatic Digest of Casein	5.0 g/L
Enzymatic Digest of Animal Tissue	5.0 g/L
Sodium Chloride	5.0 g/L
Lactose	10.0 g/L
Sucrose	10.0 g/L
Brilliant Green	0.0125 g/L
Phenol Red	0.08 g/L
Agar	20.0 g/L

Final pH: 6.9 ± 0.2 at 25°C

Formula may be adjusted and/or supplemented as required to meet performance specifications.

Precaution

Refer to SDS

Preparation

1. Suspend 58 g of the medium in one liter of purified water.
2. Mix thoroughly.
3. Autoclave at 121°C for 15 minutes.
4. Cool to 45-50°C.

Quality Control Specifications

Dehydrated Appearance: Powder is homogeneous, free flowing, beige and may have a green tint.

Prepared Appearance: Prepared medium is orange-brown and may have a green tint, and trace hazy to slightly opalescent.

Expected Cultural Response: Cultural response on Brilliant Green Agar at 35 ± 2°C after 18 - 24 hours incubation.

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Microorganism	Approx. Inoculum (CFU)	Expected Results	
		Growth	Reactions
Escherichia coli ATCC® 25922	1000	Partial to complete inhibition	Green colonies
Salmonella enteritidis ATCC® 13076	10 - 300	Fair to good	Pink colonies
Salmonella typhi ATCC® 19430	1000	None to poor	Pink colonies
Salmonella typhimurium ATCC® 14028	10 - 300	Fair to good	Pink colonies
Staphylococcus aureus ATCC® 25923	1000	Complete Inhibition	---

The organisms listed are the minimum that should be used for quality control testing.

Test Procedure

Inoculate specimens on the first quadrant of Brilliant Green Agar and streak for isolation. Incubate plates at 35°C. Examine plates after 18 – 24 hours for colonies with characteristic morphologies associated with *Salmonella* spp. Refer to appropriate references or standard methods for other applications.

Results

Typical *Salmonella* spp. colonies are opaque and pink. The few lactose and/or sucrose fermenting Organisms that grow are readily differentiated due to formation of green colonies. Brilliant Green Agar is not suitable for the isolation of *S. typhi* or *Shigella* spp., although some strains of *S. typhi* may grow.

Expiration

Refer to expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if appearance has changed from the original color. Expiry applies to medium in its intact container.

Limitations of the Procedure

1. Colonies of *Salmonella* spp. can be red, pink, or white depending on length of incubation and strain.
2. Medium is normally orange-brown, however after incubation it can turn bright red and return to normal color at room temperature.
3. Taylor showed that slow lactose fermenters, *Proteus*, *Citrobacter*, and *Pseudomonas* may grow on BG Agar as red colonies.
4. Other prepared media should be used along with Brilliant Green Agar when testing for intestinal pathogens.

Storage

Store dehydrated culture media at 2-30°C away from direct sunlight. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping container tightly closed.

References

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3. United States Pharmacopeia National Formulary 2018: USP 41 NF 35.
4. Federal Register. 1993. Chicken disease caused by *Salmonella enteritidis*; proposed rule. Fed. Regist. 58:41048-41061.
5. Taylor, W. I. 1965. Isolation of shigellae. I. Xylose lysine agars: New media for isolation of enteric pathogens. Am J. Clin. Pathol. 44:471.
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