

BG Sulfa Agar

SBG Sulfa Enrichment

Intended Use

BG Sulfa Agar is used for isolating *Salmonella*.

SBG Sulfa Enrichment is used for enriching *Salmonella* prior to isolation procedures.

Summary and Explanation

Salmonellosis continues to be an important public health problem worldwide, despite efforts to control the prevalence of *Salmonella* in domesticated animals. Infection with non-typhi *Salmonella* often causes mild, self-limiting illness. The illness results from consumption of raw, undercooked or improperly processed foods contaminated with *Salmonella*.

Many of these cases of *Salmonella*-related gastroenteritis are due to improper handling of poultry products. Various poultry products are routinely monitored for *Salmonella* before their distribution for human consumption, but in many instances, contaminated food samples elude detection.

BG (Brilliant Green) Sulfa Agar is a highly selective medium. Osborne and Stokes¹ added 0.1% sodium sulfapyridine to Brilliant Green Agar to enhance the selective properties of this medium for *Salmonella*. This formula is recommended as a selective isolation medium for *Salmonella* following enrichment.² It is also recommended for direct inoculation with primary specimens for *Salmonella* isolation.

User Quality Control

Identity Specifications

Difco™ BG Sulfa Agar

Dehydrated Appearance:	Pink, free flowing, homogeneous.
Solution:	5.9% solution, soluble in purified water upon boiling. Solution is very dark amber, very slightly to slightly opalescent.
Prepared Appearance:	Orange-brown to dark reddish-amber, slightly opalescent.
Reaction of 5.9% Solution at 25°C:	pH 6.9 ± 0.2

Difco™ SBG Sulfa Enrichment

Dehydrated Appearance:	Light beige, free-flowing, homogeneous.
Solution:	2.42% solution, soluble in purified water. Solution is green, opalescent, may have a precipitate.
Prepared Appearance:	Green, opalescent without significant precipitate.
Reaction of 2.42% Solution at 25°C:	pH 7.2 ± 0.2

Cultural Response

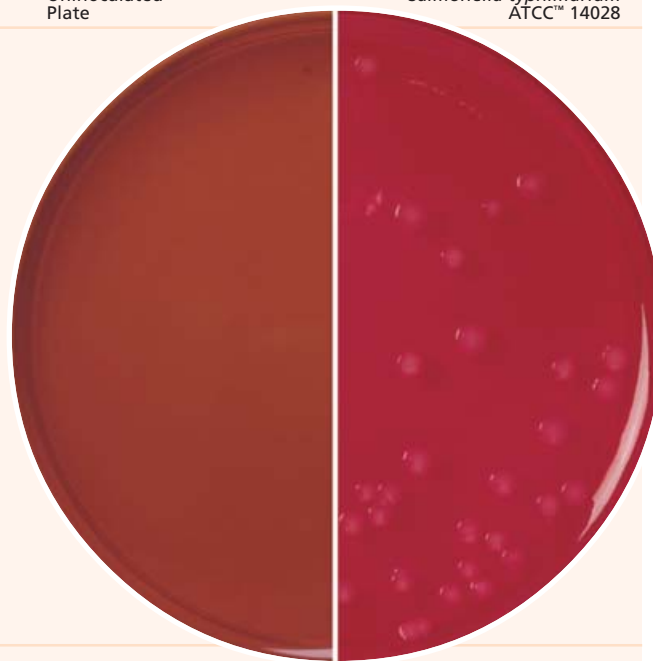
Difco™ BG Sulfa Agar

Prepare the medium per label directions. Inoculate and incubate at 35 ± 2°C for 18-48 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY	COLOR OF COLONIES/MEDIUM
<i>Enterococcus faecalis</i>	29212	10 ³ -2 × 10 ³	None	–/no change
<i>Escherichia coli</i>	25922	10 ² -3 × 10 ²	None to poor	Yellow-green/ Yellow-green
<i>Proteus vulgaris</i>	13315	10 ² -3 × 10 ²	None	–/no change
<i>Salmonella choleraesuis</i> subsp. <i>choleraesuis</i> serotype Enteritidis	13076	10 ² -3 × 10 ²	Good	Pink-white/ red
<i>Salmonella choleraesuis</i> subsp. <i>choleraesuis</i> serotype Typhimurium	14028	10 ² -3 × 10 ²	Good	Pink-white/ red

Uninoculated Plate

Salmonella typhimurium ATCC™ 14028



Difco™ SBG Sulfa Enrichment

Prepare the enrichment per label directions. Inoculate and incubate at 35 ± 2°C for 18-24 hours. After incubation, subculture onto plates of MacConkey Agar and incubate at 35 ± 2°C for 18-24 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY	COLONY COLOR ON MACCONKEY
<i>Escherichia coli</i>	25922	10 ² -3 × 10 ²	None to poor	Pink, if any
<i>Salmonella choleraesuis</i> subsp. <i>choleraesuis</i> serotype Enteritidis	13076	10 ² -3 × 10 ²	Good	Colorless
<i>Salmonella choleraesuis</i> subsp. <i>choleraesuis</i> serotype Typhimurium	14028	10 ² -3 × 10 ²	Good	Colorless
<i>Shigella sonnei</i>	9290	10 ² -3 × 10 ²	None to fair	Colorless

For food testing, BG Sulfa Agar has been used for detection of *Salmonella* in low and high moisture foods.^{3,4} It has also been used for detecting *Salmonella* in feeds and feed ingredients.⁵ This medium is recommended when testing foods for *Salmonella* following USDA guidelines.⁶⁻⁸

SBG (Selenite Brilliant Green) Sulfa Enrichment is prepared according to the formula described by Stokes and Osborne.⁹ The researchers found that whole egg and egg yolk reduced the selective properties of selenite brilliant green enrichment.¹ They also found that the addition of sulfapyridine (SBG Sulfa Enrichment) restored these selective properties.¹

SBG Sulfa Enrichment is a selective enrichment for the isolation of *Salmonella* species, especially from egg products. The shell and the contents of the egg at the time of oviposition are generally sterile or harbor very few microorganisms. Contamination of the shell occurs afterwards from nesting material, floor litter and avian fecal matter.¹⁰⁻¹²

Principles of the Procedure

In BG Sulfa Agar, peptone and yeast extract provide nitrogen, vitamins and minerals. Lactose and sucrose are the sources of carbohydrates in the medium. Brilliant green and sodium pyridine are complementary in inhibiting gram-positive bacteria and most gram-negative bacilli other than *Salmonella* spp. Phenol red is the pH indicator that turns the medium a yellow color with the formation of acid when lactose and/or sucrose is fermented. Agar is the solidifying agent.

Peptone provides the nitrogen, minerals and amino acids in SBG Sulfa Enrichment. Yeast extract is the vitamin source. D-Mannitol is the carbon source to stimulate organism growth. The phosphates act as buffers in the enrichment. Sodium taurocholate, sodium selenite and brilliant green are the selective agents. The selective agents are used to inhibit gram-positive organisms and enteric bacteria other than *Salmonella*. Sodium sulfapyridine is added to increase selectivity.

Formulae

Difco™ BG Sulfa Agar

Approximate Formula* Per Liter	
Yeast Extract	3.0 g
Proteose Peptone No. 3	10.0 g
Lactose	10.0 g
Saccharose	10.0 g
Sodium Sulfapyridine	1.0 g
Sodium Chloride	5.0 g
Agar	20.0 g
Brilliant Green	12.5 mg
Phenol Red	0.08 g

Difco™ SBG Sulfa Enrichment

Approximate Formula* Per Liter	
Yeast Extract	5.0 g
Peptone	5.0 g
D-Mannitol	5.0 g
Sodium Taurocholate	1.0 g
Sodium Sulfapyridine	0.5 g
Sodium Selenite	4.0 g
Dipotassium Phosphate	2.65 g
Monopotassium Phosphate	1.02 g
Brilliant Green	5.0 mg

*Adjusted and/or supplemented as required to meet performance criteria.

Directions for Preparation from Dehydrated Product

Difco™ BG Sulfa Agar

1. Suspend 59 g of the powder in 1 L of purified water. Mix thoroughly.
2. Heat with frequent agitation and boil for 1 minute to completely dissolve the powder.
3. Autoclave at 121°C for 15 minutes. Avoid overheating, which will decrease selectivity.
4. Test samples of the finished product for performance using stable, typical control cultures.

Difco™ SBG Sulfa Enrichment

1. Suspend 24.2 g of the powder in 1 L of purified water. Mix thoroughly.
2. Heat with frequent agitation and boil for 5-10 minutes to completely dissolve the powder. Avoid overheating. DO NOT AUTOCLAVE.
3. Test samples of the finished product for performance using stable, typical control cultures.

Procedure

Refer to appropriate references for specific procedures for the isolation and cultivation of *Salmonella* from meat, poultry and egg products and other foods.^{2,7,8}

Expected Results

BG Sulfa Agar

The typical *Salmonella* colonies appear as pink-white to red opaque colonies surrounded by a brilliant red medium. The few lactose and/or sucrose fermenting organisms that grow are readily differentiated due to the formation of a yellow-green colony surrounded by an intense yellow-green zone. BG Sulfa Agar is not suitable for the isolation of *S. typhi* or *Shigella*; however, some strains of *S. typhi* may grow forming red colonies.

SBG Sulfa Enrichment

Examine prepared media for growth. Positive tubes should be subcultured onto prepared media for isolation and identification of bacteria.

Limitations of the Procedure

1. On BG Sulfa Agar colonies of *Salmonella* spp. vary from red to pink to white depending on length of incubation and strain.¹³
2. BG Sulfa Agar is normally orange-brown in color; however, on incubation, it turns bright red and returns to normal color at room temperature.¹³
3. *S. typhi* does not grow adequately on BG Sulfa Agar. *Shigella* spp. do not grow on BG Sulfa Agar.¹³
4. Do not autoclave BG Sulfa Agar longer than 15 minutes; longer periods decrease the selectivity of the medium.
5. Since BG Sulfa Agar is highly selective, it is recommended that less selective media, such as MacConkey Agar, be used simultaneously.

6. SBG Sulfa Enrichment should be used in conjunction with a selective prepared medium for bacterial identification.

References

- Osborn and Stokes. 1955. Appl. Microbiol. 3:295.
- Downes and Ito (ed.). 2001. Compendium of methods for the microbiological examination of foods, 4th ed. American Public Health Association, Washington, D.C.
- D'Aoust, Maishment, Burgener, Conley, Loit, Milling and Purvis. 1980. J. Food Prot. 43:343.
- D'Aoust. 1984. J. Food Prot. 47:588.
- D'Aoust, Sewell and Boville. 1983. J. Food Prot. 46:851.
- Moats. 1981. J. Food Prot. 44:375.
- Federal Register. 1996. Fed. Regist. 61:38917.
- U.S. Department of Agriculture. 1998. Microbiology laboratory guidebook, 3rd ed. Food Safety and Inspection Service, USDA, Washington, D.C.
- Osborn and Stokes. 1955. Appl. Microbiol. 3:217.

- Brooks and Taylor. 1955. Rep. Rd. Invest., Bd. 60, H. M. S. O. London, England.
- Forsythe, Ayres and Radlo. 1953. Food Technol. 7:49.
- Stadelman, Ikeme, Roop and Simmons. 1982. Poultry Sci. 61:388.
- MacFaddin. 1985. Media for isolation-cultivation-identification-maintenance of medical bacteria, vol. 1. Williams & Wilkins, Baltimore, Md.

Availability

Difco™ BG Sulfa Agar

CCAM COMPE USDA

Cat. No. 271710 Dehydrated – 500 g

Difco™ SBG Sulfa Enrichment

USDA

Cat. No. 271510 Dehydrated – 500 g

BiGGY Agar

Intended Use

BiGGY (Bismuth Sulfite Glucose Glycine Yeast) is a selective and differential medium used in the detection, isolation and presumptive identification of *Candida* species.

Summary and Explanation

BiGGY Agar is based on the formulation of Nickerson.¹ Nickerson developed the medium in 1953 following a study of sulfite reduction by *Candida* species.

Differentiation of *Candida* is based on growth patterns and pigmentation of isolated colonies. The bismuth sulfite acts as an inhibitory agent to suppress bacterial growth, which enables the recovery of isolated colonies of *Candida*. *Candida* species reduce the bismuth sulfite, resulting in pigmentation of colonies and, with some species, pigmentation in the surrounding medium.

Principles of the Procedure

Candida species, through a process of substrate reduction, produce sulfide and bismuth which combine to produce brown to black pigmented colonies and zones of dark precipitate in the medium surrounding colonies of some species. Dextrose and yeast extract provide the nutrients in the formulation.

NOTE: A decrease in pH is normal and does not affect performance.

Formula

BBL™ BiGGY Agar

Approximate Formula* Per Liter	
Bismuth Ammonium Citrate	5.0 g
Sodium Sulfite	3.0 g
Dextrose	10.0 g
Glycine	10.0 g
Yeast Extract	1.0 g
Agar	16.0 g

*Adjusted and/or supplemented as required to meet performance criteria.

User Quality Control

Identity Specifications

BBL™ BiGGY Agar

Dehydrated Appearance:	Medium fine, homogeneous, free of extraneous material.
Solution:	4.5% solution, soluble in purified water upon boiling. Solution is light to medium, cream yellow, hazy to cloudy.
Prepared Appearance:	Light to medium, cream yellow, hazy to cloudy.
Reaction of 4.5% Solution at 25°C:	pH 6.8 ± 0.2

Cultural Response

BBL™ BiGGY Agar

Prepare the medium per label directions. Inoculate with fresh cultures and incubate at 25 ± 2°C for 18-24 hours (3-5 days if necessary).

ORGANISM	ATCC™	RECOVERY	COLOR OF COLONIES/MEDIUM
<i>Candida albicans</i>	10231	Good	Brown to black/–
<i>Candida kefyri</i>	8553	Good	Reddish brown/–
<i>Candida tropicalis</i>	1369	Good	Brown to black, metallic sheen/Brown to black
<i>Escherichia coli</i>	25922	Partial to complete inhibition	–/–

Candida krusei
ATCC™ 14243

Candida albicans
ATCC™ 10231

