

## BRILLIANT GREEN AGAR W/ SULFAPYRIDINE (7299)

### Intended Use

**Brilliant Green Agar w/ Sulfapyridine** is used for the selective enrichment of *Salmonella* spp.

### Product Summary and Explanation

Salmonellosis continues to be an important public health problem. Infection with non-typhi *Salmonella* spp. often causes mild, self-limiting illness.<sup>1</sup> Illness results from consumption of raw, undercooked, or improperly processed foods contaminated with *Salmonella* spp. Many cases of *Salmonella* related gastroenteritis result from improper handling of poultry products.

Brilliant Green Agar was first described by Kristensen et al.<sup>2</sup> and later modified by Kauffmann.<sup>3</sup> The outstanding selectivity of this medium permits the use of moderately heavy inocula evenly distributed over the surface. The addition of sulfonamides into Brilliant Green Agar further inhibits *Escherichia coli* and *Proteus* spp. Osborne and Stokes used 0.1% Sodium Sulfapyridine to enhance the recovery of *Salmonella* from whole egg and egg yolk.<sup>4,5</sup>

### Principles of the Procedure

Enzymatic Digest of Casein and Enzymatic Digest of Animal Tissue are the carbon and nitrogen source used for general growth requirements in this medium. Yeast Extract supplies B-complex vitamins. Lactose and Sucrose are the carbohydrates. In the presence of Phenol Red, a pH indicator, non-lactose and/or non-sucrose-fermenting *Salmonella* spp. will produce pink to red colonies. Sodium Sulfapyridine and Brilliant Green are the selective agents, inhibiting Gram-positive organisms and many Gram-negative bacteria, except *Salmonella*. Sodium Chloride maintains the osmotic balance. Agar is the solidifying agent.

### Formula / Liter

Yeast Extract .....	3 g
Enzymatic Digest of Casein .....	5 g
Enzymatic Digest of Animal Tissue .....	5 g
Sodium Chloride .....	5 g
Lactose .....	10 g
Sucrose .....	10 g
Brilliant Green .....	0.0125 g
Phenol Red .....	0.08 g
Sodium Sulfapyridine .....	1 g
Agar .....	20 g

Final pH: 6.9 ± 0.2 at 25°C

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

### Precautions

1. For Laboratory Use.
2. IRRITANT. Irritating to eyes, respiratory system, and skin. Inhalation of powder may cause respiratory irritation.

### Directions

1. Suspend 59 g of the medium in one liter of purified water.
2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
3. Autoclave at 121°C for 15 minutes. Avoid overheating.

### Quality Control Specifications

**Dehydrated Appearance:** Powder is homogeneous, free flowing, and beige to beige with a green tint.

**Prepared Appearance:** Prepared medium is brown-green to red-brown with a green tint, and trace to slightly hazy to slightly opalescent.

**Expected Cultural Response:** Cultural response on Brilliant Green Agar w/ Sulfapyridine at 35 ± 2°C and examined for growth after 18 - 24 hours incubation.

Microorganism	Approx. Inoculum (CFU)	Expected Results	
		Growth	Reaction
<i>Escherichia coli</i> ATCC® 25922	1000	Partial to complete inhibition	Yellow to green colonies
<i>Salmonella choleraesuis</i> ATCC® 13076	10 – 300	Fair to good	Pink colonies
<i>Salmonella typhi</i> ATCC® 19430	1000	None to poor	Pink colonies
<i>Salmonella typhimurium</i> ATCC® 14028	10 - 300	Fair to good	Pink colonies
<i>Staphylococcus aureus</i> ATCC® 25923	1000	Inhibited	----

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

### Test Procedure

Refer to appropriate references for instructions on specific material being tested for *Salmonella*.<sup>1,5-8</sup>

### Results

Refer to appropriate references and procedures for results.

### Storage

Store sealed bottle containing the dehydrated medium at 2 - 30°C. 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.

### 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 when stored as directed.

### Limitation of the Procedure

Due to varying nutritional requirements, some strains may be encountered that grow poorly or fail to grow on this medium.

### Packaging

Brilliant Green Agar w/ Sulfapyridine	Code No.	7299A	500 g
		7299B	2 kg
		7299C	10 kg

### References

1. **Marshall, R. T. (ed.)**. Standard methods for the examination of dairy products, 16<sup>th</sup> ed., American Public Health Association, Washington, D.C.
2. **Kristensen, M., V. Lester, and A. Jurgens**. 1925. On the use of trypsinized casein, bromthymol blue, bromcresol purple, phenol red and brilliant green for bacteriological nutrient media. Br. J. Exp. Pathol. **6**:291.
3. **Kauffmann, F.** 1935. Weitere Erfahrungen mit den kombinierten Anreicherungsverfahren für Salmonellabacillen. Z. Hyg. Infektionskr. **117**:26.
4. **Osborne, W. W., and J. L. Stokes**. 1955. The determinations of *Salmonellae* in foods. Ottawa: Food and Drug Laboratories.
5. [www.fda.gov/Food/ScienceResearch/LaboratoryMethods/BacteriologicalAnalyticalManualBAM/default.htm](http://www.fda.gov/Food/ScienceResearch/LaboratoryMethods/BacteriologicalAnalyticalManualBAM/default.htm).
6. **Cunniff, P. (ed.)**. 1995. Official Methods of Analysis AOAC International, 16<sup>th</sup> ed. AOAC International, Gaithersburg, MD.
7. **Vanderzant, C., and D. F. Splittstoesser (eds.)**. Compendium of methods for the microbiological examination of foods, 3<sup>rd</sup> ed. American Public Health Association, Washington, D.C.
8. **Eaton, A. D., L. S. Clesceri, and A. E. Greenberg (eds.)**. 1995. Standard methods for the examination of water and wastewater, 19<sup>th</sup> ed. American Public Health Association, Washington, D.C.

### Technical Information

Contact Acumedia Manufacturers, Inc. for Technical Service or questions involving dehydrated culture media preparation or performance at (517)372-9200 or fax us at (517)372-2006.