

## Brilliant Green Bile 2% Broth (NCM0048)

### Intended Use

Brilliant Green Bile Broth 2% is used for the detection of coliform bacteria in water, food, and dairy products in a laboratory setting. Brilliant Green Bile Broth 2% is not intended for use in the diagnosis of disease or other conditions in humans

### Description

The coliform group of bacteria includes aerobic and facultative anaerobic, Gram-negative, non-sporeforming bacilli that ferment lactose and form acid and gas at 35°C within 48 hours. Members of the Enterobacteriaceae comprise the majority of this group, but organisms such as *Aeromonas* spp. may also be included. Procedures to detect and confirm coliforms are used in testing water, foods, dairy products and other materials. Brilliant Green Bile Broth 2% is used to confirm a positive presumptive test result.

Brilliant Green Bile Broth 2% is also referred to as Brilliant Green Bile Broth, Brilliant Green Lactose Broth, Brilliant Green Lactose Bile Broth and Brilliant Green Lactose Bile Broth, 2%.

### Typical Formulation

Enzymatic Digest of Gelatin	10.0 g/L
Lactose	10.0 g/L
Ox Bile	20.0 g/L
Brilliant Green	0.0133 g/L

Final pH: 7.2 ± 0.2 at 25°C Formula may be adjusted and/or supplemented as required to meet performance specifications.

### Precaution

Refer to SDS

### Preparation

1. Dissolve 40 g of the medium in one liter of purified water until evenly dispersed.
2. Mix thoroughly.
3. Distribute into bottles or tubes with inverted Durham tubes.
4. Autoclave at 121°C for 15 minutes. To avoid entrapment of bubbles in the fermentation tubes, allow the autoclave to cool at least to 75°C before opening.

### Test Procedure

Refer to appropriate references for specific instructions for the material being tested.

1. Subculture from a presumptive positive coliform specimen in Lauryl Sulfate Broth or from typical coliform colonies on Violet Red Bile Agar to tubes of Brilliant Green Bile Broth 2%.
2. Incubate at 35°C for 48 ± 2 hours.
3. Examine for bubbles (gas) in the fermentation tube.

### Quality Control Specifications

**Dehydrated Appearance:** Powder is homogeneous, free flowing, and beige.

**Prepared Appearance:** Prepared medium is green and clear with no precipitate.

**Expected Cultural Response:** Cultures listed below are inoculated into Brilliant Green Bile 2% Broth and incubated at appropriate atmosphere and temperatures and examined for growth at 22 – 50 hours.

# Technical Specification Sheet



Microorganism	Approx. Inoculum (CFU)	Expected Results	
		Growth	Gas
<i>Enterococcus faecalis</i> ATCC® 29212	~10 <sup>4</sup>	Partial to complete inhibition	Negative
<i>Escherichia coli</i> ATCC® 25922	10-100	Growth	Positive
<i>Escherichia coli</i> ATCC® 8739	10-100	Growth	Positive
<i>Klebsiella pneumoniae</i> ATCC® 13883	10-100	Growth	Positive
<i>Salmonella typhimurium</i> ATCC® 14028	~10 <sup>4</sup>	Growth	Negative
<i>Citrobacter freundii</i> ATCC® 43864	10-100	Growth	Positive

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

## **Results**

Positive: Bubbles (gas) in fermentation tube.

Negative: No bubbles (gas) in fermentation tube.

## **Expiration**

Refer to expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if the 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.

## **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**

1. [www.fda.gov/Food/ScienceResearch/LaboratoryMethods/BacteriologicalAnalyticalManualBAM/default.htm](http://www.fda.gov/Food/ScienceResearch/LaboratoryMethods/BacteriologicalAnalyticalManualBAM/default.htm).
2. Cunniff, P. (ed.). 1995. Official Methods of Analysis AOAC International, 20<sup>th</sup> ed. AOAC International, Gaithersburg, MD.
3. Vanderzant, C., and D. F. Splittstoesser (eds.). Compendium of methods for the microbiological examination of foods, 4<sup>th</sup> ed. American Public Health Association, Washington, D.C.
4. Marshall, R. T. (ed.). 2004. Standard methods for the examination of dairy products, 17<sup>th</sup> ed., American Public Health Association, Washington, D.C.
5. Eaton, A. D., L. S. Clesceri, and A. E. Greenberg (eds.). 2017. Standard methods for the examination of water and wastewater, 23<sup>rd</sup> ed. American Public Health Association, Washington, D.C.

