



**BBL™ Brilliant Green Bile Broth,
2% with Durham Tube
L007443 • Rev. 08 • May 2006**

QUALITY CONTROL PROCEDURES

I INTRODUCTION

Brilliant Green Bile Broth, 2% is used for the detection of coliforms.

II PERFORMANCE TEST PROCEDURE

1. Inoculate representative samples with the cultures listed below.
 - a. Using a 0.01 mL calibrated loop, inoculate the tube samples using 10⁻¹ dilutions of 18- to 24-h **Trypticase™** Soy Broth cultures.
 - b. Incubate tubes with loosened caps at 35 ± 2°C in an aerobic atmosphere.
 - c. Include **Trypticase** Soy Broth tubes as nonselective controls for the *Enterococcus* and *Staphylococcus* strains.
2. Examine tubes after 18–24 and 42–48 h for growth, selectivity and gas production. Gas production is defined as the presence of gas in the inverted Durham tube with a corresponding effervescence produced when the tube is gently shaken.
3. Expected Results

Organisms	ATCC™	Recovery	Gas
* <i>Escherichia coli</i>	25922	Moderate to heavy growth	+
<i>Escherichia coli</i>	11775	Moderate to heavy growth	+
* <i>Enterobacter aerogenes</i>	13048	Moderate to heavy growth	+
<i>Enterococcus faecalis</i>	29212	Inhibition (partial to complete)	
* <i>Staphylococcus aureus</i>	25923	Inhibition (complete)	

*Recommended organism strain for User Quality Control.

III ADDITIONAL QUALITY CONTROL

1. Examine tubes as described under "Product Deterioration."
2. Visually examine representative tubes to assure that any existing physical defects will not interfere with use.
3. Incubate uninoculated representative tubes aerobically at 20–25°C and 30–35°C and examine after 7 days for microbial contamination.

PRODUCT INFORMATION

IV INTENDED USE

Brilliant Green Bile Broth, 2% is used for the detection of coliform organisms in foods, dairy products, water and wastewater as well as in other materials of sanitary importance.

V SUMMARY AND EXPLANATION

Brilliant Green Bile Broth, 2% is formulated according to AOAC¹ and APHA²⁻⁴ specifications for use in the confirmation of presumptive tests for coliforms.

VI PRINCIPLES OF THE PROCEDURE

Brilliant Green Bile Broth, 2% contains two inhibitors of both gram-positive and selected gram-negative organisms, namely, oxgall and brilliant green dye. Organisms, primarily coliforms, which are resistant to the action of the inhibitors and which ferment the lactose, are able to replicate in this medium. Fermentation is detected by gas production.

VII REAGENTS

Brilliant Green Bile Broth, 2%

Approximate Formula* Per Liter Purified Water

Oxgall, dehydrated	20.0 g
Lactose	10.0 g
Pancreatic Digest of Gelatin	10.0 g
Brilliant Green	13.3 mg

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

Warnings and Precautions: For Laboratory Use.

Tubes with tight caps should be opened carefully to avoid injury due to breakage of glass.

Observe aseptic techniques and established precautions against microbiological hazards throughout all procedures. After use, prepared tubes, specimen containers and other contaminated materials must be sterilized by autoclaving before discarding.

Storage Instructions: On receipt, store tubes in the dark at 2–8°C. Avoid freezing and overheating. Do not open until ready to use. Minimize exposure to light. Tubed media stored as labeled until just prior to use may be inoculated up to the expiration date and incubated for the recommended incubation times. Allow the medium to warm to room temperature before inoculation.

Product Deterioration: Do not use tubes if they show evidence of microbial contamination, discoloration, drying or other signs of deterioration.

VIII SPECIMEN COLLECTION AND HANDLING

Consult appropriate texts for detailed information and recommended procedures.¹⁻⁴

IX PROCEDURE

Material Provided: Brilliant Green Bile Broth, 2% with Durham Tube

Materials Required But Not Provided: Ancillary culture media, reagents, quality control organisms and laboratory equipment as required.

Test Procedure: Observe aseptic techniques.

For the detailed procedures for use of this medium in confirmatory testing for coliforms, refer to the various compendia.¹⁻⁴

User Quality Control: See "Quality Control Procedures."

X RESULTS

Gas production within 48 ± 2 h is considered positive evidence of fermentation by coliform bacilli.

Detailed results for the enumeration of coliforms using Brilliant Green Bile Broth, 2% are discussed in the various compendia of methods of microbiological examination of foods, dairy products and water and wastewater.¹⁻⁴

XI LIMITATIONS OF THE PROCEDURE

For identification, organisms must be in pure culture. Morphological, biochemical, and/or serological tests should be performed for final identification. Consult appropriate texts for detailed information and recommended procedures.¹⁻⁴

XII AVAILABILITY

Cat. No.	Description
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221612	BBL™ Brilliant Green Bile Broth, 2% with Durham Tube, Pkg. of 10 size A tubes
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XIII REFERENCES

1. Horwitz, W. (ed.) 2000. Official methods of analysis of AOAC International, 17th ed., vol. 1. AOAC International, Gaithersburg, Md.
2. Downes, F.P. and K. Ito (ed.). 2001. Compendium of methods for the microbiological examination of foods, 4th ed. American Public Health Association, Washington, D.C.
3. Wehr, H.M. and J.H. Frank (ed.). 2004. Standard methods for the examination of dairy products, 17th ed. American Public Health Association, Washington, D.C.
4. Clesceri, L.S., A.E. Greenberg, and A.D. Eaton (ed.). 1998. Standard methods for the examination of water and wastewater, 20th ed. American Public Health Association, Washington, D.C.

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