

Lauryl Tryptose Broth with MUG

Lauryl Sulfate Broth with MUG

Intended Use

Lauryl Tryptose Broth with MUG and Lauryl Sulfate Broth with MUG, which are also known as Lauryl Sulfate Tryptose Broth with MUG (LST-MUG), are used for the detection of *Escherichia coli* in water, food and dairy samples by a fluorogenic procedure.

Summary and Explanation

E. coli is used as an indicator organism of unsanitary conditions. A number of selective media are recommended for use in enrichment,

presumptive identification and confirmatory procedures for demonstrating the presence of coliforms in material of sanitary importance. These procedures require the incubation of test media for up to 7 days.

The presence of the fluorogenic compound, MUG (4-methylumbelliferyl- β -D-glucuronide), in this medium permits the rapid detection of *E. coli* when the medium is observed for fluorescence using a long-wave UV light source, and further confirmation is not required.^{1,2} MUG detects anaerogenic strains which may not be detected in the conventional procedure.¹

User Quality Control

NOTE: Differences in the Identity Specifications and Cultural Response testing for media offered as both **Difco™** and **BBL™** brands may reflect differences in the development and testing of media for industrial and clinical applications, per the referenced publications.

Identity Specifications

Difco™ Lauryl Tryptose Broth with MUG

Dehydrated Appearance:	Light beige, free flowing, homogeneous.
Solution:	3.57% solution, soluble in purified water upon warming. Solution is light to medium amber, clear to very slightly opalescent.
Prepared Appearance:	Light to medium amber, clear to very slightly opalescent.
Reaction of 3.57% Solution at 25°C:	pH 6.8 ± 0.2

Cultural Response

Difco™ Lauryl Tryptose Broth with MUG

Prepare the medium per label directions. Inoculate and incubate at 35 ± 2°C for 24 ± 2 hours or longer, if necessary.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY	GAS	FLUO- RESCENCE
<i>Enterobacter aerogenes</i>	13048	30-100	Good	+	–
<i>Escherichia coli</i>	25922	30-100	Good	+	+
<i>Salmonella enterica</i> subsp. <i>enterica</i> serotype Typhimurium	14028	30-100	Good	–	–
<i>Staphylococcus aureus</i>	25923	3 × 10 ² -10 ³	Marked to complete inhibition	–	–

*Gas production positive within 48 ± 3 hours.

Identity Specifications

BBL™ Lauryl Sulfate Broth with MUG

Dehydrated Appearance:	Fine, homogeneous, free of extraneous material.
Solution:	3.57% solution, soluble in purified water. Solution is pale to light, tan to yellow, clear to slightly hazy.
Prepared Appearance:	Pale to light, tan to yellow, clear to slightly hazy.
Reaction of 3.57% Solution at 25°C:	pH 6.8 ± 0.2

Cultural Response

BBL™ Lauryl Sulfate Broth with MUG

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

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY	GAS	FLUO- RESCENCE
<i>Enterobacter aerogenes</i>	13048	10 ³ -10 ⁴	Good	+	–
<i>Enterococcus faecalis</i>	29212	10 ⁴ -10 ⁵	Partial to complete inhibition	–	–
<i>Escherichia coli</i>	25922	10 ³ -10 ⁴	Good	+	+

Feng and Hartman, using a MUG-containing medium in microtitration plates, reported β -glucuronidase activity in 96% of *E. coli*, 100% of enterotoxigenic *E. coli*, 17% of *Salmonella* spp., and 40% of *Shigella* spp., while all other genera tested were negative; most reactions occurred within 4 hours, but some weakly β -glucuronidase-positive strains required overnight incubation.¹ In the presence of large numbers of *Proteus vulgaris*, which may suppress gas production by *E. coli*, fluorescence due to *E. coli* was detected within 15 hours.¹

In a comparison, with conventional methods, Robison reported 94.8% agreement, a false-positive rate of 4.8%, attributable to the presence of streptococci in the samples, and no false-negatives.²

These media are included in the compendia of methods for the detection and enumeration of coliform organisms in food³ and dairy⁴ products and in the *Official Methods of Analysis of AOAC International*.⁵

Principles of the Procedure

Lactose is a source of energy for organisms. Peptone provides additional nutrients. The phosphate compounds provide buffering capacity. Sodium lauryl sulfate is inhibitory to many organisms but not for coliforms.

The substrate 4-methylumbelliferyl- β -D-glucuronide is hydrolyzed by an enzyme, β -glucuronidase, possessed by most *E. coli* and a few strains of *Salmonella*, *Shigella* and *Yersinia*, to yield a fluorescent end product, 4-methylumbelliferone.^{1,2} Development of fluorescence permits the detection of *E. coli* in pure or mixed cultures within 4-24 hours following inoculation and incubation of the medium.

Formulae

Difco™ Lauryl Tryptose Broth with MUG

Approximate Formula* Per Liter

Tryptose	20.0	g
Lactose	5.0	g
Dipotassium Phosphate	2.75	g
Monopotassium Phosphate	2.75	g
Sodium Chloride	5.0	g
Sodium Lauryl Sulfate.....	0.1	g
MUG (4-methylumbelliferyl- β -D-glucuronide)	0.05	g

BBL™ Lauryl Sulfate Broth with MUG

Approximate Formula* Per Liter

Pancreatic Digest of Casein	20.0	g
Lactose	5.0	g
Dipotassium Phosphate	2.75	g
Monopotassium Phosphate	2.75	g
Sodium Chloride	5.0	g
Sodium Lauryl Sulfate.....	0.1	g
4-Methylumbelliferyl- β -D-glucuronide	0.05	g

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

Directions for Preparation from Dehydrated Product

Difco™ Lauryl Tryptose Broth with MUG

1. Suspend 35.7 g of the powder in 1 L of purified water and warm slightly to dissolve completely.
2. Dispense into test tubes containing inverted fermentation vials.
3. Autoclave at 121°C for 15 minutes.
4. Test samples of the finished product for performance using stable, typical control cultures.

BBL™ Lauryl Sulfate Broth with MUG

1. Dissolve 35.7 g of the powder in 1 L of purified water.
2. Dispense in test tubes, containing inverted Durham tubes, in 10 mL amounts for testing samples of 1 mL or less. For testing 10 mL quantities of samples, dissolve 71.4 g of the powder in 1 L of purified water and distribute in 10 mL amounts. The concentration of the medium should be varied according to the size of the test samples.
3. Autoclave at 121°C, preferably for 12 minutes, but not exceeding 15 minutes. After autoclaving, cool the broth as quickly as possible.
4. Test samples of the finished product for performance using stable, typical control cultures.

NOTE: Refrigerated broth generally becomes cloudy or forms precipitates but clears upon warming to room temperature. However, clarity is not important because only gas production is significant.

Procedure

Follow standard methods for the test being performed.³⁻⁵ Observe the medium periodically during the incubation period for the development of fluorescence, using a long-wave UV light source (approximately 366 nm) as well as for characteristic growth and/or gas production.

Expected Results

The presence of *E. coli* is detected by the appearance of fluorescence throughout the tube.

References

1. Feng and Hartman. 1982. Appl. Environ. Microbiol. 43:1320.
2. Robison. 1984. Appl. Environ. Microbiol. 48:285.
3. Downes and Ito. 2001. Compendium of methods for the microbiological examination of foods, 4th ed. American Public Health Association, Washington, D.C.
4. Wehr and Frank (ed.). 2004. Standard methods for the examination of dairy products, 17th ed. American Public Health Association, Washington, D.C.
5. Horwitz (ed.). 2007. Official methods of analysis of AOAC International, 18th ed., online. AOAC International, Gaithersburg, Md.

Availability

Difco™ Lauryl Tryptose Broth with MUG

AOAC BAM COMPF SMD SMWW

Cat. No. 211740 Dehydrated –100 g
211744 Dehydrated – 500 g

BBL™ Lauryl Sulfate Broth with MUG

AOAC BAM COMPF SMD SMWW

Cat. No. 298076 Dehydrated –500 g