BD BBL™ Prepared Tubed Medium for Detection of Fecal Coliform Bacteria

EC Broth with Durham Tube

8806881JAA
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INTENDED USE

EC Broth with Durham tube is used in the detection of fecal coliform bacteria as an indication of fecal pollution of food and water from sources other than potable water. A Durham tube is provided to detect the production of gas.

SUMMARY AND EXPLANATION

Hajna and Perry developed EC Broth for improved selective detection of coliform bacteria and presumptive detection of *E. coli* as evidence of fecal coliforms in water, foods, shellfish, milk and other materials.[1,2]

Tennant et al. reported using EC Broth at an incubation temperature of 44.5°C for the estimation of *E. coli* densities in sea water and shellfish.[3]

Fishbein and Surkiewicz compared the recovery of *E. coli* from frozen foods and nutmeats using EC Broth incubated at 44.5 and 45.5°C.[4] The higher incubation temperature (45.5°C) enhanced the specificity of the test for *E. coli*, while the lower incubation temperature (44.5°C) provided a 4% greater recovery of *E. coli*.

EC Broth is used as part of the confirmatory test of the total coliform MPN (most probable number) procedure.[5,6] The EC Broth is inoculated with broth from positive presumptive tubes and incubated at 44.5 ± 0.2°C (for waters and shellfish) or 45.5 ± 0.2°C (for foods) for 24 ± 2 h. If gas is produced, the test is positive, indicating the presence of coliforms of fecal origin (usually *E. coli*).

PRINCIPLES OF THE PROCEDURE

Enzymatic digest of casein provides amino acids and other nitrogenous substances. Bile salts inhibit fecal streptococci and sporeformers. Phosphate buffer maintains the pH of the medium, and sodium chloride maintains the osmotic equilibrium.

Gas produced during lactose fermentation at an incubation temperature of 35 ± 2°C indicates the presence of coliform bacteria. Gas production at the elevated temperatures indicates the presence of fecal coliforms, usually *E. coli*. Growth (turbidity) at the elevated temperatures without gas production indicates a source other than the feces of warm-blooded animals. A Durham tube collects the gas produced during lactose fermentation.

REAGENTS

Formula:

EC Broth

Approximate Formula* Per Liter Purified Water

Pancreatic Digest of Casein	20.0 g
Lactose	5.0 g
Bile Salts	1.5 g
Dipotassium Phosphate	4.0 g
Monopotassium Phosphate	1.5 g
Sodium Chloride	5.0 g

*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.

Storage Instructions: On receipt, store tubes in the dark at 2 – 25°C. Avoid freezing or 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 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, precipitation, evaporation or other signs of deterioration.

SPECIMEN COLLECTION AND HANDLING

This medium is not for use directly with samples containing mixed microbial flora. Consult appropriate texts for information.[5,6]

Observe established precautions against microbiological hazards throughout all procedures. After use, sterilize prepared tubes and other contaminated materials by autoclaving.

PROCEDURE

Material Provided: EC Broth 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.

Inoculate EC Broth with a 3 mm loopful of broth from tubes of Lauryl Sulfate Broth showing gas production. Consult texts for information about the processing of water, food and other samples.[5,6]

Remove air bubbles from the Durham tube prior to incubation by inverting the test medium and gently tapping the side to dislodge the bubble. Carefully return the test medium to an upright position to avoid re-introducing air bubbles into the Durham tube.

Place the inoculated tubes in a water bath at 35 ± 2°C for detection of coliforms or at 44.5 ± 0.2°C (waters and shellfish) or 45.5 ± 0.2°C (food) for detection of fecal coliforms (*E. coli*) for 24 ± 2 h.

User Quality Control:

1. Examine the tubes for signs of deterioration as described under "Product Deterioration."
2. Check performance by inoculating a representative sample of tubes with pure cultures of stable control organisms that give known, desired reactions. Incubate one set of the inoculated tubes at 35 ± 2°C and a duplicate tube of the *E. coli* strain at 44 ± 2°C. The following test strains are recommended:

	EXPECTED RESULTS	
TEST STRAIN	35 ± 2°C	44 ± 2°C
Escherichia coli ATCC™ 25922	Growth, gas production.	Growth, gas production.
Salmonella enterica subsp. *enterica* serotype Typhimurium ATCC 13311	Growth, no gas produced.	
Staphylococcus aureus ATCC 25923	Inhibited.	

RESULTS

At an incubation temperature of 35 ± 2°C, growth (turbidity) with gas production is a positive reaction, indicating the presence of coliforms. Gas production at the elevated temperatures is a positive test for *E. coli*, indicating fecal pollution.

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.[5-9]

AVAILABILITY

Cat. No.	Description
295605	**BBL™** EC Broth with Durham Tube, Pkg. of 10 size A tubes.

REFERENCES

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