

# TECHNICAL DATA SHEET

## EC BROTH

### DETECTION AND ENUMERATION OF PRESUMPTIVE *ESCHERICHIA COLI*

#### 1 INTENDED USE

EC Broth (abbreviation for *Escherichia coli*) is a selective media used for the confirmation of *E. coli* in water, milk and other food products, as well as live shellfish.

The typical composition of the broth responds to that defined in the normalized method of presumptive *Escherichia coli* enumeration, MPN technique (NF ISO 7251).

#### 2 HISTORY

In 1943, the media was developed by Hajna, Perry and Darby for the detection of coliforms, and more specifically, for *E. coli* in food products. These authors had analyzed the results from 11 laboratories that tested a wide variety of water, milk and shellfish samples. The results obtained had demonstrated a very high specificity for the media to coliforms. In 1964, Fishbein and Surkiewicz used the EC broth for the confirmation of *Escherichia coli* in frozen products and nutmeats. They found that the test was optimal when the incubation was limited to a duration of 24 hours at 45,5 °C.

#### 3 PRINCIPLES

The media corresponds to the buffered lactose broth, with added bile salts, whose objective is to inhibit the development of spore-formers and enterococci, while at the same time promoting *Escherichia coli* growth.

The development of coliforms and notably *E. coli* is demonstrated through the appearance of turbidity associated with gas production in a Durham tube following lactose fermentation..

#### 4 TYPICAL COMPOSITION

The composition can be adjusted in order to obtain optimal performance.

For 1 liter of media :

- Tryptone .....	20,00 g
- Lactose.....	5,00 g
- Bile salts n°3 .....	1,50 g
- Dipotassium phosphate.....	4,00 g
- Monopotassium phosphate.....	1,50 g
- Sodium chloride .....	5,00 g

pH of the ready-to-use media at 25 °C : 6,8 ± 0,2.

#### 5 PREPARATION

- Dissolve 37,0 g of rehydrated media (BK162) in distilled or demineralized water.
- Mix slowly until complete dissolution.
- Dispense into 10 mL tubes, each containing a Durham tube.
- Sterilize in an autoclave at 121 °C for 15 minutes.
- After cooling, the Durham tubes should not contain trapped air.

✓ **Reconstitution :**  
37,0 g/L

✓ **Sterilization :**  
15 min at 121 °C

## 6 INSTRUCTIONS FOR USE

- From a tube of selective enrichment media, (Laurylsulfate BK010) presenting an opacity, cloudiness or gas production, inoculate a tube of EC broth with a loop.
- Incubate in a temperature-controlled water bath for 24 hours at  $44 \pm 1$  °C. In the absence of gas production, extend the incubation up to 48 hours.

✓ **Inoculation :**  
A loop of inoculum

✓ **Incubation :**  
24 h to 48 h at 44 °C

## 7 RESULTS

Observe the production of gas in the Durham tubes.

Should the EC broth present gas production, confirm the strain through the indole test in Peptone Water (BK084). Consider as positive for *Escherichia coli* all tubes that present a gas development in EC broth as well as being indole positive.

## 8 QUALITY CONTROL

**Dehydrated media :** white powder, free-flowing and homogeneous..

**Prepared media :** clear, limpid amber solution.

Typical culture response after 24 and 48 hours of incubation at 44 °C, qualitative method (NF EN ISO 11133) :

Microorganisms		Growth
<i>Escherichia coli</i>	WDCM 00012	Good, score 2
<i>Escherichia coli</i>	WDCM 00013	Good, score 2
<i>Pseudomonas aeruginosa</i>	WDCM 00025	Inhibited

## 9 STORAGE / SHELF LIFE

**Dehydrated media :** 2-30 °C.

The expiration date is indicated on the label.

**Prepared media in tubes (\*) :** 180 days at 2-25 °C.

(\*) Benchmark value determined under standard preparation conditions, following manufacturer's instructions.

## 10 PACKAGING

**Dehydrated media :**

500 g bottle ..... BK162HA

## 11 BIBLIOGRAPHY

Hajna, A.A., and Perry, C.A. 1943. Comparative study of presumptive and confirmative media for bacteria of the coliform group and for fecal streptococci. Am. J. Public Health, 33: 550-556.

Fishbein M.A, and Surkiewicz B.F.. 1964. Comparison of the recovery of *Escherichia coli* from frozen food and nutmeats by confirmatory incubation in EC-medium at 44.5 and 45.5°C. Appl. Microb., 12: 127-131.

NF ISO 7251. Juillet 2005. Microbiologie des aliments. Méthode horizontale pour la recherche et le dénombrement d'*Escherichia coli* présumés. Technique du nombre le plus probable.

## 12 ADDITIONAL INFORMATION

The information provided on the labels take precedence over the formulations or instructions described in this document and are susceptible to modification at any time, without warning.

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