

BROMOCRESOL PURPLE LACTOSE BROTH

DETECTION AND ENUMERATION OF COLIFORMS

1 INTENDED USE

Bromocresol Purple (BCP) Lactose broth is used as enrichment media for the enumeration of coliforms and of thermotolerant coliforms in water, according to the standard NF T90-413 (MPN method).

2 PRINCIPLES

The media is used in single and in double strength varieties.

Tryptone and meat extract supply nitrogen nutrients required for bacterial growth.

The fermentation of lactose is shown by the acidification of the medium that causes the pH indicator (bromocresol purple) to turn yellow, as well as by gas production in Durham tubes.

In order to confirm the presence of coliform bacteria, subcultures must be prepared using the appropriate confirmation media.

3 TYPICAL COMPOSITION

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

For 1 liter of media :

- Tryptone 5,0 g
- Meat extract..... 3,0 g
- Lactose..... 5,0 g
- Bromocresol purple 25,0 mg

pH of the ready-to-use media at 25 °C : 6,7 ± 0,2.

4 PREPARATION

Preparation of single strength media :

- Dissolve 13,0 g of dehydrated media (BK119) in 1 liter of distilled or demineralized water.
- Stir slowly until complete dissolution.
- Dispense 10 mL into 16 x 160 mm tubes each containing a Durham tube.
- Sterilize in an autoclave at 121 °C for 15 minutes.
- Cool to room temperature.

✓ **Reconstitution :**
13,0 g/L

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

Preparation of double strength media :

- Dissolve 26,0 g of dehydrated media (BK119) in 1 liter of distilled or demineralized water.
- Stir slowly until complete dissolution.
- Dispense 10 mL into 20 x 200 mm tubes each containing a Durham tube.
- Sterilize in an autoclave at 121 °C for 15 minutes
- Cool to room temperature.

✓ **Reconstitution :**
26,0 g/L

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

5 INSTRUCTIONS FOR USE

- Inoculate 3 tubes of double strength broth with 10 mL of the primary dilution.
- Inoculate 3 tubes of single strength broth with 1 mL of the primary dilution and its subsequent serial dilutions.
- Incubate at 30 ± 1 °C for 24 and 48 hours.

✓ **Inoculation** : MPN
10 mL in double strength broth
1 mL in single strength broth

✓ **Incubation** :
24 and 48 h at 30 ± 1 °C

NOTE

Insure that the Durham tubes contain no air before incubation.

6 RESULTS

The fermentation of lactose results in the appearance of cloudiness due to bacterial development, as well as the release of gas in the Durham tubes.

Re-inoculate the presumptive positive tubes into BGBB tubes for confirmation (BM011).

7 QUALITY CONTROL

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

Prepared media : violet, limpid solution.

Typical culture response after 48 hours of incubation à 30 °C, inoculum $\leq 10^2$ microorganisms (FD T90-461).

Microorganisms		Growth	Gas production (Durham tube)
<i>Escherichia coli</i>	WDCM 00012	Good, score 2	≥ 5 mm
<i>Staphylococcus aureus</i>	WDCM 00035	Partially inhibited, score 0-1	Negative

8 STORAGE / SHELF LIFE

Dehydrated media : 2-30 °C.

The expiration date is indicated on the label.

Prepared media in tubes (*) : 30 days at 2-8 °C.

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

9 PACKAGING

Dehydrated media :

500 g bottle BK119HA

10 BIBLIOGRAPHY

Rodier, J. 1984. L'analyse de l'eau. Dénombrement des coliformes, coliformes fécaux, et *Escherichia coli* présumés. Dunod 7ème Ed., 793-798.

NF T90-413. Octobre 1985. Essais des eaux. Recherche et dénombrement des coliformes et des coliformes thermotolérants. Méthode générale par ensemencement en milieu liquide (NPP).

11 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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