# E.COLI COLIFORMS CHROMOGENIC AGAR BASE (BOE)

**CAT Nº: 1491**

Selective medium for the simultaneous detection of *E.coli* and other coliforms in water and food samples

## FORMULA IN g/l

| | | | |
|---|---|---|---|
| Sodium Chloride | 5.00 | Sorbitol | 1.00 |
| Casein Peptone | 3.00 | X-Glucuronide | 0.20 |
| Disodium Hydrogenphosphate | 2.70 | Salmon-Gal | 0.20 |
| Sodium Dihygrogenphosphate | 2.20 | Tergitol-7 | 0.15 |
| Sodium Pyruvate | 1.00 | Bacteriological Agar | 10.00 |
| Tryptophan | 1.00 | | |

**Final pH 6.8 ± 0.2 at 25°C**

## PREPARATION

Suspend 13.25 grams of the medium in 500 ml of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. AVOID OVERHEATING. DO NOT AUTOCLAVE. Cool to 45-50ºC and aseptically add one vial of the E.coli-coliforms supplement (Cat. 6041) reconstituted in 5 ml of sterile distilled water. Homogenize gently and dispense into Petri dishes. The prepared medium should be stored at 8-15°C, protected from light. The color of the prepared medium is amber.

The dehydrated medium should be homogeneous, free-flowing and beige in color. If there are any physical changes, discard the medium.

**E.coli-coliforms Supplement (Cat. 6041)**
(1 vial for 500 ml of the medium)
Vancomycin .....2.5 mg
Cefsulodine..... 2.5 mg

## USES

E.COLI-COLIFORMS CHROMOGENIC AGAR BASE is a selective medium for the detection of *E.coli* and other coliforms in waters and foods.

The interaction of ingredients in the medium, such as peptone, sorbitol, etc, grants a quick colony growth, including infectious Coliforms. Tergitol-7 inhibits Gram positive bacteria. Sodium chloride maintains the osmotic balance and Phosphate salts act as a buffer system. Bacteriological agar is the solidifying agent. The chromogenic mixture contains chromogenic substrates as Salmon-GAL and X-glucuronide. Coliform enzymes produced, such as galactosidase and glucuronidase, cleave these substrates, resulting in the different coloration of certain bacteria colonies.
The ß-D-galactosidase cleaves Salmon-GAL substrate, and gives a salmon to red color to the coliform colonies.
The cleaves both substrates Salmon-Gal and X-glucuronide, giving a dark blue to violet color to the colonies, easily distinguishable from other coliform colonies that have a salmon to red color.
The addition of tryptophan to the medium allows the performance of the Indole test for further *E. coli* confirmation.

Inoculate and incubate at 36 ± 2°C for 18 - 24 hours. Incubate until 24 hours to observe possible retarded β-Galactoside and β-Glucurinidase reactions.

Note: Some *Shigella* strains contains the enzyme ß-D-glucuronidase and can grow asl ight blue colonies.
The negative *E. coli* ß-glucuronidase colonies are colorless, e.g. *E. coli* O157:H7.

## MICROBIOLOGICAL TEST

The following results were obtained in the performance of the medium from type cultures after incubation at a temperature of 36 ± 2°C and observed after 21±3 hours.

| Microorganisms | Growth | Colony Color | Inóculo (cfu/ml) | Recovery rate % |
|---|---|---|---|---|
| *Escherichia coli* ATCC 25922 | Good | Blue-dark violet | $10^2$ -$10^3$ | ≥80 |
| *Escherichia coli* ATCC 8739 | Good | Blue-dark violet | $10^2$ -$10^3$ | ≥80 |
| *Citrobacter freundii* ATCC 8090 | Good | Salmon | $10^2$ -$10^3$ | ≥80 |
| *Salmonella enteritidis* ATCC 13076 | Good | Colorless | $10^2$ -$10^3$ | ≥50 |
| *Enterococcus faecalis* ATCC 19433 | Null | - | $>10^5$ | ≤0.01 |

## BIBLIOGRAPHY

Alonso, J.L. Soriano, K., Amoros I., Ferrus, M.A. 1998 Cevartitatine determination of *E. coli* and fecal coliforms in water using a chromogenic medium.

BOLETÍN OFICIAL DEL ESTADO. Num. 78 Martes 31 de marzo de 2009 Sec. I. Pág. 30417. Orden SCO/778/2009, de 17 de marzo, sobre métodos alternativos para el análisis microbiológico del agua de consumo humano.

## STORAGE

Once opened keep powdered medium closed to avoid hydration.