



## HiColiform Broth, Modified

M1850

HiColiform Broth, Modified is used for the detection and confirmation of *Escherichia coli* and total coliforms from water samples, using a combination of chromogenic and fluorogenic substrates.

### Composition\*\*

| Ingredients                    | Gms / Litre |
|--------------------------------|-------------|
| Peptone                        | 5.000       |
| Sodium chloride                | 5.000       |
| Potassium sulfate              | 1.000       |
| Dipotassium hydrogen phosphate | 4.000       |
| Potassium dihydrogen phosphate | 1.000       |
| Sodium lauryl sulphate         | 0.100       |
| Sodium puruvate                | 1.000       |
| Chromogenic substrate          | 0.100       |
| Fluorogenic substrate          | 0.100       |
| IPTG                           | 0.100       |
| Final pH ( at 25°C)            | 6.8±0.2     |

\*\*Formula adjusted, standardized to suit performance parameters

### Directions

Suspend 17.4 grams in 1000 ml distilled water. Heat if necessary to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Mix well and dispense as desired.

### Principle And Interpretation

HiColiform Broth, Modified was designed for detection and confirmation of *Escherichia coli* and total coliforms from water samples using a combination of chromogenic and fluorogenic substrates. *Escherichia coli* can be distinguished from other coliforms by its unique ability to fluoresce in the presence of fluorogenic substrate (1, 2). The fluorogenic substrate is split by enzyme beta-glucuronidase especially present in *Escherichia coli*. The reaction is indicated by the development of a blue fluorescence under UV light. The presence of total coliforms is indicated by blue-green colourations due to the cleavage of the chromogenic substrate. IPTG amplifies enzyme synthesis and increases the activity of beta-galactosidase.

Peptone provides essential growth nutrients and is useful for the simultaneous detection of indole production. The phosphate salts provide buffering action for rapid growth of coliforms. Sodium chloride helps to maintain the osmotic balance. Sodium lauryl sulphate makes the medium selective by inhibiting accompanying microflora, especially the gram-positive organisms.

### Quality Control

#### Appearance

Cream to yellow homogeneous free flowing powder

#### Colour and Clarity of prepared medium

Light yellow coloured, clear to slightly opalescent solution in tubes

#### Reaction

Reaction of 1.74% w/v aqueous solution at 25°C. pH : 6.8±0.2

#### pH

6.60-7.00

#### Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

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| Organism                                 | Inoculum (CFU) | Growth    | Colour of medium | Fluorescence (under uv) |
|--|----------------|-----------|------------------|-------------------------|
| <b>Cultural Response</b>                 |                |           |                  |                         |
| <i>Enterobacter aerogenes</i> ATCC 13048 | 50-100         | luxuriant | blue-green       | negative reaction       |
| <i>Escherichia coli</i> ATCC 25922       | 50-100         | luxuriant | blue-green       | positive reaction       |

### Storage and Shelf Life

Store dehydrated and prepared medium at 2-8°C in tightly closed container. Use before expiry date on the label.

### Reference

- 1.Feng P.C.S. and Hartman P.A. ,1982, J.Appl. Environmental Microbiol. 43. 1320-1323.
- 2.Harsen W., and Yourassowsky, 1984, J. Clin. Microbiol. 20. 1177-1179.

Revision : 2 / 2015

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