



HiCrome™ Rapid ECC Broth

M2011

Intended use

Recommended for rapid detection of *Escherichia coli* and other *Enterobacteriaceae* from water samples.

Composition**

Ingredients	g / L
Peptone special	24.000
Sodium chloride	5.000
Disodium hydrogen phosphate	1.000
Sodium thiosulphate	5.000
Ferric citrate	1.000
Lactose	5.000
Phenol red	0.018
Selective mix	1.500
Chromogenic substrate	3.830
Final pH (at 25°C)	7.4±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 46.35 gram in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely.

DO NOT AUTOCLAVE. Cool to 45-50°C. Dispense into sterile tubes or flasks as desired.

Principle And Interpretation

HiCrome™ Rapid ECC Broth is designed for detection and confirmation of *Escherichia coli* and other coliforms from water samples. The major microbial water contaminants are coliforms that includes *Escherichia coli*, *Klebsiella pneumoniae*, *Salmonella*, *Citrobacter*, *Vibrio* and *Pseudomonas* (1). This test was designed for the rapid detection and differentiation of these organisms.

Peptone special provides nitrogen and carbon source, long chain amino acids, vitamins and other essential growth nutrients. Phosphates buffer the medium. Lactose is the fermentable carbohydrate and phenol red is the indicator. Lactose fermenting organisms gives yellow colour to the medium while lactose non-fermenters gives pink to red colour. The chromogenic substrate is used to detect the presence of β -D-glucuronidase produced by *E.coli* thus imparting blue colour to the medium. However since *E.coli* also ferments lactose, the presence of *E.coli* is indicated by bluish green to green colour. The detection of H_2S production is enhanced by the presence of specific H_2S detectors. The medium turns black in case of H_2S producers such as *Salmonella*, *Citrobacter* etc are present. The phosphate salts provide buffering action for rapid growth of coliforms. Sodium chloride helps to maintain the osmotic balance. Selective mix present in the medium suppresses the growth of gram positive microorganisms. Recovery of these pathogens is faster and reliable.

Type of specimen

Water samples

Specimen Collection and Handling:

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards(2). After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations :

1. 97% of *E.coli* are β glucuronidase positive, and will give green colour. Strains producing less amount of β glucuronidase enzyme will give yellow to yellowish green colour to the medium.
2. Further biochemical and serological test are necessary for confirmation.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Light yellow to pink homogeneous free flowing powder

Colour and Clarity of prepared medium

Red coloured clear solution in tubes

Reaction

Reaction of 4.63% w/v aqueous solution at 25°C. pH : 7.4±0.2

pH

7.20-7.60

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 12-18 hours.

Organism	Inoculum (CFU)	Growth	Colour change in medium
<i>Escherichia coli</i> ATCC 25922 (00013*)	50-100	luxuriant	green
<i>Klebsiella pneumoniae</i> ATCC 13883 (00097*)	50-100	luxuriant	yellow
<i>Citrobacter freundii</i> ATCC 8090	50-100	luxuriant	black
<i>Salmonella</i> Typhimurium ATCC 14028 (00031*)	50-100	luxuriant	black
<i>Enterococcus faecalis</i> ATCC 29212 (00087*)	≥10 ⁴	Inhibition	-
<i>Staphylococcus aureus</i> subsp. <i>aureus</i> ATCC 25923 (00034*)	≥10 ⁴	Inhibition	-

Key: (*) Corresponding WDCM numbers

Storage and Shelf Life

Store between 15-25°C in a tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (3,4).

Reference

1. Methods for Examination of Waters and Associated Materials, Environment Agency, 1998, Standing Committee of Analysts.
2. Lipps WC, Braun-Howland EB, Baxter TE, eds. Standard methods for the Examination of Water and Wastewater, 24th ed. Washington DC:APHA Press; 2023.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
4. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

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Disclaimer :

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