



## HiCrome™ EC Broth w/ RUG

M2073

### Intended Use:

Recommended for detection of *Escherichia coli* in water and food samples by a chromogenic and fluorogenic method.

### Composition\*\*

Ingredients	Gms / Litre
Yeast extract	2.000
Acicase #	1.000
Buffers	4.500
Sodium chloride	0.500
Salts	2.450
Chromogenic mixture	0.112
Final pH ( at 25°C)	7.0±0.2

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

# Equivalent to Casein acid hydrolysate

### Directions

Suspend 10.56 grams in 1000 ml purified / distilled water. Heat to boiling to dissolve the medium completely. DO NOT AUTOCLAVE. Cool to 45-50°C. Dispense into sterile tubes or flasks as desired.

### Principle And Interpretation

*Escherichia coli* is a member of faecal coliform group of bacteria. It is a member of the indigenous faecal flora of warm blooded animals. *E.coli* is considered a specific indicator of faecal contamination and the possible presence of enteric pathogens. *E.coli* can be reliably detected with media that contain a chromogenic or fluorogenic substrate for beta-glucuronidase, an enzyme that occurs almost exclusively in *E. coli*.

Resorufin-beta-D-glucuronic acid methyl ester (RUG) is a highly sensitive chromogenic and fluorogenic indicator for *E.coli*. In contrast to MUG, RUG is more specific and does not require fluorescent detection. The released dye Resorufin itself gives intense pink color which can be visually detected. Additional confirmation can be done by observation of fluorescence under UV light.

Yeast extract and Acicase provides carbonaceous, nitrogenous substances, long chain amino acids, vitamins and other essential nutrients. Sodium chloride maintains osmotic equilibrium. The medium has a strong buffering system to control the pH in the medium Sodium lauryl sulphate inhibit gram-positive bacteria especially *Staphylococcus*, *Bacillus* species and faecal *Streptococcus*.

\$ Resorufin-beta-D-glucuronic acid methyl ester (RUG) is a patent of BIOSYNTH.

### Type of specimen

Food sample; Water samples

### Specimen Collection and Handling

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (4).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (1).

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. Slight variation in intensity of colour may be observed depending on the isolates.

## 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

Pale yellow to orange homogeneous free flowing powder

### Colour and Clarity of prepared medium

Yellow coloured clear solution with slight precipitate.

### Reaction

Reaction of 1.05% w/v aqueous solution at 25°C. pH : 7.0±0.2

### pH

6.80-7.20

### Cultural Response

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

Organism	Inoculum (CFU)	Growth	Colour of medium	*Fluorescence (at 366 nm)
<i>Escherichia coli</i> ATCC 25922 (00013*)	50-100	luxuriant	bright pink	positive, throughout the tube
<i>Escherichia coli</i> ATCC 10536	50-100	luxuriant	bright pink	positive throughout the tube
<i>Citrobacter freundii</i> ATCC 8090	50-100	luxuriant	pale orange	negative
<i>Salmonella</i> Enteritidis ATCC 10376 (00030*)	50-100	luxuriant	pale orange	negative
<i>Staphylococcus aureus</i> subsp. <i>aureus</i> ATCC 25923 (00034*)	≥10 <sup>4</sup>	inhibited	no colour change	
<i>Bacillus subtilis</i> subsp. <i>spizizenii</i> ATCC 6633 (00003*)	≥10 <sup>4</sup>	inhibited	no colour change	

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 (2,3).

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## Reference

1. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.
2. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
3. 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.
4. Salfinger Y., and Tortorella M.L., 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.

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### Disclaimer :

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