



## HiCrome™ EC O157:H7 Agar, Modified

M1574A

### Intended Use

Recommended for isolation and differentiation of *Escherichia coli* O157:H7 from food and environmental samples.

### Composition\*\*

| Ingredients            | Gms / Litre |
|------------------------|-------------|
| Tryptone               | 8.000       |
| Sorbitol               | 7.000       |
| Bile salts mixture     | 1.500       |
| Sodium lauryl sulphate | 0.100       |
| Chromogenic mixture    | 0.250       |
| Agar                   | 12.000      |
| Final pH ( at 25°C)    | 6.8±0.2     |

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

### Directions

Suspend 28.85 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45- 50°C. Mix well and pour into sterile Petri plates. This medium can be made more selective by aseptically adding 0.25 ml of rehydrated contents of one vial of FD052 (1% Potassium Tellurite Solution) to 1000 ml molten and cooled medium (45-50°C).

### Principle And Interpretation

*Escherichia coli* O157:H7 belongs to the Enterohemorrhagic *Escherichia coli* (EHEC) group and it predominates as a food borne pathogen. *E.coli* O157: H7 was first recognized as a human pathogen in 1982 when two outbreaks of hemorrhagic colitis were associated with consumption of undercooked ground beef that has been contaminated with this organism (1).

HiCrome™ EC O157:H7 Agar is a chromogenic medium recommended for the isolation and differentiation of *E.coli* O157:H7 from food and environmental samples. HiCrome™ EC O157:H7 Agar is based on the formulation described by Rappaport and Henigh (2). The medium contains sorbitol as a fermentable carbohydrate and a chromogenic mixture instead of lactose and indicator dyes respectively. The chromogenic substrate is specifically and selectively cleaved by *Escherichia coli* O157: H7 resulting in a dark purple to magenta coloured moiety. *E.coli* give bluish green coloured colonies.

Tryptone provides carbonaceous, nitrogenous and growth nutrients. Sodium chloride maintains osmotic equilibrium. Bile salts mixture and Sodium lauryl sulphate inhibits gram-positive organisms. Potassium tellurite selects the serogroups and inhibits *Aeromonas* species and *Providencia* species.

### Type of specimen

Food

### Specimen Collection and Handling:

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (3).

### Warning and Precautions:

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

### Limitations :

Due to variable nutritional requirements, some strains show poor growth on this medium.

## 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

Cream to yellow homogeneous free flowing powder

### Gelling

Firm, comparable with 1.2% Agar gel.

### Colour and Clarity of prepared medium

Light amber coloured, clear to slightly opalescent gel forms in Petri plates

### Reaction

Reaction of 2.88% 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.

| Organism  | Inoculum (CFU)   | Growth    | Recovery | Colour of Colony         |
|---|------------------|-----------|----------|--------------------------|
| <i>Bacillus subtilis</i> ATCC 6633 (00003*)       | ≥10 <sup>3</sup> | inhibited | 0 %      |                          |
| <i>Escherichia coli</i> O157:H7 (NCTC 12900)      | 50-100           | luxuriant | ≥50%     | dark purple-magenta      |
| <i>Escherichia coli</i> ATCC 25922 (00013*)       | 50-100           | luxuriant | ≥50%     | bluish green             |
| <i>Klebsiella pneumoniae</i> ATCC 13883           | 50-100           | luxuriant | ≥50%     | colourless-mauve, mucoid |
| <i>Pseudomonas aeruginosa</i> ATCC 27853 (00025*) | 50-100           | luxuriant | ≥50%     | colourless               |
| <i>Staphylococcus aureus</i> ATCC 25923 (00034*)  | ≥10 <sup>3</sup> | inhibited | 0%       |                          |

Key : \*Corresponding WDCM numbers.

## Storage and Shelf Life

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

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 clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (4,5).

## Reference

- Downes F. P. and Ito K., (Ed.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., American Public Health Association, Washington, D.C.
- Rappaport F. and Henigh E., 1952, J. Clin. Pathol., 5:361.

3. Downes F. P. and Ito K., (Ed.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., American Public Health Association, Washington, D.C.
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2<sup>nd</sup> Edition.
5. 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.

Revision : 01 / 2017

**Disclaimer :**

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