

## HiCrome™ E. coli Agar Plate

MP1295I

### Intended Use:

Recommended for the detection and enumeration of *Escherichia coli* in foods without further confirmation on membrane filter or by indole reagent. The composition and performance of this media are as per specifications laid down in in ISO 16649-2:1999.

### Composition\*\*

Ingredients	Gms / Litre
Tryptone	20.000
Bile salts mixture	1.500
X-Glucuronide	0.075
Agar	15.000
Final pH ( at 25°C)	7.2±0.2

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

### Directions

Either streak, inoculate or surface spread the test inoculum (50-100 CFU) aseptically on the plate.

### Principle And Interpretation

HiCrome™ E.coli Agar is based on Tryptone Bile Agar to detect *Escherichia coli* in foods (1), where recovery of *E.coli* is faster, more reliable and accurate. Most of the *E.coli* strains can be differentiated from other coliforms by the presence of enzyme glucuronidase, which is highly specific for *E.coli* (2). The chromogenic agent X-glucuronide used in this medium helps to detect glucuronidase activity of *E.coli*. *E.coli* cells absorb X-glucuronide and the intracellular glucuronidase enzyme splits the bond between the chromophore and the glucuronide. The released chromophore gives bluish green colouration to the *E.coli* colonies. The formulation is in accordance with ISO 16649-2 (3). It can also be used to isolate *E.coli* from samples.

Tryptone provides carbon, nitrogen compounds, long chain amino acids, vitamins and other essential growth nutrients to the organisms. Bile salts mixture inhibits gram-positive organisms. The surface of the plated medium is dried before use. Dilute food samples 1:5 or 1:10 with 0.1% (w/v) sterile Peptone Water (M028) and homogenize in a blender or a stomacher. Pipette 0.5 ml or 1.0 ml of the homogenized food sample on to the plate and spread with sterile glass spreader. Incubate the plates at 30°C for 4 hours and then at 44°C for 18 hours.

### Type of specimen

Food samples

### Specimen Collection and Handling

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (3). 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.β-glucuronidase is present in 97% of *E.coli* strains, however few *E.coli* may be negative.
- 2.Certain species of *Salmonella* are β-glucuronidase positive.
- 3.Individual organisms differ in their growth requirement and may show variable growth patterns on the medium.
- 4.Each lot of the medium has been tested for the organisms specified on the COA. It is recommended to users to validate the medium for any specific microorganism other than mentioned in the COA based on the user's unique requirement.
- 5.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

Sterile HiCrome™ E. coli Agar Plate in 90 mm disposable plates with smooth surface and absence of black particles/cracks/bubbles

#### Colour of medium

Light yellow coloured medium

#### Quantity of medium

25 ml of medium in 90 mm disposable plates.

#### pH

7.00-7.40

#### Sterility Check

Passes release criteria

#### Cultural Response

Cultural characteristics observed after an incubation at 44°C (35-37 in coa) for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Colour of Colony
<i>Escherichia coli</i> ATCC 25922 (00013*)	50-100	luxuriant	>=50%	bluish green
<i>Salmonella</i> Enteritidis ATCC 13076 (00030*)	50-100	luxuriant	>=50%	colourless
<i>Staphylococcus aureus</i> subsp. <i>aureus</i> ATCC 25923 (00034*)	>=10 <sup>4</sup>	inhibited	0%	

Key : \*Corresponding WDCM numbers.

### Storage and Shelf Life

On receipt store between 2-8°C. 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 sample must be decontaminated and disposed of in accordance with current laboratory techniques (5,6).

### Reference

1. Anderson J.M. and Baird-Parker A.C., (1975), J. Appl. Bact., 39:111.
2. Hansen W. and Yourassawsky E.,(1984), J.Clin. Microbiol., 20:1177.
3. International Standard ISO 16649-2: 1999. Microbiology of food and animal feeding stuffs- Horizontal method for the enumeration of  $\beta$ -glucuronidase-positive *Escherichia coli*.
4. Salfinger Y., and Tortorello M.L. Fifth (Ed.),2001, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition
6. 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 : 00/2023

#### Disclaimer :

User must ensure suitability of the product(s) in their application prior to use. Products conform solely to the information contained in this and other related HiMedia™ publications. The information contained in this publication is based on our research and development work and is to the best of our knowledge true and accurate. HiMedia™ Laboratories Pvt Ltd reserves the right to make changes to specifications and information related to the products at any time. Products are not intended for human or animal or therapeutic use but for laboratory,diagnostic, research or further manufacturing use only, unless otherwise specified. Statements contained herein should not be considered as a warranty of any kind, expressed or implied, and no liability is accepted for infringement of any patents.