

# **Technical Data**

### HiCrome<sup>TM</sup> Coliform Agar w/ SLS Intended use

## HiCrome Coliform Agar w/ SLS is a selective agar recommended for the simultaneous detection of *Escherichia coli* and total coliforms in water, food and clinical samples.

#### Composition\*\*

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Ingredients	Gms / Litre
Peptone, special	3.000
Sodium chloride	5.000
Dipotassium hydrogen phosphate	3.000
Potassium dihydrogen phosphate	1.700
Sodium pyruvate	1.000
L-Tryptophan	1.000
Sodium lauryl sulphate	0.100
Chromogenic mixture	0.200
Agar	12.000
Final pH ( at 25°C)	$6.8 \pm 0.2$
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\*\*Formula adjusted, standardized to suit performance parameters

#### Directions

Suspend 27 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. Add 5mg/l novobiocin before autoclaving the medium, when a high number of gram positive accompanying bacteria are expected. Mix well and pour into sterile Petri plates.

#### **Principle And Interpretation**

HiCrome<sup>TM</sup> Coliform Agar w/ SLS is a selective medium recommended for the simultaneous detection of *Escherichia coli* and total coliforms in water and food samples (7). Peptone special and sodium pyruvate provide essential growth nutrients to the organisms. The phosphates buffer the medium well. The medium composition helps even the sub lethally injured coliforms to grow rapidly. Sodium lauryl sulphate inhibits gram-positive organisms. The chromogenic mixture contains two chromogenic substrates. The enzyme  $\beta$ -galactosidase produced by coliforms cleaves one chromogen, resulting in the salmon red colouration of coliform colonies. The enzyme  $\beta$ -galactosidase produced by *E. coli*, cleaves X-glucuronide. *E. coli* forms dark blue to violet coloured colonies due to cleavage of both the chromogens (2,5,6). The addition of L-Tryptophan improves the indole reaction, thereby increasing detection reliability in combination with the two chromogens. To confirm *E. coli*, add a drop of Kovacs reagent (R008) on the dark-blue to violet colony. Formation of cherry-red colour indicates positive reaction.

#### Type of specimen

Clinical samples - faeces, urine , food samples ; Water samples.

#### **Specimen Collection and Handling**

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (3,4). For food samples, follow appropriate techniques for sample collection and processing as per guidelines (8). 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

In Vitro diagnostic use only. 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 clinical specimens. Safety guidelines may be referred in individual safety data sheets.

#### **M1300**

#### **Limitations :**

- 1. ß-glucuronidase is present in 97% of *E.coli* strains, however few *E.coli* may be negative.
- 2. Certain species of *Shigella* and *Salmonella* are β-glucuronidase positive

#### **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 beige homogeneous free flowing powder

#### Gelling

Firm, comparable with 1.2% Agar gel.

#### Colour and Clarity of prepared medium

Light yellow, clear to slightly opalescent gel forms in Petri plates

#### Reaction

Reaction of 2.7% w/v aqueous solution at 25°C. pH : 6.8±0.2

#### pН

6.60-7.00

#### **Cultural Response**

Cultural characteristics observed after an incubation at 35-37°C for 24 hours (48 hours if necessary).

Organism	Inoculum (CFU)	Growth	Recovery	Colour of Colony	Indole production
<i>Citrobacter freundii ATCC</i> 8090	50-100	good-luxuriant	>=50%	salmon to red	negative reaction
Escherichia coli ATCC 8739 (00012)*	50-100	good-luxuriant	>=50%	dark blue/viole	t positive, confirmation of red colour around the colony by addition of Kovacs reagent (R008)
Enterobacter cloacae ATCC 23355 (00082*)	50-100	good-luxuriant	>=50%	salmon to red	negative reaction
Enterococcus faecalis ATCC 29212 (00087*)	C>=10 <sup>3</sup>	inhibited	0%		
Klebsiella pneumoniae ATCC 13883 (00097*)	50-100	good-luxuriant	>=50%	light pink	negative reaction
Salmonella Enteritidis ATCO 13076 (00030*)	C50-100	good	40-50%	colourless	negative reaction
Shigella flexneri ATCC 12022 (00126*)	50-100	good	40-50%	colourless	negative reaction

Key: \*Corresponding WDCM numbers.

#### **Storage and Shelf Life**

Store dehydrated powder and prepared medium on receipt at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder 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.

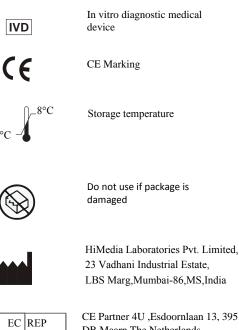
#### **Disposal**

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 (3,4).

#### Reference

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- 3. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2<sup>nd</sup> Edition.
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- 8. 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.

Revision : 02/2019



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#### Disclaimer :

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