

HiCrome™ EC 0157:H7 Selective Agar Base, Modified

Recommended for selective isolation and easy detection of *Escherichia coli* O157:H7 from food samples.

M1575A

Composition **

Ingredients	Grams/Litre
Tryptone	5.00
Yeast extract	3.00
Sorbitol	7.00
Bile salts mixture	1.50
Sodium lauryl sulphate (SLS)	0.10
Chromogenic mixture	0.25
Agar	15.00

Final pH (at 25°C) 6.8 ± 0.2

** Formula adjusted, standardized to suit performance parameters

Directions

Suspend 31.85 grams in 990 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 rehydrated contents of 1 vial of HiCrome EC 0157:H7 Selective Supplement (FD187) aseptically. Mix well and pour into sterile Petri plates.

Principle and Interpretation

Enterohaemorrhagic *E. coli* strains are also termed as verocytotoxin-producing *E. coli* (VTEC/ EHEC). Although many different serotypes of *Escherichia coli* are known to produce verocytotoxin (2) those of *Escherichia coli* O157:H7 and O157:H are so far the common types causing human infections. O157 VTEC strains have several unusual biochemical characters that are exploited in methods for their laboratory identification. They belong to the minority of *E. coli* that are β -glucuronidase negative and do not ferment sorbitol or rhamnose within 24 hours. These can be isolated from faecal specimens by plating on media containing D-sorbitol instead of lactose.

HiCrome™ EC 0157:H7 Selective Agar Base, Modified is based on the formulation described by Rappaport and Henigh (1). The medium contains sorbitol and a proprietary 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* forms bluish green coloured colonies.

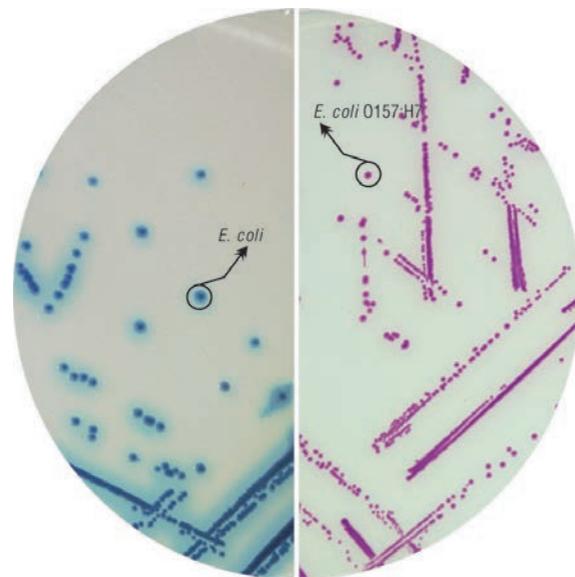
Tryptone and yeast extract provides carbonaceous, nitrogenous and growth nutrients. Addition of HiCrome™ EC 0157:H7 Selective Supplement (FD187) makes the medium selective (3). Potassium tellurite selectively inhibits *Aeromonas* and *Providencia* species. Novobiocin inhibits gram-positive bacteria. Sodium lauryl sulphate helps to inhibit the accompanying gram-positive flora.

Type of specimen

Food and dairy samples.

Specimen Collection and Handling

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (4, 5, 6).



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HiCromeVeg™ Freedom from BSE / TSE worries
Single Streak Rapid Differentiation Series

HiCrome™ EC 0157:H7 Selective Agar Base (M1575A) is also available as HiCrome™ EC 0157:H7 Selective HiVeg Agar Base (MV1575A) wherein all the animal origin nutrients have been replaced by vegetable based nutrients.

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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. Some species may show poor growth due to varying nutritional requirements.
2. Further biochemical test must be carried out for confirmation.

Performance and Evaluation

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

Quality Control

- Appearance of Powder** : Cream to yellow coloured, homogeneous, free flowing powder.
- Gelling** : Firm, comparable with 1.5% Agar gel.
- Colour and Clarity of prepared medium** : Light amber coloured, clear to slightly opalescent gel forms in Petri plates.
- Reaction** : Reaction of 3.18% w/v aqueous solution at 25°C. pH : 6.8 ± 0.2.
- Cultural Response** : Cultural characteristics observed with added HiCrome™ EC 0157:H7 Selective Supplement (FD187). after an incubation at 35-37°C for 18-24 hours

Organisms (ATCC)	Inoculum (CFU)	Growth	Recovery	Colour of colony
<i>Escherichia coli</i> O157:H7 (NCTC 12900)	50-100	luxuriant	≥50%	dark purple-magenta
<i>Escherichia coli</i> (25922) (00013*)	50-100	none to poor	≤10%	bluish green
<i>Pseudomonas aeruginosa</i> (27853) (00025*)	50-100	fair to good	30-40%	colourless
<i>Klebsiella pneumoniae</i> (13883) (00097*)	>10 ³	fair to good	30-40%	colourless - mauve (mucoid)
<i>Bacillus subtilis</i> sub spizizenii (6633) (00003*)	≥10 ³	inhibited	0%	-
<i>Staphylococcus aureus</i> subsp aureus (25923) (00034*)	≥10 ³	inhibited	0%	-

Key : * : corresponding WDCM Numbers

Storage and Shelf-life

Store between 2-8°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. 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 (7, 8).

References

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5. 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.
6. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed APHA Inc., Washington, D.C.
7. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
8. 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.