HiCrome™ Enrichment Broth Base for EC O157:H7

M1598

Intended Use

Recommended for isolation and selective differentiation of *Escherichia coli* O157:H7 from food, environmental by chromogenic method.

Composition**

Ingredients	g / L
Tryptone	10.000
Sorbitol	10.000
Bile salts mixture	1.500
Chromogenic mixture	1.300
Final pH (at 25°C)	7.1±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 11.4 gram in 500 ml purified/distilled water. Heat if necessary to dissolve the medium completely. Sterilize by autoclaving at 15lbs pressure (121°C) for 15 minutes. For selective isolation of *E.coli* O157:H7, aseptically add the rehydrated contents of 1vial of NPt Selective Supplement (FD230). Mix well and dispense into sterile test tubes.

Principle And Interpretation

March and Ratnam (1) reported the inability of *Escherichia coli* O157:H7 to ferment sorbitol while developing Sorbitol MacConkey medium. Subsequently Thomson et al (2) observed the absence of ß-glucoronidase activity in *E.coli* O157:H7 from a variety of samples by direct culture. The medium contains tryptone that provides nitrogenous, carbonaceous compounds, long chain amino acids, vitamins and other essential growth nutrients. Sorbitol is the fermentable carbohydrate; bile salt mixture inhibits most of the gram-positive organisms. Addition of tellurite (FD230) makes the medium more specific and selective. The bluish colour development by colonies of *E.coli* and *Klebsiella* in the medium is due to the enzymes ß-glucoronidase and ß-D-galactosidase that cleaves the chromogenic substrates present in chromogenic mixture respectively. However *E.coli* O157:H7 gives a purple colour to the medium due to the absence of ß-glucoronidase and its inability to ferment sorbitol.

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. ß-glucuronidase is present in 97% of *E.coli* strains, however few *E.coli* may be negative.
2. Some species may show poor growth due to nutritional variations.
3. 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

Cream to yellow homogeneous free flowing powder

Colour and Clarity of Prepared Medium

Light yellow coloured, clear solution which may have slight precipitate.

Reaction

Reaction of 2.28% w/v aqueous solution at 25°C. pH : 7.1±0.2

pH

6.90-7.30

Cultural Response

Cultural characteristics observed with added NPt Selective Supplement (FD230), after an incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Colour of Medium	Growth (after addition of FD230	Colour of Medium (after addition of FD230
Klebsiella pneumoniae ATCC 13883 (00097*)	50-100	good-luxuriant	bluish-green may show slight precipitation of growth	good	bluish green may show slight precipitation of growth
Escherichia coli ATCC 25922 (00013*)	50-100	good-luxuriant	blue may show slight precipitation of growth	inhibited	
Enterococcus faecalis ATCC 29212 (00087*)	>=10^4	inhibited	-	inhibited	-
Escherichia coli 0157:H7 (NCTC 12900) (00014*)		good-luxuriant	purple may show slight precipitation of growth	good-luxuriant	purple may show slight precipitation of growth
#*Cronobacter sakazakii* ATCC 12868	50-100	good-luxuriant	white may show slight precipitation of growth	none-poor	colourless may show slight precipitation of growth
Staphylococcus aureus subsp *aureus* ATCC 25923 (00034*)	>=10^4	inhibited	-	inhibited	-
Salmonella Enteritidis ATCC 13076 (00030*)		good-luxuriant	light green may show slight precipitation of growth	good	light green may show slight precipitation of growth
Shigella flexneri ATCC 12022 (00126*)	50-100	good	colourless	inhibited	-

Key : * Corresponding WDCM numbers, # Formerly known as *Enterobacter sakazakii.*

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 (4,5).

Reference

1. March S. B and Ratnam S., 1986, J. Clin. Microbiol., 23:869-872.
2. Thompson J. S., Hodge D. S., Borczyk A. A., 1990, J. Clin. Microbiol.28, 2165-2168.
3. Salfinger Y., and Tortorello M.L., 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd 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.

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