

## Sorbitol Iron HiVeg™ Agar

MV299

### Intended Use:

Recommended for cultural identification and differentiation of enteropathogenic *Escherichia coli* which do not ferment sorbitol.

### Composition\*\*

Ingredients	g / L
HiVeg™ extract	3.000
HiVeg™ peptone No. 3	15.000
D-Sorbitol	2.000
Sodium chloride	5.000
Ferric ammonium citrate	0.500
Sodium thiosulphate	0.500
Phenol red	0.030
Agar	20.000
Final pH ( at 25°C)	7.6±0.2

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

### Directions

Suspend 46.03 grams in 1000 ml purified/distilled water. Heat to boiling to dissolve the medium completely. Dispense in test tubes and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C. Allow the tubes to cool in a slanted position.

### Principle And Interpretation

*Escherichia coli* is the most common bacterium isolated in clinical samples, the most prevalent facultative gram-negative rods in faeces, the most common cause of urinary tract infection and a common cause of both intestinal and extra-intestinal infections (1). Strains of *E. coli* that are primary intestinal pathogens of man are described in four groups namely Enterotoxigenic *E. coli* (ETEC), Enteroinvasive *E. coli* (EIEC), Verocytotoxin-producing *E. coli* (VTEC) and Enteropathogenic *E. coli* (EPEC) (2). EPEC causes infantile diarrhea (1).

Sorbitol Iron Agar is a differential tube medium described by Rappaport and Henig (1). It is a modification of Kligler Iron Agar where dextrose and lactose is substituted with D-sorbitol. The pathogenic strain of *E. coli* is identified on the basis of inability to ferment sorbitol and hydrogen sulfide production. Sorbitol Iron HiVeg™ Agar is prepared by using vegetable peptones in place of animal based peptones which make the media free of BSE/TSE risks.

HiVeg™ extract and HiVeg™ peptone No. 3 in the medium provide carbon, nitrogen, vitamins and minerals required for the growth of organisms. D-Sorbitol is the fermentable carbohydrate source. Sodium chloride provides essential ions. The combination of ferric ammonium citrate and sodium thiosulphate enables the detection of hydrogen sulphide production, which is evidenced by a black colour formation. Phenol red is the pH indicator, detecting the fermentation of sorbitol and subsequent formation of acidic conditions.

### Type of specimen

Food samples

### Specimen Collection and Handling

Colourless colonies from Sorbitol HiVeg™ Agar (MV298) are inoculated into Sorbitol Iron Agar by stabbing the butts and streaking the slants. After 18-24 hours, freshly isolated pathogenic strains of *E. coli* show neither acid nor blackening of the medium. *Proteus* species may or may not blacken the medium, may produce acid in the butt; and on transfer to urease test medium, will give a positive urease test. Ordinary strains of *E. coli* produce acid and gas on Sorbitol Iron Agar, some pathogenic strains after laboratory cultivation may develop the capacity to ferment sorbitol and produce acid. Subsequently transfer of such cultures on Kligler Iron HiVeg™ Agar (MV078) or Triple Sugar Iron HiVeg™ Agar (MV021), Urease Test Medium will help in identification.

### 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 .Further biochemical and serological tests need to 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 expiry period when stored at recommended temperature.

## Quality Control

### Appearance

Light yellow to pink homogeneous free flowing powder

### Gelling

Firm, comparable with 2.0% agar gel.

### Colour and Clarity of prepared medium

Red coloured clear to slightly opalescent gel forms in tubes as slants

### Reaction

Reaction of 4.6% w/v aqueous solution at 25°C. pH : 7.6±0.2

### pH

7.40-7.80

### Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Sorbitol	H <sub>2</sub> S
<b>Growth Promoting</b>				
<i>Escherichia coli</i> ATCC 25922 (00013*)	50-100	luxuriant	positive reaction, yellow colour with gas formation	negative reaction
# <i>Klebsiella aerogenes</i> ATCC 13048 (00175*)	50-100	luxuriant	positive reaction, yellow colour	negative reaction
<i>Enterococcus faecalis</i> ATCC 29212 (00087*)	50-100	luxuriant	positive reaction, yellow colour	negative reaction
<i>Klebsiella pneumoniae</i> ATCC 13883 (00097*)	50-100	luxuriant	positive reaction, yellow colour	negative reaction
\$ <i>Proteus hauseri</i> ATCC 13315	50-100	luxuriant	negative reaction	positive reaction, blackening of medium
<i>Salmonella</i> Typhimurium ATCC 14028 (00031*)	50-100	luxuriant	positive reaction, yellow colour	positive reaction, blackening of medium
<i>Shigella flexneri</i> ATCC 12022 (00126*)	50-100	luxuriant	negative reaction	negative reaction
<i>Escherichia coli serotype</i> 011 and 055	50-100	luxuriant	negative reaction	negative reaction
<i>Escherichia coli O157:H7</i> NCTC 12900 (00014*)	50-100	luxuriant	negative reaction	negative reaction

Key : (\*) Corresponding WDCM numbers. (#) Formerly known as *Enterobacter aerogenes* (\$) Formerly known as *Proteus vulgaris*

## Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°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 (3,4).

## Reference

1. Rappaport F. and Henig E., 1952, J. Clin. Pathol., 5:361.
2. Collee J. G., Fraser A. G., Marmion B. P., Simmons A., (Eds.), Mackie and McCartney, Practical Medical Microbiology, 1996, 14th Edition, Churchill Livingstone.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
4. 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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### Disclaimer :

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