### **Hugh Leifson Medium**

### Intended Use

Hugh Leifson Medium is used to distinguish between anaerobic and aerobic breakdown of carbohydrate (glucose).

### Summary

Hugh Leifson Medium is used for isolation and cultivation of *Vibrio cholerae* and other *Vibrio* species which causes food poisoning. This medium was formulated by Hugh and Leifson. They described the taxonomic significance of fermentative and oxidative metabolism of carbohydrates in Gram-negative intestinal bacteria.

### **Principle**

The medium contains a high concentration of carbohydrate and low concentration of peptone to avoid the possibility of an aerobic organism utilizing peptone and producing an alkaline condition which would neutralize slight acidity produced by an oxidative organism. Dipotassium phosphate promotes fermentation and acts as pH controlling buffer. Agar concentration enables the determination of motility and aids in distribution of acid throughout the tube produced at the surface of medium. Oxidative organisms produce acid in unsealed tube with little or no growth and no acid formation in sealed tube while fermentative organisms produce acid in both sealed and unsealed tubes.

#### Formula\*

Ingredients	g/L
Peptone	2.0
Dipotassium phosphate	0.3
Sodium chloride	5.0
Glucose (Dextrose)	10.0
Bromothymol blue	0.05
Agar	2.0
Final pH (at 25°C)	$6.8 \pm 0.2$
*Adjusted to suit performance parameters.	

#### **Storage and Stability**

Store dehydrated medium below 30°C in tightly closed container and the prepared medium at 2°C-8°C. Avoid freezing and overheating. Use before expiry date on the label. Once opened keep powdered medium closed to avoid hydration.

### Type of specimen

Clinical samples- Blood Water sample Food and dairy sample

### **Specimen Collection and Handling**

Ensure that all samples are properly labelled.

Follow appropriate techniques for handling samples as per established guidelines.

Some samples may require special handling, such as immediate refrigeration or protection from light, follow the standard procedure.

The samples must be stored and tested within the permissible time duration.

After use, contaminated materials must be sterilized by autoclaving before discarding.

### **Directions**

- 1. Suspend 19.35 g of the powder in 1000 mL purified / distilled water.
- 2. Heat to boiling to dissolve the powder completely.
- 3. Dispense into test tubes in duplicate for aerobic and anaerobic fermentation.
- 4. Sterilize by autoclaving at 121°C (15 psi) for 15 minutes as per validated cycle.
- 5. Cool the tubed medium in an upright position.

# **Quality Control**

**Dehydrated Appearance:** Light yellow to bluish green coloured, homogenous, free flowing powder. **Prepared Appearance:** Bluish green coloured, clear to slightly opalescent semisolid gel forms in tubes as slants. **Cultural Response:** Cultural characteristics observed after an incubation of 18-48 hours at 35°C-37°C.

<b>Organism (ATCC)</b> <i>Klebsiella</i> <i>aerogenes</i> (13048)	<b>Motility</b> Positive, growth away from stabline causing turbidity	Aerobic fermentation Acid (yellow) and gas production, positive reaction	Anaerobic fermentation Acid (Yellow) and gas production, positive reaction
Escherichia coli (25922)	Positive, growth away from stabline causing turbidity	Acid (yellow) and gas production, positive reaction	Acid (Yellow) and gas production, positive reaction
<i>Pseudomonas aeruginosa</i> Strain Boston 41501 (27853)	Positive, growth away from stabline causing turbidity	Acid (yellow) production, positive reaction	unchanged (green) or alkaline(blue) negative reaction
Salmonella Typhi (6539)	Positive, growth away from stabline causing turbidity	Acid (yellow) and gas production, positive reaction	Acid (Yellow) and gas production, positive reaction
Shigella sonnei (25931)	Negative growth along the stabline, surrounding medium	Acid (yellow) production, positive reaction	Acid (Yellow) and gas production, positive reaction

## Performance and Evaluation

Performance of the product is dependent on following parameters as per product label claim:

- 1. Directions
- 2. Storage
- 3. Expiry

# **Precautions/Limitations**

This medium is general purpose medium and may not support the growth of fastidious organisms.

# Warranty

This product is designed to perform as described on the label and package insert. The manufacturer disclaims any implied warranty of use and sale for any other purpose.

# Reference

- 1. Hugh and Leifson, 1953, J. Bacteriol., 66:24.
- 2. Bureau of Indian Standards, IS:5887 (Part V) 1976, reaffirmed 1986.
- 3. MacFaddin J.F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. I, Williams and Co., St. Louis. Wilkins, Baltimore.
- 4. Downes F. P. and Ito K., (Ed.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., American Public Health Association, Washington, D.C.
- 5. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D.C.
- 6. Greenberg A. E., Clesceri L. S. and Eaton A. D., (Eds.), 2005, Standard Methods for the Examination of Water and Wastewater, 21st ed., APHA, Washington, D.C.
- 7. Isenberg, H.D. Clinical Microbiology Procedures Handb0ook. 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.
- 9. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
- 10. Data on file: Microxpress<sup>®</sup>, A Division of Tulip Diagnostics (P) Ltd.

## **Product Presentation:**

**Cat No.** 201080030500 Product description Dehydrated Culture Media

Pack Size

#### Disclaimer

Information provided is based on our inhouse technical data on file, it is recommended that user should validate at his end for suitable use of the product.