TM 742- HUGH LEIFSON MEDIUM (IS : 5887 (Part V) 1976, reaffirmed 2005)

INTENDED USE

For detecting aerobic and anaerobic breakdown of glucose.

PRODUCT SUMMARY AND EXPLANATION

Hugh Leifson Medium is recommended by BIS for the isolation and cultivation of *Vibrio cholerae* and other *Vibrio* species which cause food poisoning. It was formulated by Hugh and Leifson. They described the taxonomic significance of fermentative and oxidative metabolism of carbohydrates in gram-negative intestinal bacteria.

COMPOSITION

Ingredients	Gms / Ltr
Glucose	10.000
Sodium chloride	5.000
Agar	3.000
Peptic digest of animal tissue	2.000
Dipotassium phosphate	0.300
Bromothymol blue	0.030

PRINCIPLE

The medium contains a high concentration of added carbohydrates relative to the Peptic digest of animal tissue concentration to avoid the utilization of peptone by an aerobic organism and the production of an alkaline reaction which would neutralize slight acidity produced by an oxidative organism. The dipotassium phosphate adds buffering capacity to the medium. The agar permits the property of semi solid media, which helps in determination of motility and aids in the even distribution of any acid produced at the surface of the medium.

INSTRUCTION FOR USE

1. Dissolve 20.33 grams in 1000ml distilled water.
2. Gently heat to boiling with gentle swirling and dissolve the medium completely.
3. Dispense in test tubes in duplicate for aerobic and anaerobic fermentation.
4. Sterilize by autoclaving at 10 psi (115°C) for 20 minutes.
5. Cool the tubed medium in upright position.

QUALITY CONTROL SPECIFICATIONS

Appearance of Dehydrated Powder : Light yellow to bluish green, Homogeneous free flowing powder
Appearance of Prepared medium : Greenish blue coloured, clear to slightly opalescent gel as butts
pH (at 25°C) : 7.1± 0.2

INTERPRETATION

Cultural characteristics observed after incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Motility	Aerobic fermentation	Anaerobic fermentation	Incubation Temperature	Incubation period
Enterobacter aerogenes	13048	50-100	Positive, growth away from stab line causing turbidity	Acid (yellow) and gas production, positive reaction	Acid (yellow) and gas production, positive reaction	35-37°C	18-48 Hours
Pseudomonas aeruginosa	27853	50-100	Positive, growth away from stab line causing turbidity	Acid (yellow) production, positive reaction	Unchanged (green) or alkaline (blue), negative reaction	35-37°C	18-48 Hours
Escherichia coli	25922	50-100	Positive, growth away from stab line causing turbidity	Acid (yellow) and gas production, positive reaction	Acid (yellow) and gas production, positive reaction	35-37°C	18-48 Hours
Salmonella typhi	6539	50-100	Positive, growth away from stab line causing turbidity	Acid (yellow) and gas production, positive reaction	Acid (yellow) and gas production, positive reaction	35-37°C	18-48 Hours
Shigella sonnei	25931	50-100	Negative, growth along the stab line, surrounding medium	Acid (yellow) production, positive reaction	Acid (yellow) and gas production, positive reaction	35-37°C	18-48 Hours

PACKAGING

In 100 & 500 gm packaging size.

STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers below 25°C and protect from direct Sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

Product Deterioration: Do not use powder if they show evidence of microbial contamination, discoloration, drying, or other signs of deterioration.

DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

REFERENCES

1. MacFaddin, J.F. Media for isolation-cultivation-identification-maintenance of medical bacteria, vol. I. Williams & Wilkens, Baltimore. (1985).
2. Shigei, J. Test methods used in the identification of commonly isolated aerobic gram-negative bacteria. Oxidation-fermentation test, p. (1992).
3. Hugh, R., and E. Leifson. The taxonomic significance of fermentative versus oxidative metabolism of carbohydrates by various gram-negative bacteria. J. Bacteriol. 66:24-26. (1953).
4. Bureau of Indian Standards, IS:5887 (Part V) 1976, reaffirmed 1986.
5. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D.C.

NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.

***For Lab Use Only**

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