



## Hugh Leifson Glucose Medium

M871

### Intended Use:

Recommended for the differentiation of Staphylococci from Micrococci on the basis of their ability to ferment glucose anaerobically.

### Composition\*\*

Ingredients	g / L
Peptone	2.000
Yeast extract	0.500
Sodium chloride	30.000
Dextrose (Glucose)	10.000
Bromocresol purple	0.015
Agar	3.000
Final pH ( at 25°C)	7.4±0.2

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

### Directions

Suspend 45.52 grams in 1000 ml purified/distilled water. Heat to boiling to dissolve the medium completely. Dispense into test tubes in duplicate for aerobic and anaerobic fermentation. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool the tubed medium in an upright position.

### Principle And Interpretation

Hugh Leifson Glucose Medium is formulated by Hugh and Leifson (1). Hugh Leifson Glucose Medium is prepared as described by FDA (2) for differentiation of Staphylococci from Micrococci. They described the taxonomic significance of fermentative and oxidative metabolism of carbohydrates in gram-negative intestinal bacteria. There are two ways of utilizing carbohydrates by microorganisms, namely fermentation and oxidation. This property may be frequently used for the differentiation of some bacteria.

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 (3,4). Agar concentration enables the determination of motility and aids in distribution of acid throughout the tube produced at the surface of medium.

Hugh Leifson Glucose Medium contains high salt concentration thus it is used for the identification of pathogenic and halophilic organisms and for testing aerobic and anaerobic breakdown of glucose by Staphylococci and Micrococci (5).

### Type of specimen

Clinical samples- Swabs of mouth, mucosae, oropharynx and upper respiratory tract; Food and dairy samples

### Specimen Collection and Handling:

The tubes for aerobic and anaerobic fermentation are inoculated and the agar surface of one tube of duplicate is covered with layer of sterile paraffin oil, about 25 mm thickness and incubated at 37°C. 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. If acid is produced, it is indicated by change in colour from purple to yellow throughout the medium. Liquid paraffin tube used should be dry sterilized at 160-170°C for 2 hours. Wet sterilization with high pressure is not sufficient for the purpose. Inoculate the culture under test into two tubes of the medium by stabbing throughout their length with a long wire loop. Cover one tube of the pair with layer of sterile liquid paraffin and incubate at 37°C. Read yellow colouration as acid production from glucose. Staphylococci produce acid by fermentation throughout the depth of the medium both in the anaerobic tubes sealed with paraffin and the aerobic unsealed tube. Micrococci either fail to produce acid in either tube or produce it only by oxidation in the upper part of the aerobic tube.

## Warning and Precautions :

In Vitro diagnostic use. For professional use only. 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 clinical specimens. Safety guidelines may be referred in individual safety data sheets.

## Limitations :

1. Wet sterilization with high pressure is not sufficient for the purpose.

## 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 bluish grey homogeneous free flowing powder

### Gelling

Semisolid, comparable with 0.3% Agar gel.

### Colour and Clarity of prepared medium

Purple coloured, clear to slightly opalescent gel forms in tubes as butts

### Reaction

Reaction of 4.55% w/v aqueous solution at 25°C. pH : 7.4±0.2

### pH

7.20-7.60

### Cultural Response

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

Organism	Growth	Colour of Medium (Aerobic)	Colour of Medium (Anaerobic)
<i>Micrococcus luteus</i> ATCC 10240	good	yellow	pink-purple
<i>Staphylococcus aureus</i> subsp. <i>aureus</i> ATCC 25923 (00034*)	good	yellow	yellow

Key : \*Corresponding WDCM numbers.

## Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-30°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 clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (6,7).

## Reference

1. Hugh and Leifson, 1953, J. Bacteriol., 66:24.
2. Bacteriological Analytical Manual, 1995, 8th Ed., Food & Drug Administration, AOAC International, USA.
3. Finegold S. M., Martin W. J., and Scott E. G., 1978, Bailey and Scotts Diagnostic Microbiology, 5th Ed., The C.V. Mosby Co., St. Louis.
4. MacFaddin J.F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol.I, Williams and Wilkins, Baltimore.
5. Baird Parker, 1966, International subcommittee on Staphylococci and Micrococci.

6.Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.

7.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.

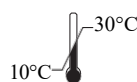
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