

Technical Information

Hugh Leifson Glucose Medium

Product Code: DM 1871

Application: Hugh Leifson Glucose Medium is recommended for the differentiation of Staphylococci from Micrococci on the basis of their ability to ferment glucose anaerobically.

Composition**

Ingredients	Gms / Litre
Peptic digest of animal tissue	2.000
Yeast extract	0.500
Sodium chloride	30.000
Glucose	10.000
Bromocresol purple	0.0 15
Agar	3.000
Final pH (at 25°C)	7.4±0.2

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

Hugh Leifson Glucose Medium formulated by Hugh and Leifson ⁽²⁾ is prepared as described by FDA ⁽¹⁾ for differentiation of Staphylococci from Micrococci. They described the taxonomic significance of fermentative and oxidative metabolism of carbohydrates in gram-negative intestinal bacteria also 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.

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

The medium contains a high concentration of carbohydrate and low concentration of peptic digest of animal tissue to avoid the possibility of an aerobic organism utilizing peptic digest of animal tissue 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 helps in distribution of acid throughout the tube produced at the surface of medium. 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.

Methodology

Suspend 45.52 grams of powder media in 1000 ml distilled water. Shake well & heat 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.

Quality Control

Physical 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 Range: 7.2-7.6

Cultural Response/Characteristics

DM1871: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Colour of Medium (Aerobic)	Colour of Medium (Anaerobic)
<i>Micrococcus luteus</i> ATCC 1024	50-100	Good	yellow	Pink-purple
<i>Staphylococcus aureus</i> ATCC 25923	50-100	Good	Yellow	yellow

Storage and Shelf Life

Dried Media: Store below 30°C in tightly closed container and use before expiry date as mentioned on the label.

Prepared Media: 2-8° in sealable plastic bags for 2-5 days.

Further Reading

1. Bacteriological Analytical Manual, 1995, 8th Ed., Food & Drug Administration, AOAC International, USA.
2. Hugh and Leifson, 1953, J. Bacteriol., 66:24.
3. MacFaddin J.F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol.I, Williams and Wilkins, Baltimore.
4. 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.
5. Baird Parker, 1966, International subcommittee on Staphylococci and Micrococci.

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- User must ensure suitability of the product(s) in their application prior to use.
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