



Hoyle Medium Base

M015

Intended use

A highly selective medium used for the isolation and differentiation of *Corynebacterium diphtheriae* types from clinical and non-clinical specimens.

Composition**

Ingredients	g / L
Peptone	10.000
HM peptone B [#]	10.000
Sodium chloride	5.000
Agar	15.000
Final pH (at 25°C)	7.8±0.2

**Formula adjusted, standardized to suit performance parameters

- Equivalent to Beef extract

Directions

Suspend 40.0 grams in 940 ml purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C and aseptically add 50 ml of laked blood and 10 ml of PTe 3.5% Selective Supplement (1 ml per vial) (FD047). Mix well and pour into sterile Petri plates.

Principle And Interpretation

The most common disease caused by *Corynebacterium diphtheriae* is diphtheria, an acute communicable disease manifested by both local infection of the upper respiratory tract and the systemic effects of the toxin, which are most notable in the heart and peripheral nerves (1). Hoyle Medium Base, formulated by Hoyle (2), is the modification of the original formulation of Neill, for the isolation and differentiation of *C. diphtheriae*. This medium is not inhibitory to some mitis types of *Corynebacterium*, as the original formulation.

Peptone and HM peptone B supply carbon, nitrogen substances, amino acids, vitamins and other essential growth nutrients. Potassium tellurite is a selective agent, which inhibits most of the normal flora of the upper respiratory tract except *Corynebacterium*. Hoyle's Medium is a highly selective medium and should be used in conjunction with a non-selective media such as Loeffler Serum Medium (M537) and Blood Agar Base (M089) with 10% horse blood (3). *C. diphtheriae* are usually present in small numbers permitting the formation of well isolated colonies. So, inoculation is done by directly rubbing the swab over the entire surface of the medium. Incubation should be carried out till 72 hours if the results are negative. To study the morphology, gentian violet staining is done. To demonstrate the characteristic morphology and staining reactions of *C. diphtheriae* by Neissers or Alberts method, it is advisable to use colonies from Loeffler Medium. The toxigenicity of *C. diphtheriae* strains can be determined by Eleks (4) method.

Type of specimen

Clinical samples - Throat swab, nasal swab, wound swab

Specimen Collection and Handling:

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (3,5).

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

Warning and Precautions :

In Vitro diagnostic Use only. 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. Each lot of the medium has been tested for the standard organisms specified on the COA. However, hospital isolates may show variable growth depending on nutritional requirement.
2. Biochemical and serological tests must 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

Cream to yellow homogeneous free flowing powder

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Basal Medium: Amber coloured, clear to slightly opalescent gel . After Addition of blood & Tellurite : Brownish red coloured opaque gel forms in Petri plates

Reaction

Reaction of 4.0% w/v aqueous solution at 25°C. pH : 7.8±0.2

pH

7.60-8.00

Cultural Response

Cultural characteristics observed with added 50 ml of laked blood and tellurite solution after an incubation at 35-37°C for 18-24 hours .

Organism	Inoculum (CFU)	Growth	Recovery	Colony characteristics
** <i>Bacillus spizizenii</i> ATCC 6633 (00003*)	≥10 ⁴	inhibited	0%	
<i>C. diphtheriae</i> type <i>intermedius</i> 14779	50-100	good-luxuriant	≥50%	grey colonies with darker centers
<i>Corynebacterium diphtheriae</i> type <i>mitis</i>	50-100	good-luxuriant	≥50%	grey colonies with shining surface
<i>Escherichia coli</i> ATCC 25922 (00013*)	≥10 ⁴	inhibited	0%	
<i>Enterococcus faecalis</i> ATCC 29212 (00087*)	50-100	good-luxuriant	≥50%	black minute colonies

Key : (*) Corresponding WDCM numbers.

**Formerly known as *Bacillus subtilis* subsp. *spizizenii*

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 2-8°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 (3,5).

Reference

- 1.Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Tenover F. C., 8th Ed., American Society for Microbiology, Washington, D.C. (Ed.), 2003, Manual of Clinical Microbiology.
- 2.Hoyle I., 1941, Lancet., 1:175.
- 3.Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
- 4.Elek S. D., 1948, Brit. Med. A1:493.Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.

5 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.

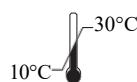
Revision : 05/2024



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**In vitro diagnostic
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Storage temperature



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