

Columbia HiCynth™ Broth

MCD145

Intended Use:

Recommended used as a general purpose medium and also for the cultivation of fastidious organisms various samples.

Composition**

Ingredients	Gms / Litre
HiCynth™ Peptone No.3*	23.000
L-Cysteine hydrochloride	0.100
Dextrose (Glucose)	2.500
Sodium chloride	5.000
Magnesium sulphate	0.100
Ferrous sulphate	0.020
Sodium carbonate	0.600
Tris (hydroxymethyl) aminomethane	0.830
Tris (hydroxymethyl) aminomethane hydrochloride	2.860
Final pH (at 25°C)	7.5±0.2

**Formula adjusted, standardized to suit performance parameters

*Chemically defined peptone

Directions

Suspend 35.01 grams in 1000 ml purified / distilled water. Heat to boiling to dissolve the medium completely. If desired, SPS (Sodium polyanethol sulphonate) may be added in a final concentration of 0.01%. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Principle And Interpretation

Morello and Ellner in 1969 devised a liquid medium for the recovery of microorganisms from blood cultures (1). This medium was devised from Columbia Blood Agar Base previously formulated by Ellner et al (2). While studying they found that Columbia Broth was superior to a commonly used general-purpose broth for faster growth of *Staphylococcus aureus*, *Escherichia coli*, viridans Streptococci and *Enterococcus* groups. In the formulation the increased concentration of cystine is provided for improved recovery of both aerobic and anaerobic microorganisms from blood specimens. Columbia HiCynth™ Broth is prepared by replacing animal and vegetable peptones with chemically defined peptones to avoid BSE/TSE risks associated with animal peptones.

The presence of CO₂ is stimulatory for many organisms. It differs from the agar base in that the cornstarch is omitted to reduce opalescence (1) and salts have been included.

Medium contains HiCynth™ Peptone No.3 to support luxurious growth of the organisms. Dextrose is added as a carbon and energy source. The medium is buffered with tris buffer. The addition of salts was found to be beneficial for the recovery of organisms. L-Cysteine HCL is the reducing agent. Magnesium & iron are added to facilitate organism growth. SPS (Sodium Polyanethol Sulphonate), a polyanionic anticoagulant inhibits complement and lysozyme activity, interferes with phagocytosis and inactivates aminoglycosides (3). Tube media should be inoculated with 1 to 2 drops of the liquid specimen using a sterile pipette. Swab specimens may be inserted into the broth after inoculation of the plated media. Liquid media should be reduced by placing the tubes with caps loosened under anaerobic conditions for 18-24 hours prior to inoculation for anaerobic incubation. Alternatively, it can be reduced immediately before use by boiling with caps loosened and cooling to room temperature with tightened caps, before inoculation. Growth in tubes is indicated by presence of turbidity compared to an uninoculated control. If growth appears, cultures should be subcultured onto appropriate media. Addition of SPS is inhibitory to *Neisseria* species, and thus 1.2% gelatin addition may counteract the inhibitory effect.

Type of specimen

Food samples

Specimen Collection and Handling

For food samples follow appropriate techniques for handling specimens as per established guidelines (4). After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

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 specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations :

1. As organisms differ in their nutritional requirements, some fastidious organisms may be inhibited or may show poor growth.
2. Each lot of the medium has been tested for the organisms specified on the COA. It is recommended to users to validate the medium for any specific microorganism other than mentioned in the COA based on the user's unique requirement.
3. Further biochemical and serological tests must be carried out for complete identification.

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

Colour and Clarity of prepared medium

Light amber coloured, clear to slightly opalescent solution, may have a fine precipitate.

Reaction

Reaction of 3.5% w/v aqueous solution at 25°C. pH : 7.5±0.2

pH

7.30-7.70

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours

Organism	Inoculum (CFU)	Growth
<i>Clostridium perfringens</i> ATCC 12924	50-100	good-luxuriant
<i>Neisseria meningitidis</i> ATCC 13090	50-100	good-luxuriant
<i>Staphylococcus aureus</i> <i>subsp. aureus</i> ATCC 25923 (00034*)	50-100	good-luxuriant
<i>Streptococcus mitis</i> ATCC 9811	50-100	good-luxuriant
<i>Streptococcus pyogenes</i> ATCC 19615	50-100	good-luxuriant

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 sample must be decontaminated and disposed of in accordance with current laboratory techniques (5,6).

Reference

1. Morello J. A. and Ellner P. D., 1969, Appl. Microbiol. 17:68.
2. Ellner P. D., Stoessel C. J., Drakeford E. and Vasi F., 1966, Am. J. Clin. Pathol., 45:502
3. Reller, Murray and MacLowry, 1982, Cumitech 1A, Blood cultures II, Coord. Ed., ASM, Washington D.C.
4. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
6. 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 : 01/2023

Disclaimer :

User must ensure suitability of the product(s) in their application prior to use. Products conform solely to the information contained in this and other related HiMedia™ publications. The information contained in this publication is based on our research and development work and is to the best of our knowledge true and accurate. HiMedia™ Laboratories Pvt Ltd reserves the right to make changes to specifications and information related to the products at any time. Products are not intended for human or animal or therapeutic use but for laboratory, diagnostic, research or further manufacturing use only, unless otherwise specified. Statements contained herein should not be considered as a warranty of any kind, expressed or implied, and no liability is accepted for infringement of any patents.