

Technical Information

CHO Medium Base

Product Code: DM 1351

Application: - CHO Medium Base is a basal medium to which carbohydrates may be added for use in fermentation studies of anaerobic bacteria.

Composition**

Ingredients	Gms / Litre
Casein enzymic hydrolysate	15.000
Yeast extract	7.000
L-Cystine	0.250
Sodium chloride	2.500
Ascorbic acid	0.100
Sodium thioglycollate	0.500
Bromo thymol blue	0.010
Agar	0.750
Final pH (at 25°C)	7.0±0.2

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

Primary identification of anaerobes is based on cellular morphology and colony characteristics on blood agar which is further confirmed by biochemical tests⁽¹⁾. Carbohydrate utilization patterns play a key role in the identification of anaerobes. Metabolism of anaerobes is less efficient and therefore require auxiliary growth factors. For the anaerobic microorganisms, proper collection and transport of suspected specimens is of utmost importance. Exposure of the specimens to air should be restricted to the possible minimum level and should be promptly cultured in the laboratory under proper anaerobic conditions. CHO Medium Base⁽²⁾ is recommended for studying fermentation of anaerobic bacteria⁽³⁻⁵⁾ for their final identification.

Casein enzymic hydrolysate and ascorbic acid enhance growth of oxygen sensitive and fastidious anaerobes⁽⁴⁾. Sodium chloride maintains the osmotic equilibrium of the medium. Yeast extract serves as a source of B-complex nutrients. L - Cystine and thioglycollate help in maintaining reduced oxygen atmosphere in the medium. Small amount of agar also aids in creating anaerobiosis. Bromothymol blue is the pH indicator.

Methodology

Suspend 26.11 grams of powder media in 1000 ml distilled water. Shake it well & heat to boil to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C. Aseptically add 6.25 ml of 10% sterile carbohydrate solution. Mix well and dispense in sterile tubes containing inverted Durhams tubes.

Quality Control

Physical Appearance

Cream to light green homogeneous free flowing powder

Colour and Clarity of prepared medium

Light green coloured, clear to very slightly opalescent solution without any precipitate

Reaction

Reaction of 2.6% w/v aqueous solution at 25°C. pH : 7.0±0.2

pH range 6.80-7.20

Cultural Response/ characteristics

DM 1351: Cultural characteristics observed when incubated anaerobically, after an incubation at 35-37°C for upto 7 days

Organism	Inoculum (CFU)	Growth	Fermentation w/Dextrose	Fermentation w/Lactose
<i>Bacteroides melaninogenicus</i> ATCC 25611	50-100	luxuriant	negative reaction, no colour change	Positive reaction, yellow colour
<i>Bacteroides vulgatus</i> ATCC 8482	50-100	luxuriant	negative reaction, no colour change	negative reaction, no colour change
<i>Bacteroides fragilis</i> ATCC 25285	50-100	luxuriant	Positive reaction, yellow colour	Positive reaction, yellow colour
<i>Clostridium botulinum</i> ATCC 25763	50-100	luxuriant	Positive reaction, yellow colour	negative reaction, no colour change
<i>Clostridium perfringens</i> ATCC 12924	50-100	luxuriant	Positive reaction, yellow colour	Positive reaction, yellow colour
<i>Escherichia coli</i> ATCC 35218	50-100	luxuriant	Positive reaction, yellow colour	Positive reaction, yellow colour

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. Laboratory Methods in Anaerobic Bacteriology, 1974, CDC Laboratory Manual, U.S. Dept. HEW, Pub. No. 74-8262.
2. Atlas, R. M., 2004, A Handbook of Microbiological Media, 3rd Ed, CRC Press.
3. MacFaddin, J. F., 1985, (Ed), Media for Isolation-Cultivation-Identification of Medical Bacteria. Vol. I., Williams and Wilkins, Baltimore.
4. Dowell V. R. Jr., Lombad G. L., Thompson F. S., Armfield A. Y., Media for Isolation, Characterization and Identification of Obligately Anaerobic Bacteria, USDHEW Atlanta, CA: Centers for Disease Control, 1977:22
5. Washington J. A., Laboratory Procedures in Clinical Microbiology, Cd 2 New York: Springer-Verlag, 1985: 774, 801-802.

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