

# **Technical Data**

CHO Medium Base M351

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

<sup>\*\*</sup>Formula adjusted, standardized to suit performance parameters

#### **Directions**

Suspend 26.11 grams in 1000 ml 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. Aseptically add 6.25 ml of 10% sterile carbohydrate solution. Mix well and dispense in sterile tubes containing inverted Durhams tubes.

## **Principle And Interpretation**

Identification of anaerobes is based on cellular morphology and colony characteristics on blood agar and confirmation by biochemical tests (1). Carbohydrate utilization patterns play a key role in the identification of anaerobes. Metabolism of anaerobes is less efficient and therefore they require auxiliary growth factors. For the anaerobic microorganisms, proper collection and transport of suspected specimens is of pivotal importance. Exposure of the specimens to air should be minimized to the possible extent and they should be promptly cultured in the laboratory under proper atmospheric conditions. CHO Medium Base (2) is recommended for studying fermentation of anaerobic bacteria (3, 4, 5).

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 atmosphere in the medium. Small amount of agar also aids in creating anaerobiosis. Bromothymol blue is the pH indicator.

## **Quality Control**

### 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 :  $7.0\pm0.2$ 

# pН

6.80-7.20

#### **Cultural Response**

M351: Cultural characteristics observed when incubated anaerobically, after an incubation at 35-37°C for upto 7 days

Organism Inoculum Growth Fermentation Fermentation (CFU) w/Dextrose w/Lactose

**Cultural Response** 

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Bacteroides melaninogenicus ATCC 25611	50-100	luxuriant	negative reaction, no colour change	positive reaction, yellow colour
Bacteroides vulgatus ATCC 8482	50-100	luxuriant	negative reaction, no colour change	negative reaction, no colour change
Bacteroides fragilis ATCC 25285	50-100	luxuriant	positive reaction, yellov colour	positive wreaction, yellow colour
Clostridium botulinum ATCC 25763	50-100	luxuriant	positive reaction, yellov colour	negative wreaction, no colour change
Clostridium perfringens ATCC 12924	50-100	luxuriant	positive reaction, yellov colour	positive wreaction, yellow colour
Escherichia coli ATCC 35218	50-100	luxuriant	positive reaction, yellov colour	positive wreaction, yellow colour

# **Storage and Shelf Life**

Store below 30°C in tightly closed container and prepared medium below 2-8°C. Use before expiry period on the label.

#### Reference

- 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.

Revision: 1 / 2011

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