

Medium used for the isolation and cultivation of *Clostridium difficile*.

CONTENTS (Liter)

Proteose Peptone	40.0 g
Disodium Phosphate	5.0 g
Monopotassium Phosphate	1.0 g
Magnesium Sulfate	0.1 g
Sodium Chloride	2.0 g
Fructose	6.0 g
Agar	15.0 g
Final pH = 7.4 \pm 0.2 at 25°C	

PROCEDURE

Suspend 69.1 G of powder in 922 mL of distilled or deionized water. Heat to boiling until completely dissolved. Sterilize by autoclave at 121°C for 15 minutes. Cool to 45 - 50°C in water bath. Aseptically add 2 vials of Clostridium Difficile supplement (MB-C1807) and 70 mL of Horse Blood Defibrinated (MB-H1883) or Sheep Blood Defibrinated (MB-S1876). Mix well. Pour into petri dishes.

Clostridium Difficile supplement

1 vial contents (each vial is sufficient for 500 mL of medium)D-Cycloserine0.125 gCefoxitin0.004 g

INTERPRETATION

Clostridium Difficile Agar is a medium used for the isolation and cultivation of *Clostridium difficile*. Proteose peptone provides nitrogen, vitamins, and amino acids. Phosphates are the buffering agents. Magnesium sulfate is a source of ions to stimulate growth. Sodium chloride maintains the osmotic balance of the medium. Fructose is a fermentable carbohydrate used to enhance recovery and growth of *Clostridium difficile*. Agar is the solidifying agent. D-Cycloserine and cefoxitin inhibit the growth of a large variety of microorganisms such as enterobacteria, faecal streptococci, anaerobic non-spore forming Gram-negative bacilli and Clostridium spp. except *Clostridium difficile*. Sheep blood defibrinated may be used instead of horse blood defibrinated but some strains of the organism will show slight reduced growth.

TECHNIC

Inoculate the specimen using a sterile loop to the medium. Incubate at $36 \pm 1^{\circ}$ C for 48 - 72 hours under anaerobic condition. Refer appropriate references for recommended test procedure.

QUALITY CONTROL FOR USE

 $\label{eq:product} \begin{array}{l} \underline{Dehydrated\ medium} \\ \\ Appearance:\ free-flowing,\ homogeneous \\ Color:\ light\ beige \\ \underline{Prepared\ medium} \\ \\ \\ Appearance:\ clear\ to\ slightly\ opalescent \\ Color:\ cherry\ red \\ \\ \\ Incubation\ conditions:\ 36\ \pm\ 1^\circ C\ /\ 48\ -\ 72\ hours\ under\ anaerobic\ condition \\ \end{array}$

Microorganism	ATCC	Growth	Characteristics
Clostridium difficile	9689	good	grayish white colonies
Escherichia coli	25922	partially inhibited	-
Staphylococcus aureus	25923	partially inhibited	-

STORE

The powder is very hygroscopic. Store the powder at room temperature, in a dry environment, in its original container tightly closed and use it before the expiry date on the label. Store prepared medium at 2 - 8°C.

REFERENCES

- 1. Levett (1985) J. Clin. Pathol. 38: 233-234.
- 2. Barlett, J.G. et al. (1978) N. Eng. J. Med., 298 , 531.
- 3. Boriello, S.P. et al (1981) J. Antimicrob. Chemother. 7 Supp. A. 53-62.
- 4. George, R.H. et al (1976) J. Clin. Microbiol. 6, 214-219.

PACKAGE

Cat. No : MB-C1115 Clostridium Difficile Agar

500 G

