It is used for cultivation of *Clostridium difficile* from certain clinical specimens.

**Composition**

**Ingredients**

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Proteose peptone</td>
<td>40.000</td>
</tr>
<tr>
<td>Disodium hydrogen phosphate</td>
<td>5.000</td>
</tr>
<tr>
<td>Potassium dihydrogen phosphate</td>
<td>1.000</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>2.000</td>
</tr>
<tr>
<td>Magnesium sulfate</td>
<td>0.100</td>
</tr>
<tr>
<td>Mannitol</td>
<td>6.000</td>
</tr>
<tr>
<td>Neutral red</td>
<td>0.030</td>
</tr>
<tr>
<td>Sodium taurocholate</td>
<td>1.000</td>
</tr>
<tr>
<td>Cysteine</td>
<td>0.500</td>
</tr>
</tbody>
</table>

**Final pH (at 25°C)**: 7.3±0.2

**Formula adjusted, standardized to suit performance parameters**

**Directions**

Suspend 55.63 grams in 1000 ml distilled water. Heat if necessary 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 rehydrated contents of 1 vial of Clostridium difficile Selective Supplement (FD320). Mix well and dispense into sterile tubes.

**Principle And Interpretation**

*Clostridium difficile* Mannitol Taurocholate Broth Base is used for the primary isolation of *C. difficile* from faecal specimens. The spectrum of disease caused by *Clostridium difficile* (a pathogenic *Clostridium* affecting the bowel) ranges from pseudomembranous colitis (PMC) through antibiotic associated colitis (AAC). It also includes chronic inflammatory bowel diseases, post-operative diarrhoea and non-antibiotic associated diarrhoea. Smith and King first reported the presence of *C. difficile* in human infections.

The medium composition is designed so as to obtain luxuriant growth of *C. difficile*. The selective agents D-cycloserine and cefoxitin used in the medium inhibit the growth of majority of *Enterobacteriaceae* and also *Enterococcus faecalis*, gram-negative anaerobic bacilli and *Clostridium* species other than *C. difficile*, which may be found abundantly in faecal samples. Proteose peptone provides essential growth factors and trace nutrients. Mannitol is the fermentable carbohydrate, fermentation of which leads to acid production, detected by neutral red indicator. Taurocholate and lysozyme are added as spore germination stimulators. Inorganic salts supply the necessary growth requirements. Sodium chloride maintains the osmotic equilibrium.

**Quality Control**

**Appearance**

Cream to yellow homogeneous free flowing powder

**Colour and Clarity of prepared medium**

Red coloured clear solution in tubes.

**Reaction**

Reaction of 5.56% w/v aqueous solution at 25°C. pH: 7.3±0.2

**Cultural Response**

Cultural characteristics observed under anaerobic condition with added Clostridium difficile Selective Supplement (FD320) after an incubation at 35-37°C for 48 hours.

**Cultural Response**

Please refer disclaimer Overleaf.
Organism | Inoculum (CFU) | Growth | Acid |
--- | --- | --- | --- |
**General Response**
Clostridium difficile ATCC 11204 | 50-100 | luxuriant | positive reaction, yellow colour |
Clostridium sporogenes ATCC 11437 | $\geq 10^3$ | inhibited | negative reaction, no colour change |
Clostridium perfringens ATCC 12924 | $\geq 10^3$ | inhibited | negative reaction, no colour change |
Staphylococcus aureus ATCC 25923 | $\geq 10^3$ | inhibited | negative reaction, no colour change |
Bacteroides fragilis ATCC 25285 | $\geq 10^3$ | inhibited | negative reaction, no colour change |
Streptococcus faecalis ATCC 29212 | $\geq 10^3$ | inhibited | negative reaction, no colour change |
Proteus mirabilis ATCC 25933 | $\geq 10^3$ | inhibited | negative reaction, no colour change |

**Storage and Shelf Life**
Store below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label.

**Reference**

Revision: 0 / 2014