Modified Duncan Strong (DS) Medium

Modified Duncan Strong (DS) Medium is used for isolation and differentiation of \textit{Clostridium perfringens} from other clostridia from foods on the basis of raffinose fermentation.

**Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Proteose peptone</td>
<td>15.000</td>
</tr>
<tr>
<td>Yeast extract</td>
<td>4.000</td>
</tr>
<tr>
<td>Sodium thioglycollate</td>
<td>1.000</td>
</tr>
<tr>
<td>Disodium phosphate</td>
<td>10.000</td>
</tr>
<tr>
<td>Raffinose</td>
<td>4.000</td>
</tr>
<tr>
<td>Final pH (at 25°C)</td>
<td>7.8±0.2</td>
</tr>
</tbody>
</table>

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

**Directions**

Suspend 34 grams in 1000 ml distilled water and mix thoroughly. Heat if necessary to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Dispense into sterile tubes. Check one or two tubes for measuring the pH.

**Principle And Interpretation**

\textit{Clostridium perfringens}, a gram-positive, rod shaped, anaerobic, spore-forming bacteria, is the major cause of food poisoning in humans. A heat labile enterotoxin produced only by sporulating cells induces the major symptoms of diarrhea in perfringens poisoning. \textit{C. perfringens} is commonly found in raw meats, poultry, dehydrated soups and sauces, raw vegetables and certain other foods or food ingredients.

Modified Duncan Strong (DS) Medium is formulated as per Duncan and Strong (1), and is recommended by APHA (2) for the isolation and differentiation of \textit{C. perfringens} from other Clostridia from foods on the basis of raffinose fermentation. It is also recommended for the rapid detection of the \textit{Clostridium perfringens} enterotoxin (3).

Proteose peptone and yeast extract provide nitrogenous compounds and other nutrients for bacterial growth. Sodium thioglycollate helps to create anaerobic conditions suitable for clostridial growth. Disodium phosphate acts as a buffering agent. Raffinose in the medium is fermented by \textit{C. perfringens} to produce acid within 72 hours, but not by culturally similar species like \textit{Clostridium baratii}, \textit{Clostridium celatum}, \textit{Clostridium sardiniense} etc. To test for acid, transfer 1 ml of culture to a test tube or spot plates and add 2 drops of 0.04% bromothymol blue. A yellow colour indicates acid production.

Inoculate about 2 gm of the food sample into 15-20 ml of Chopped Liver Broth (M606). After an incubation at 35-37°C for 20-24 hours, tubes showing turbidity are streaked on Perfringens Agar Base (M837) containing Egg Yolk Emulsion (FD045) to obtain presumptive \textit{C. perfringens}. These presumptive colonies can be confirmed by inoculating into Motility Nitrate Medium, Buffered (M630), Lactose Gelatin Medium (M628) and Modified Duncan Strong (DS) Medium (M1237) (2).

**Quality Control**

**Appearance**
Cream to yellow homogeneous free flowing powder

**Colour and Clarity of prepared medium**
Yellow coloured clear solution without any precipitate

**Reaction**

Reaction of 3.4% w/v aqueous solution at 25°C. pH : 7.8±0.2

**pH**
7.60-8.00

**Cultural Response**

M1237: Cultural characteristics observed after an incubation at 35-37°C for 48-72 hours.

Please refer disclaimer Overleaf.
<table>
<thead>
<tr>
<th>Organism</th>
<th>Inoculum (CFU)</th>
<th>Growth</th>
<th>Raffinose fermentation</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Clostridium perfringens</em> ATCC 12924</td>
<td>50-100</td>
<td>good-luxuriant</td>
<td>positive reaction</td>
</tr>
<tr>
<td><em>Clostridium sporogenes</em> ATCC 11437</td>
<td>50-100</td>
<td>good-luxuriant</td>
<td>negative reaction</td>
</tr>
</tbody>
</table>

**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**