

# TM 840 - REINFORCED CLOSTRIDIAL BROTH W/O AGAR

#### **INTENDED USE**

For cultivation and enumeration of Clostridia.

#### PRODUCT SUMMARY AND EXPLANATION

Reinforced Clostridial Broth is formulated by Hirsch and Grinsted. It can be used to initiate growth from small inocula and to obtain the highest viable count of Clostridia. Barnes and Ingram used the broth medium for diluting an inoculum of vegetative cells of Clostridium perfringens. It can be used in studies of spore forming anaerobes, especially Clostridium butyricum in cheese, for enumeration of Clostridia in tube dilution counts or for preparation of plates for isolation. Other spore forming anaerobes, Streptococci and Lactobacilli also grow in this media. This is a nonselective enrichment media.

### **COMPOSITION**

Ingredients	Gms / Ltr	
Casein enzymic hydrolysate	10.000	
Beef extract	10.000	
Yeast extract	3.000	
Dextrose	5.000	
Sodium chloride	5.000	
Sodium acetate	3.000	
Starch, soluble	1.000	
L-Cysteine hydrochloride	0.500	

## **PRINCIPLE**

This medium consists of Casein enzymic hydrolysate, yeast extract, beef extract, starch, L-cysteine and sodium acetate which provide all the necessary nutrients for the growth of Clostridia. Dextrose is a fermentable carbohydrate in the medium while sodium chloride maintains osmotic equilibrium. This media can be made selective by addition of 15-20 mg Polymyxin B per litre of media.

## **INSTRUCTION FOR USE**

- Dissolve 37.5 grams in 1000 ml distilled water.
- Heat if necessary to dissolve the medium completely. Dispense as desired.
- Sterilize by autoclaving at 10 psi pressure (115°C) for 15 minutes.

## **QUALITY CONTROL SPECIFICATIONS**

Appearance of Powder : Light yellow coloured homogeneous free flowing powder. Appearance of prepared medium : Medium amber coloured slightly opalescent solution.

pH (at 25°C)  $: 6.8 \pm 0.2$ 

# **INTERPRETATION**

Cultural characteristics observed in an anaerobic atmosphere after incubation.













Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Incubation Temperature	Incubation Period
Clostridium butyricum	13732	50-100	Good-luxuriant	35-37°C	24-48 Hours
Bacteroides vulgatus	8482	50-100	Good-luxuriant	35-37°C	24-48 Hours
Bacteroides fragilis	23745	50-100	Good-luxuriant	35-37°C	24-48 Hours
Clostridium perfringenes	12924	50-100	Good-luxuriant	35-37°C	24-48 Hours

#### **PACKAGING:**

In pack size of 100 gm and 500 gm bottles.

# **STORAGE**

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

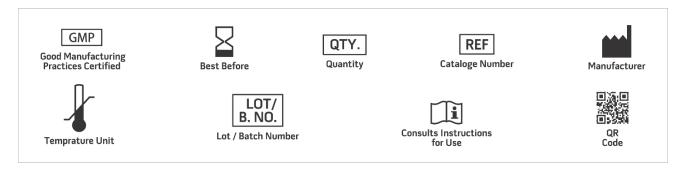
**Product Deterioration:** Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

# **DISPOSAL**

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

# **REFERENCES**

- 1. Hirsch and Grinsted, 1954, J. Dairy Res., 21:101.
- 2. Barnes and Ingram, 1956, J. Appl. Bact., 19:117.
- 3. Lewis and Angelotti (Eds.), 1964, Examination of Foods for Enteropathogenic and Indicator Bacteria, Dept. of HEW, PHS Publication, 1142, Washington.















**NOTE:** Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices. \*For Lab Use Only Revision: 08 Nov., 2019









