

## REINFORCED CLOSTRIDIAL MEDIUM

Medium for the cultivation and enumeration of *Clostridium* and other anaerobes, according to E.P.

### TYPICAL FORMULA (g/L)

Beef Extract	10.00
Dextrose	5.00
Yeast Extract	3.00
Soluble Starch	1.00
Casein Peptone	10.00
Sodium Chloride	5.00
Sodium Acetate	3.00
L-Cysteine Hydrochloride	0.50
Agar	0.50
Final pH 6.8 ± 0.2	

### DESCRIPTION

REINFORCED CLOSTRIDIAL MEDIUM is a semisolid medium formulated by Hirsch and Grinstead. It is used for cultivating and enumerating clostridia, other anaerobes, and other species of bacteria from foods and clinical specimens, according to E.P.

### PRINCIPLE

Yeast extract provide nitrogen, vitamins, amino acids and carbon. The Casein Peptone and Beef extract are sources of nitrogen, vitamins and amino acids and carbon. Dextrose is a complex carbohydrate and Sodium chloride maintains the osmotic balance. In low concentrations, the Starch detoxifies metabolic by-products. L-Cysteine hydrochloride is the reducing agent and Sodium acetate is the buffer. The small amount of agar makes the medium semisolid. The medium is a nonselective enrichment medium and grows various anaerobic and facultative bacteria when incubated anaerobically.

### PREPARATION

Suspend 38 grams of the medium in one liter of distilled water. Heat with frequent agitation until completely dissolved. Dispense into tubes and sterilize at 121 °C for 15 minutes.

### TECHNIQUE

Inoculate tubes with a pure culture by stabbing the center of the column of medium to greater than half the depth. Incubate tubes for 18-48 hours at 35 ± 2 °C in an anaerobic atmosphere.

### INTERPRETATION OF RESULTS

The turbidity indicates bacterial growth.

### STORAGE

The powder is very hygroscopic: store the powder at 10-30°C, in a dry environment, in its original container tightly closed until the expiry date indicated on the label or until signs of deterioration or contamination are evident.  
Store prepared media at 2-8°C.

### WARNING AND PRECAUTIONS

The product is not classified as hazardous by current legislation and does not contain harmful substances in concentrations of ≥ 1%.  
The product must be used only by properly trained operators.

### DISPOSAL OF WASTE

Disposal of waste must be carried out according to national and local regulations in force.

### REFERENCES

1. Hirsch A. and Grinstead E. (1954) J. Dairy Res. 21. 101-110
2. Mundt J. O. and Jones V. W. (1952) Bact. Proc. p. 106
3. Gibbs B. M. and Hirsch A. (1956) J. Appl. Bact. 19. 129-141
4. Gibbs B. M. and Freame B. (1965) J. Appl. Bact. 28. 95-111
5. The Microbiology of Water 1994 Part 1 – Drinking Water. Report on Public Health and Medical Subjects Number 71: Subjects Number 71: Methods for the Examination of Waters and Associated Materials. HMSO London.
6. Weenk G., Fitzmaurice E. and Mossel D.A.A. (1991) J. Appl. Bact. 70. 135-143
7. European Pharmacopoeia. (2002) 4<sup>th</sup> Edition.



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## PRODUCT SPECIFICATIONS

### NAME

REINFORCED CLOSTRIDIAL MEDIUM

### PRESENTATION

Dehydrated culture medium

### STORAGE

10-30 °C

### PACKAGING

Code	Content	Packaging
610340	500 gr	500 gr of powder in plastic bottle
610340	100 gr	100 gr of powder in plastic bottle

### pH OF THE MEDIUM

6.8 ± 0.2

### USE

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### TECHNIQUE

Refer to technical sheet of the product.

### APPEARANCE OF THE MEDIUM

Dehydrated medium

Appearance: free-flowing, homogeneous.

Colour: light tan

Prepared medium

Appearance: slightly opalescent

Colour: amber, opalescent

### SHELF LIFE









4 years

### QUALITY CONTROL

- Control of general characteristics, label and print
- Sterility control  
7 days at 25 ± 1°C, in aerobiosis  
7 days at 36 ± 1°C, in aerobiosis
- Microbiological control  
Inoculum for productivity: 10-100 UFC/ml  
Inoculum for specificity: ≤ 10<sup>4</sup> UFC/ml  
Incubation conditions: 18-48 hours at 35 ± 2°C, anaerobically

Microorganisms		Growth
<i>Bacteroides fragilis</i>	ATCC 23745	Good
<i>Clostridium bifermentans</i>	ATCC 19299	Good
<i>Clostridium perfringens</i>	ATCC 13124	Good
<i>Clostridium perfringens</i>	ATCC 10543	Good

### TABLE OF SYMBOLS

 LOT	Batch code	 Temperature limitation	 Manufacturer	 Contains sufficient for <n> tests
 REF	Catalogue number	 Keep away from heat	 Use by	 Caution, consult accompanying documents



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