



# Technical Data

## Clostridium Broth Base

M1315

### Intended Use:

Recommended for identification of spores of *Clostridium tyrobutyricum* which is usually responsible for "late blowing" in cheese.

### Composition\*\*

Ingredients	Gms / Litre
Casitose ▲	15.000
HM extract	10.000
Yeast extract	5.000
Sodium acetate	5.000
L-Cysteine	0.500
Final pH ( at 25°C)	6.0±0.2

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

▲- Equivalent to Casein peptone

### Directions

Suspend 35.5 grams in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely. Add 10 ml of 50% sodium lactate. Cool at 45-50°C. Mix well and dispense into tubes. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

### Principle And Interpretation

Ripening of cheese under controlled conditions of temperature and humidity determine the final flavour and body characteristics of the product. Microbial spoilage in cheese is generally limited because of the combined effect of acid and salt and is less likely in the lower moisture cheese. Spores of *Clostridium tyrobutyricum* in the milk used for the manufacture of Emmentaler, Edam and Gouda can survive the heat treatment used for cheese milk and cause late gas formation (blowing defect) and related off-flavours during ripening (9). Even low spore densities of this anaerobe in milk used for cheese production can bring about this phenomenon, if the growth conditions are suitable. Clostridium Broth Base is recommended for the identification of spores of *C. tyrobutyricum*.

*C. tyrobutyricum* ferments lactate and acetate to butyrate, CO<sub>2</sub> and H<sub>2</sub> (3,4). As against *C. butyricum*, *C. tyrobutyricum* grows in media with lower acidic pH (2,7). Low pH value of the media helps in inhibiting other microbial flora thereby favoring growth of *C. tyrobutyrium*. Casitose, HM extract and yeast extract provide the essential nutrients mainly the nitrogen compounds whereas L-cysteine promotes the growth of Clostridial species by maintaining a low oxygen tension in the medium.

### Type of specimen

Dairy samples - Gouda cheese, Gruye're cheeses (Natural or processed cheese.) (8)

### Specimen Collection and Handling:

For dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (1,10). After use, contaminated materials must be sterilized by autoclaving before discarding.

### Warning and Precautions :

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

### Limitations :

1. Further biochemical and serological tests must be carried out for further identification.
2. Some organism may show poor growth due to nutritional variation.

## Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

## Quality Control

### Appearance

Cream to yellow homogeneous free flowing powder

### Colour and Clarity of prepared medium

Yellowish brown coloured, clear solution without any precipitate

### Reaction

Reaction of 3.55% w/v aqueous solution at 25°C. pH : 6.0±0.2

### pH

5.80-6.20

### Cultural Response

Cultural characteristics observed under anaerobic condition, after an incubation at 35-37°C for upto 7 days

Organism	Inoculum (CFU)	Growth	Gas
<i>Clostridium perfringens</i> ATCC 10543 (00174*)	50-100	luxuriant	positive reaction
<i>Clostridium tyrobutyricum</i> ATCC 25755	50-100	luxuriant	positive reaction
<i>Escherichia coli</i> ATCC 25922 (00013*)	50-100	good	variable reaction
<i>Pseudomonas aeruginosa</i> ATCC 27853 (00025*)	≥10 <sup>4</sup>	inhibited	
<i>Staphylococcus aureus</i> subsp. <i>aureus</i> ATCC 25923 (00034*)	50-100	good	negative reaction

Key : \*Corresponding WDCM numbers.

## Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

## Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (5,6).

## Reference

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