

### Specification

Solid medium used for the biochemical confirmation of *Clostridium perfringens*, according to ISO 7937 standard.

### Formula \* in g/L

Tryptone.....	15,00
Yeast Extract.....	10,00
Lactose.....	10,00
Gelatin.....	120,00
Phenol red.....	0,05

Final pH 7,50 ±0,2 at 25 °C

\* Adjusted and /or supplemented as required to meet performance criteria

### Directions

Dissolve 155 g of powder in 1 L distilled water, heating if necessary. Adjust pH to 7.5 ± 0.2 (add 10-15 ml of sodium carbonate solution at 10%). Dispense in tubes in suitable volumes and sterilize at 121°C for 15 minutes. If not used the same day, store in the refrigerator. Just prior to use heat in a boiling water bath or flowing steam for 15 minutes, then cool rapidly to the incubation temperature. Discard unused medium 1 month after preparation. Note: Before preparing the medium, shake the container vigorously until the powder is homogeneous. The different granulometry of gelatin, can cause disaggregation of the medium in transport.

### Description

This medium with the Nitrate Motility Medium (Art. No. 03-612) are used in the confirmation technique for *Clostridium perfringens* according to the 7937:2004 ISO Standard.

### Technique

Inoculate each selected colony from the Tryptose-Sulfite-Cycloserine Agar (Art. No. 01-278) into the Lactose Gelatine Medium and incubate under anaerobic conditions for 24 hours at 37°C. Examine the tubes of Lactose Gelatine Medium for the presence of gas and a yellow colour due to acid formation indicating fermentation of lactose. Chill the tubes for 1 hour at 5 ± 3°C and check for gelatine liquefaction. If the medium has solidified, re-incubate for an additional 24 hours and again check for gelatine liquefaction.

### Interpretation

Non-motile bacteria that produce black colonies in Tryptose Sulfite Cycloserine Agar (Art. No. 01-278) liquefy gelatine in 48 hours are considered to be *Clostridium perfringens*.

Cultures that show a faint reaction for nitrite should be eliminated, since *C. perfringens* consistently gives an intense and immediate reaction.

### Quality control

**Incubation temperature:** 35°C ±2,0      **Incubation time:** 24 h

**Inoculum:** Pure cultures using and inoculating needle

### Microorganism

*Staphylococcus aureus* ATCC® 25923  
*Escherichia coli* ATCC® 25922  
*Clostridium perfringens* ATCC® 10543  
*Clostridium perfringens* ATCC® 13124  
*Clostridium sporogenes* ATCC® 11437

### Growth

Poor  
 Good  
 Good  
 Good  
 Good

### Remarks

-  
 -  
 L (+) Gas (+) Gelatinase (+)  
 L (+) Gas (+) Gelatinase (+)  
 L (+) Gas (D) Gelatinase (+)

### References

· ISO 7937 Standard (2004) Microbiology of food and animal feeding stuffs - Horizontal method for the enumeration of *Clostridium perfringens* - Colony count technique.

### Storage

For laboratory use only. Keep tightly closed, away from bright light, in a cool dry place (+4 °C to 30 °C).