

Specification

Fluid medium used for the identification of *Clostridium perfringens* according to ISO standard 7937:2004.

Formula * in g/L

Meat extract.....	3.00
Peptone.....	5.00
Potassium nitrate.....	1.00
Bi-Sodium phosphate.....	2.50
Galactose.....	5.00
Agar.....	5.00

Final pH 7,3 ±0,2 at 25 °C

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

Directions

Suspend 21,5 g of powder in 1 L of distilled water containing 5 mL of glycerol. Bring to the boil and distribute in suitable containers. Sterilize in the autoclave at 121°C for 15 minutes.

Description

This medium is produced according to the formulation of the US Food & Drug Administration and the ISO Standard 7937 for the identification of *Clostridium perfringens* in cosmetics and food.

Technique

The final containers, usually flat or standard tubes, are degassed by heating in a water bath à 100°C for 10 minutes. Then, they are cooled and they are inoculated with a needle in the middle of the medium. The inocula must be obtained from black colonies growth on TSN Agar. The inoculated tubes are incubated à 37°C for 18-20 hours without any seal or reduced atmosphere.

If the growth in this medium appears à a deep of 5-7 mm from the surface this is evidence of anaerobiosis and the absence of motility is shown by a clear and limpid growth along the streak.

For verification of nitrate reduction, several drops of a mixture of Nitrate A Reagent and Nitrate B Reagent must be added. Development of a cherry red colour shows a positive nitrate to nitrite reduction.

Clostridium perfringens is an anaerobic, non-motile and nitrate-reducing microorganism.

Quality control

Incubation temperature: 37°C ±1

Incubation time: 18 -20 h

Inoculum: Pure cultures using and inoculating needle, according to ISO 11133:2014/Amd 1:2018 & Adm 2:2020

Microorganism
Growth
Remarks

<i>Clostridium perfringens</i> ATCC® 13124	Good	Nitrate (+). Nonmotile
<i>Clostridium sporogenes</i> ATCC® 11437	Good	Nitrate (-). Motile
<i>Clostridium perfringens</i> ATCC® 10543	Good	Nitrate (+). Nonmotile

References

- FDA (Food and Drug Administrations) (1998) Bacteriological Analytical Manual. 8th ed. Revision A. AOAC International. Gaithersburg. MD. USA.
- ISO 7937 Standard (2004) Microbiology of food and animal feeding stuffs - Horizontal method for the enumeration of *Clostridium perfringens* - Colony count. Technique.
- ISO 11133:2014/ Adm 1:2018/ Adm 2:2020/ Microbiology of food, animal feed and water. Preparation, production, storage and performance testing of culture media.

Storage

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