

# 66759 Lactose sulfite medium (LS) NutriSelect® Plus

For the confirmation of *Clostridium perfringens* based on lactose fermentation and production of hydrogen sulfide according to ISO 7937:2004.

### **Composition:**

Ingredients	Grams/Litre	
Cysteine hydrochloride	0.3	
Pancreatic digest of casein	5.0	
Sodium chloride	2.5	
Yeast extract	2.5	
Lactose monohydrate	10.0	

Final pH 7.1 +/- 0.2 at 25°C

Store granulated media between 10-30°C in tightly closed container and the prepared medium at 20-30°C. Avoid freezing and overheating. Once opened keep powdered medium closed to avoid hydration. Use before expiry date on the label.

Appearance(color): Faint yellow to light yellow to light beige, free flowing powder

Color and Clarity: Very light to dark yellow and very light brown-yellow to dark brown-yellow

coloured, clear to slightly hazy

## **Directions:**

Suspend 20,3 grams of the medium in one liter of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Dispense 8 ml portions into tubes with Durham gas collecting tubes for gas detection. Sterilize in autoclave at 121 °C for 15 minutes. Before using add to each tube 0,5 ml of a 12 g/L solution of sodium metabisulfite and 0,5 ml of a solution of 10 g/L of ferric ammonium citrate. Both solutions have to be freshly prepared and sterilized.

# ISO 7937:2004

- Inoculate each presumtive colony into Thioglycollate Medium.
- Incubate under anaerobic conditions at 37 °C for 18-24 hours. The turbidity should be 1-2 F.T.U.
- Transfer 5 drops of the thioglycollate culture to the Lactose Sulfite Broth.
- Incubate aerobically at 46 °C for 18 to 24 hours
- Examine the tubes of Lactose Sulfite Broth for the production of gas and the presence of a black color (iron sulfite precipitate).

#### **Principle and Interpretation:**

Pancreatic digest of casein and yeast extract provides nitrogenous, carbonaceous compounds and other essential growth nutrients like vitamine B. Sodium chloride maintains the osmotic equilibrium and Lactose is the fermentable carbohydrate source. *C. perfringens* can ferment lactose and release CO<sub>2</sub> which is visible as gas in the Durham tube. Cysteine hydrochloride is the reducing agent. Sodium bisulfite is converted from *C. perfringens* to hydrogen sulfide which reacts with ferric ammonium citrate resulting in a black precipitate (iron sulfite).



Cultural characteristics observed after an incubation aerobically at 46°C for 18-24 hours.

Organisms	Growth	Characteristic reaction
Clostridium perfringens ATCC 13124	+++	Gas prduction (+), blackennig (+)
Clostridium perfringens (NCTC 13170)	+++	Gas prduction (+), blackennig (+)
Clostridium perfringens (WDCM 00007)	+/-	Gas prduction (+), blackennig (+)

#### References:

- 1. ISO Standard 7937 Microbiology of food and animals feeding stuffs. Horizontal method for enumeration of Clostridium perfringens. Colony count technique.
- 2. ISO 11133:2014. Microbiology of food, animal feed and water. Preparation, production, storage and performance testing of culture media

#### **Precautions and Disclaimer**

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

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