

LACTOSE SULFITE BROTH BASE EUROPEAN PHARMACOPOEIA

Selective medium recommended for the detection and enumeration of spores of *Clostridium perfringens* in food products.

CAT Nº: 21009

FORMULA IN g/l

Lactose Monohydrate	10.00	Yeast Extract	2.50
Pancreatic Digest of Casein	5.00	Cysteine Hydrochloride	0.30
Sodium Chloride	2.50		

Final pH 7.1 ± 0.2 at 25°C

PREPARATION

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 into tubes with Durham gas collecting tubes for gas detection. Sterilize in autoclave at 121°C for 15 minutes. Store at 4°C. Before using add to each tube 0.5 ml of a 12 g/liter solution of Sodium Metabisulfite and 0.5 ml of a solution of 10 g/liter of Ferric Ammonium Citrate. Both solutions have to be freshly prepared and sterilized. The color of the prepared medium is amber.

The dehydrated medium should be homogeneous, free-flowing and beige in color. If there are any physical changes, discard the medium.

USES

LACTOSE SULFITE BROTH BASE is a selective medium used to detect and enumerate spores of *Clostridium perfringens* based on lactose fermentation and production of hydrogen sulfide.

The nutrient base provides optimal conditions for the development of Clostridia. Casein peptone provides nitrogen, vitamins, minerals and amino acids essential for growth. Yeast extract is the source of vitamins, particularly of the B-group. Lactose is a complex carbohydrate energy source. Sodium chloride supplies essential electrolytes for transport and osmotic balance. Cysteine hydrochloride is the reducing agent.

European Pharmacopoeia recommends to prepare samples using 1:100 and 1:1000 dilutions with Buffered Peptone Water (Cat. 21401). Determine the most

probable number of bacteria (MPN) in tubes or other suitable containers with small Durham tubes. Mix the sample and the medium with minimum shaking and incubate at 45.5 - 46.5°C for 24–48 hours.

Colonies producing hydrogen sulfide are characterized by a blackening due to the reaction of Sodium bisulfite and the Ferric ammonium citrate salt. The containers showing a blackening and abundant formation of gas in the Durham tube (at least 1/10 of the volume) indicate the presence of *C. perfringens*. Estimate the most probable number using the appropriate table (MPN Table).

0.30 MICROBIOLOGICAL TEST

The following results were obtained in the performance of the medium, from type cultures, with additives added, after incubation at a temperature of 45.5 – 46.5°C and observed after 24 - 48 hours. According to European Pharmacopoeia 6.5.

Microorganisms	Growth	Gas production	Blackening
<i>Clostridium perfringens</i> ATCC 13124	Good	+	+

BIBLIOGRAPHY



European Pharmacopoeia 6th Edition 2007

STORAGE

Once opened keep powdered medium closed to avoid hydration.

