

## Specification

Solid medium for the detection of *Clostridium perfringens* in water and food samples.

## Presentation

	Packaging Details	Shelf Life	Storage
30 Prepared Plates 55 mm Plates for filtration purposes with: 9 ± 1 ml	1 box containing: 5 plastic bags with 6 plates of 55 mm/ bag.	6 months	2-25°C

## Composition

Composición (g/l):

Sodium sulfite.....	0.50
Polymyxin B sulfate.....	0.01
Sodium Sulfadiazine.....	0.12
Yeast Extract.....	10.0
Iron(III) Citrate.....	0.50
Casein Peptone.....	15.00
Agar.....	13.90

## Description /Technique

### Description

SPS Agar (Sulfite Polymyxin Sulfadiazine) is a modification of the original Wilson & Blair medium for the detection of clostridia. The present medium better the formulation of Mossel and also the later modification of Angelotti et al.. It achieves a higher selectivity for *C. perfringens* with the addition of Sulfadiazine and Polymyxin.

The differential system consists of sodium sulfite and ferric citrate which allows the detection of sulfite reducing organisms, which form black colonies due to ferrous sulfide precipitate.

### Technique

The usual technique for the use of this medium is as follows:

Collect, dilute and prepare samples and volumes to be filtered as required according to specifications, directives, official standard regulations and/or expected results.

Filter the sample through a 0.45 µm pore membrane and apply it onto the surface of the agar SPS.

Incubate the plates anaerobically at 35±/- 2°C for 24-48h. If a more selective culture medium is desired to incubate at 44 ° C ± 1

(Incubation times greater than those mentioned above or different incubation temperatures may be required depending on the sample, on the specifications,...)

After incubation, enumerate all the colonies that have appeared onto the surface of the membrane.

After incubation, enumerate the colonies with a black iron sulfide precipitate.

90% of the black colonies which are formed can usually be attributed to *Clostridium perfringens*.

Since the medium is not extremely selective, it is advisable to verify black colonies by checking that they are Gram positive sporulated non-motile organisms incapable of reducing nitrates to nitrites.

Most clostridia are sulfite reducers. Among them are *C. perfringens* and *C. botulinum* which along with *C. bifermentans* are the species most frequently involved in food poisoning.

**Quality control****Physical/Chemical control**

Color : Straw-coloured yellow      pH:  $7 \pm 0.2$  at 25°C

**Microbiological control**

Membrane Filtration /Practical range  $100 \pm 20$  CFU; Min. 50 CFU (Productivity)./ $10^4$ - $10^6$  CFU for Selectivity.

Anaerobiosis. Incubation at  $35 \pm 2.5^\circ\text{C}$ , reading after 24-48 hours

**Microorganism****Growth**

*Clostridium perfringens* ATCC® 13124, WDCM 00007, NCTC® 8237      Good - H<sub>2</sub>S positive . Black colonies

*Clostridium perfringens* ATCC® 10543, WDCM 00174      Good - H<sub>2</sub>S positive . Black colonies

*Escherichia coli* ATCC® 8739, WDCM 00012      Inhibited

**Sterility Control**

Incubation 48 hours at 30-35°C and 48 hours at 20-25°C: NO GROWTH

Check at 7 days after incubation in same conditions

**Bibliography**

- ANGELOTTI, HALL, FOSTER & LEWIS (1962) Quantisation of *Clostridium perfringens* in foods. Appl. Microbiol., 10:193.
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- ISO 11133:2014/ Adm 1:2018. Microbiology of food, animal feed and water. Preparation, production, storage and performance testing of culture media.
- MOSSEL, D.A.A. (1959) Enumeration of sulfite-reducing bacteria occurring in foods. J. Sci. Food Agric. 19:662.