

Specification

Sterile selective supplement used for *Listeria* isolation in food samples.

Presentation

10 Freeze-dried vials
Vial
with: 9 ± 0.1 g

Packaging Details

$22 \pm 0.25 \times 55 \pm 0.5$ mm glass vials, tag labelled, White plastic cap - 10 vials per box.

Shelf Life

49 months

Storage

2-25 °C

Composition

Compositon (g/vial)

Cyclohexymide.....	0.2000
Colistin sulphate.....	0.0100
Acriflavine.....	0.0025
Cefotetan.....	0.0010
Phosphomycin sodium salt.....	0.0050

NOTE: Each vial is enough to supplement
500ml of OXFORD medium Base.

Reconstitute the original freeze-dried vial

by adding:

Sterile solvent (50% Ethanol/water)..... 9 ml

Description /Technique

Description:

Listeria Agar Base Oxford (Cat. 1133) is a selective medium for *Listeria* according to the Oxford formula and it is recommended for the detection of *Listeria monocytogenes* from clinical samples and food products. It is used for a direct sample inoculation or for confirmation after using *Listeria* Enrichment Broth Base (Cat.1120).

All *Listeria* species hydrolyze the esculin to esculetin, which reacts with the iron ions producing black colonies and a blackening of the medium. Another advantage of this medium is that peptones and maize starch provides a rich nutrient base for growth, and the addition of Ferric ammonium citrate improves the growth of *L. monocytogenes*. Lithium chloride is an inhibiting agent, together with the other antibiotics from the supplement, which inhibit the growth of gram-negative bacteria and a large part of Gram-positive ones. Cycloheximide inhibits yeasts.

Technique:

Aseptically reconstitute 1 vial with 9 ml of 1:1 sterile distilled water/Ethanol. Mix gently until complete dissolution. Aseptically add to 500 ml of *Listeria* Agar Base Oxford (Cat. 1133), autoclaved and cooled to 50 °C. Mix well and distribute into sterile containers.

Instructions for use:

For clinical diagnosis, the type of sample is amniotic fluid.

- Inoculate on the surface making parallel striae with the handle or hyssop. - Incubate in aerobic conditions at 37 °C for 48 hours.

- Reading and interpretation of the results.

For the detection and enumeration of *Listeria monocytogenes* and *Listeria* spp. according to ISO 11290:

- Weigh 25 g (or 25 ml) of the sample and add 225 ml of 1/2 Fraser Broth (Cat. 1183). Homogenize and incubate at 30 °C for 25±1 hours.

- Inoculate 0,1 ml of culture of the 1/2 Fraser Broth incubated (independently of the color) in 10 ml of Fraser Broth (Cat.1182). Incubate at 37 °C for 24±2 hours under aerobic conditions.

- The primary enrichment culture is inoculated on the *Listeria* Agar surface according to Ottaviani and Agosti (Cat. 1345) and in another selective medium of the laboratory (Oxford), to obtain the well separated colonies.

- From the secondary enrichment culture, repeat the procedure, inoculate the surface of the *Listeria* Agar according to Ottaviani and Agosti (Cat. 1345) and the Oxford Agar. Incubate for a total of 48±2 h.

- Select presumptive colonies and carry out confirmatory tests for *L. monocytogenes* or *Listeria* spp.

Although typical colonies of *L. monocytogenes* are almost always visible after 24 hours of incubation, the incubation should be continued for a further 24 hours to obtain strains of slower growth.

Quality control

Physical/Chemical control

Color : Yellow- orangey

pH: at 25°C

Microbiological control

Distribute the complete medium, cooled at 50°C, in plates

Inoculate: Practical range 100 ± 20 CFU. min. 50 CFU (productivity)/ 10⁴-10⁶ (selectivity).

Aerobiosis. Incubation at 35°C ± 2 °C, reading at 24-48 hours

Microorganism

Escherichia coli ATCC® 25922, WDCM 00013*L. monocytogenes* ATCC® 13932, WDCM 00021*Listeria monocytogenes* ATCC® 35152*Enterococcus faecalis* ATCC® 29212, WDCM 00087

Sterility Control

Add 5 ml of the sample to:

100 ml TSB and 100 ml Thioglycollate.

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

Growth

Inhibited

Good - Esculin Positive reaction

Good - Esculin Positive reaction

Inhibited

Bibliography

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