ALLIANCE BIO EXPERTISE

Reference: DSHB3051 **A.B.E. - Technical Data Sheet**

Product: **Listeria Oxford Selective Supplement**

Specification

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

Presentation

	Packaging Details	Shelf Life	Storage
10 Freeze dried vials Vial with: 6 ± 0.1 g	23x60 mm glass vials, tag labelled, White plastic cap - 10 vials per box.	49 months	2-25 ºC

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 500 ml of Oxford medium Base.

Reconstitute the original freeze-dried vial by adding:
Sterile solvent (50% Ethanol/water)..... 9 ml

Description /Technique

Description:

Listeria Selective Supplement (Oxford Formulation) is added to Oxford agar base in order to obtain a complete selective medium for the detection of *Listeria monocytogenes* from clinical and food specimens.
Listeria monocytogenes plays an important role in human and animal illness and the sources of infections are numerous.
In the last years the lack of an effective selective medium has been a gap in the detection of *Listeria*, as it can be easily and completely overgrown by competing flora.
With his supplement, which supplies the selective inhibitory components acriflavine, colistin sulphate, cefotetan, cycloheximide and fosfomycin, the competing flora is inhibited. *Listeria monocytogenes* is differentiated becasue it hydrolyses aesculin, producing black zones around the colonies .
Gram-negative bacteria are completely inhibited and also most of the unwanted Gram-positive species. Some strains of enterococci grow poorly and exhibit a weak aesculin reaction, usually after 40 hours incubation. Some staphylococci may grow as aesculin-negative colonies.

Technique:

Collect, dilute and prepare samples and volumes as required according to specifications, directives, official standard regulations and/or expected results.
Reconstitute the vial with 9 ml of sterile diluent (50% Ethanol:water) in aseptic conditions and add it to 500 ml of Oxford Agar base cooled to 50ºC.
Do not overheat once suplemented.
Pour the complete medium into Petri dishes and, once solidified on a flat surface, spread the plates by streaking or spyral method.
Incubate the plates in aerobic atmosphere at 37 ± 1ºC for 44 ±4h.
Incubation times longer than those mentioned above or different incubation temperatures may be requied depending on the sample or the specifications.
After incubation, enumerate all the colonies that have appeared onto the surface of the agar, observing any blackening of the medium due to esculin hydrolysis, typical for Listeria strains.
Presumptive isolation of *Listeria* must be confirmed by further microbiological and biochemical tests.

Quality control

Physical/Chemical control

Color : Yellow- orangey

Microbiological control

Reconstitute 1 vial as indicated in COMPOSITION; shake and dissolve completely

Add 1 vial to 500 ml of medium base. DO NOT HEAT once supplemented.

Inoculate 30-300 CFU (productivity) 1.000-10.000 CFU (selectivity)

Analytical methodology according to ISO 11133:2014/A1:2018; A2:2020.

Distribute the complete medium, cooled to 50 ºC, into 90 mm plates

Incubate according instructions for complete medium indicated in COMPOSITION.

Aerobiosis. Incubation at 37 ± 1 ºC, reading after 24/44 ± 4 h

Microorganism	Growth
L. monocytogenes ATCC® 13932, WDCM 00021	Good - Esculin Positive reaction
Escherichia coli ATCC® 25922, WDCM 00013	Inhibited
Enterococcus faecalis ATCC® 29212, WDCM 00087	Inhibited
L. monocytogenes ATCC® 35152, WDCM 00109	Good - Esculin Positive reaction

Sterility control

Add 5mL of the sample to 100 mL of TSB and to 100 mL Thioglycollate.
Incubation 48 h at 30-35 ºC and 48 h at 20-25 ºC: NO GROWTH.
Check at 7 days after incubation in same conditions.

Bibliography

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