

Specification

A sterile selective supplement used for the isolation of *Listeria spp.*

Presentation

10 Freeze dried vials
Vial
with: 3 ± 0.1 g

Packaging Details

23x60 mm glass vials, tag labelled, White plastic cap - 10 vials per box.

Shelf Life

49 months

Storage

2-25 °C

Composition

Composition (g/vial)

Polymyxin B.....0.0050
Acriflavine.....0.0025
Ceftazidime.....0.0100

NOTE : Each vial is sufficient to supplement 500 ml of PALCAM medium Base.

Reconstitute the original freeze-dried vial
by adding
Sterile Distilled Water..... 6 ml

Description /Technique

Description:

Listeria PALCAM selective supplement is added to PALCAM Medium base in order to obtain a complete selective medium used for the detection and the isolation of *Listeria monocytogenes* from foods.

Palcam Agar is based on the formulation described initially by van Netten *et al.* which has a high selectivity and produces good colonial differentiation. Selectivity is achieved by the inclusion of lithium chloride, acriflavine, polymyxin B and ceftazidime, since they inhibit the growth of almost all the Gram negative and most of the Gram positive companion bacteria.

Listeria hydrolyze esculin to esculetin, which reacts with ferric ammonium citrate producing a dark precipitate and green-grey colonies with beige halos. If colonies of enterococci or staphylococci do grow on this medium they can be easily recognized, since they utilise mannitol and produce yellow colonies and haloes, contrasting with the cherry-red colour of medium. However, when there are many *Listeria* colonies, the entire medium darkens, which can cause interference in differentiation. In these cases it is advisable to perform the inoculation with a more diluted sample.

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 the 6 ml sterile distilled water in aseptic conditions and add it to 500 ml of sterilized PALCAM agar base cooled to 50°C. Do not overheat once supplemented.

Pour the complete medium into Petri dishes and, once solidified on a flat surface, spread the plates by streaking methodology or by spiral method.

Incubate the plates in aerobic atmosphere at 37 ± 1°C for 44 ± 4h.

(Incubation times longer 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 agar, observing any blackening of the medium due to esculin hydrolysis, typical for *Listeria* strains.

Presumptive isolation of *Listeria sp.* must be confirmed by further microbiological and biochemical tests.

Quality control

Physical/Chemical control

Color: Orange

pH: at 25°C

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.

Isolation by loop spreading

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

Aerobiosis. Incubation at 37 °C ± 1, reading after 44 ± 4h

Microbiological control according to the current version of the ISO 11133:2014/A1:2018.

Microorganism

L. monocytogenes ATCC® 13932, WDCM 00021*Escherichia coli* ATCC® 25922, WDCM 00013*Enterococcus faecalis* ATCC® 29212, WDCM 00087*L. monocytogenes* ATCC® 7644

Sterility Control

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

Check at 7 days after incubation in same conditions.

Add 5mL of the sample to 100 mL of TSB and to 100 mL Thioglycollate.

Growth

Good - Esculin Positive reaction

Inhibited

Inhibited

Good - Esculin Positive reaction

Bibliography

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