

## Specification

Selective supplement for isolation and confirmation of *Listeria monocytogenes* formulated according to ISO 11290-1 and 2:1996 Amd 2004

## Presentation

10 Freeze dried vials  
Vial  
with: 3 ± 0.1 g

### Packaging Details

22±0.25 x 55±0.5 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.....38350 IU  
Cycloheximide.....0.025  
Ceftazidime.....0.010  
Nalidixic acid.....0.010

**NOTE :** Each vial is sufficient to supplement

476 ml of Listeria selective Agar Base according to Ottaviani and Agosti

Reconstitute the original  
freeze-dried vial

by adding 1 vial with

Sterile distilled water..... 6 ml

## Description /Technique

### Description:

Completed with all its supplements the Agar Listeria Ottaviani & Agosti is a selective and differential medium for the detection of *Listeria* species and the presumptive identification of *Listeria monocytogenes*.

The selectivity is achieved by the high concentration of lithium chloride and the mixture of antimicrobics. The differential activity is due to the chromogenic substrate to detect the  $\beta$ -glucosidase, enzyme that is present in all *Listeria* species.

The specific identification is obtained by the L- $\alpha$ -phosphatidylinositol, that acts as substrate for a phospholipase C present only in *Listeria monocytogenes* and some strains of *Listeria ivanovii*. The combination of both substrates allows the differentiation *L. monocytogenes*, which produces colonies blue-green in colour and surrounded by an opaque zone, from the other *Listeria* species, which growth with blue-green colonies but without any halo. This differentiation is evident after incubate the plates for 24 ± 2 hours at 37 °C.

Sometimes, especially with highly contaminated samples, it is possible that some colonies, white in colour, that are not *Listeria* growth. In this case it is recommended an enrichment step previous to the plate inoculation.

Observations: Most *Listeria ivanovii* also produce an opaque halo around the colonies after 48 h of incubation. This presumptive evidence must be confirmed by performing the biochemical or serological identification tests (Rhamnose / Xylose sugar fermentation, hemolysis tests, CAMP test, etc.) or any test confirming the species without hesitation.

### Technique:

Add 1 bottle Enrichment supplement Ottaviani & Agosti (L- $\alpha$ -phosphatidylinositol - 24ml) and 1 vial Selective supplement Ottaviani & Agosti for complete 500ml medium. Homogenize by mixing and distribute in Petri dishes. The solidified cool medium appears homogeneously turbid.

There are a lot of standardized methodology (ISO, FDA-BAM, AOAC, AFNOR, etc.) The technician must follow the protocol validated in his laboratory.

## Quality control

### Physical/Chemical control

Color : White-Gray

pH: at 25°C

### Microbiological control

Spiral Spreading: Practical range 100 ± 20 CFU. min. 50 CFU (productivity) / 10<sup>4</sup>-10<sup>6</sup> CFU (selectivity).

Microbiological control according to ISO 11133:2014/A1:2018.

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

### Microorganism

*Escherichia coli* ATCC® 25922, WDCM 00013*Enterococcus faecalis* ATCC® 29212, WDCM 00087*L. monocytogenes* ATCC® 13932, WDCM 00021*Listeria innocua* ATCC® 33090, WDCM 00017*Listeria monocytogenes* ATCC® 35152

### Growth

Inhibited

Inhibited

Good - Blue colonies with white halo

Blue colonies without white halo

Good - Blue colonies with white halo

### 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

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