

# Listeria Agar Base Palcam ISO

Cat. 1141

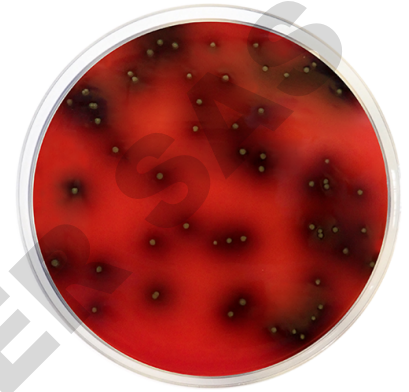
Selective and differential medium for the detection of *Listeria* spp, particularly *Listeria monocytogenes*.

## Practical information

Applications	Categories
Selective isolation	Listeria

Industry: Clinical / Food

Regulations: ISO 11290



## Principles and uses

Listeria Agar Base Palcam, used with supplements, is a selective and differential medium for *Listeria* spp. It is recommended by ISO 11290 for the detection and enumeration of *Listeria monocytogenes* in food products and clinical samples, and can also be used for environmental samples.

It is used after a primary and secondary enrichment stage, using Listeria Enrichment Broth Base (Cat.1120). It allows the easy differential diagnosis of *Listeria monocytogenes* using a double-system indicator: Esculin/Iron and Mannitol/Phenol red. All *Listeria* species hydrolyze the esculin to esculetin, which reacts with iron ions producing a blackening of the medium.

Lithium chloride included in the medium, along with ceftazidime, polymyxin B sulfate and Acryflavine from the supplement, inhibit the growth of the non-*Listeria* accompanying bacteria present in foods, which can hydrolyze the esculin. Peptones and maize starch provide a rich nutrient base for growth. Yeast extract is the source of vitamins, particularly of the B-group. Glucose is the fermentable carbohydrate. Ferric ammonium citrate improves the growth of *L. monocytogenes*.

The Mannitol/Phenol red differentiation system is used to differentiate *Listeria* spp that do not ferment mannitol from other species that occasionally grow in the medium such as enterococci or staphylococci. Differentiation is achieved by the acid increase in the media, causing the phenol red indicator to change the color of the medium from red to yellow. Confirmation of *Listeria* is done by biochemical and serological identifications tests.

## Formula in g/L

Glucose	0,5	Bacteriological agar	10
Esculin	0,8	Ferric ammonium citrate	0,5
Maize starch	1	Mannitol	10
Peptone	23	Phenol red	0,08
Sodium chloride	5	Yeast extract	3
Lithium chloride	15		

Typical formula g/L \* Adjusted and/or supplemented as required to meet performance criteria.

## Preparation

Suspend 34,4 grams of the medium in 500 ml of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Sterilize in autoclave at 121 °C for 15 minutes. Cool to 45-50 °C and aseptically add one vial of Palcam Listeria Selective Supplement (Cat. 6004). Homogenize gently and dispense into Petri dishes.

## Instructions for use

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

- Inoculate on the surface making parallel striae with the handle or swab.

- Incubate in aerobic conditions at 35±2 °C for 24-48 hours.
- Reading and interpretation of the results.

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

Primary enrichment:

- Weigh 25 g (or 25 ml) of the sample and add 225 ml of *Listeria* 1/2 Fraser Broth (Cat. 1120 + Cat. 6002). Homogenize and incubate at 30 °C for 25±1 h.

Secondary enrichment:

- Inoculate 0,1 ml of the culture of the *Listeria* 1/2 Fraser Broth incubated (regardless of its color) in 10 ml of *Listeria* Fraser Broth (Cat. 1120 + Cat. 6001).

Incubate at 37 °C for 24±2 hours under aerobic conditions.

Plaque and identification:

- From the primary enrichment culture, the *Listeria* Agar surface is inoculated according to Ottaviani and Agosti (Cat. 1345), to obtain well separated colonies.
- From the secondary enrichment culture, the procedure is repeated, inoculate the surface of the *Listeria* Agar according to Ottaviani and Agosti, the Palcam *Listeria* Agar (Cat. 1141) and another medium such as the Oxford Agar (Cat. 1133).
- For *Listeria* Agar according to Ottaviani and Agosti incubate for a total of 48±2 h.
- For Agar *Listeria* Palcam incubate at 35±2 °C for 24-48 h.
- For Oxford agar incubate at 35±2 °C for 24-48 h.

Confirmation:

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

## Quality control

Solubility	Appearance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25°C)
w/o rests	Fine powder	Beige	Red	7,2±0,2

## Microbiological test

According to ISO 11133:

Incubation conditions: (35±2 °C / 24-48 h).

Inoculation conditions: Productivity quantitative (100±20. Min. 50 CFU) / Selectivity (10<sup>4</sup>-10<sup>6</sup> CFU) / Specificity (10<sup>3</sup>-10<sup>4</sup> CFU).

Reference media: TSA.

Microorganisms	Specification	Characteristic reaction
<i>Listeria monocytogenes</i> 4b ATCC 13932	Good growth (2)	Green-gray colonies with black center and black halo.
<i>Escherichia coli</i> ATCC 25922	Total inhibition (0)	
<i>Enterococcus faecalis</i> ATCC 29212	Total inhibition (0)	
<i>Listeria monocytogenes</i> ATCC 7644	Good growth (2)	Green-gray colonies with black center and black halo

## Storage

Temp. Min.: 2 °C

Temp. Max.: 25 °C

## Bibliography

ISO NORMATIVE 11290-2: Microbiology of food and animal feeding stuffs - Horizontal method for the detection and enumeration of *Listeria monocytogenes* -- Part 2: Enumeration method.

Van Netten, P., I. Perales A. Van de Moosalijk G.D.W. Curtis and DAA Mossel 1989 Liquid and solid selective differential media for the detection and enumeration of *L. monocytogenes* and other *Listeria* spp. Int. J. of Food Microbiol 8: 299-317.

Farber JMDW Warburton and T. Babiuk, 1994 Isolation of *Listeria monocytogenes* from all food and environmental samples.