

CONFIRM' L. MONO AGAR®

CONFIRMATION OF *LISTERIA MONOCYTOGENES*

1 INTENDED USE

CONFIRM' L. mono Agar® is a solid culture media designed for the confirmation of the genus & species *Listeria monocytogenes*, isolated from a single characteristic colony from **COMPASS® Listeria Agar**, within the context of the detection method (certified AFNOR Certification, under the reference number BKR 23/2-11/02 and enumeration methods (certified AFAQ AFNOR Certification under the reference number BKR 23/05-12/07).



BKR 23/02-11/02, BKR 23/05-12/07
METHODES ALTERNATIVES D'ANALYSE POUR L'AGROALIMENTAIRE
Certifié par AFNOR Certification <http://nf-validation.afnor.org>

2 HISTORY

Standardized confirmation methods (hemolysis detection, sugar utilization and the CAMP test) are rather fastidious to perform and are poorly adapted to newer chromogenic media, which offer greater specificity and selectivity for *Listeria monocytogenes*.

In 1977, Groves et Welshimer demonstrated the correlation between pathogenicity and the acidification of rhamnose in bacteria belonging to the genus *Listeria*.

14 years later, Notermans et al (1991) proved that the enzymatic activity of specific phospholipase C phosphatidyl-inositol (PI-PLC) was one of the fundamental markers of pathogenicity for *Listeria monocytogenes*.

The principle of CONFIRM' L. mono Agar® is based on the expression of these two elements, when combined are specific to *Listeria monocytogenes* taken from a single colony isolated on **COMPASS® Listeria Agar**.

3 PRINCIPLES

The unique composition of the media in peptones and growth factors insure proliferation of strains belonging to the genus *Listeria*, isolated from colonies taken from **COMPASS® Listeria Agar** (refer to the technical data sheet for this product).

A judiciously developed inhibition system provides the selectivity needed to prevent proliferation of secondary microflora, which could be isolated along with the characteristic colonies from **COMPASS® Listeria Agar**.

The detection of rhamnose fermentation is visualized by a color change to yellow, the result of a localized drop in pH.

PI-PLC detection is observed through the development of an opaque halo around the inoculation streak.

4 TYPICAL COMPOSITION

The composition can be adjusted in order to obtain optimal performance.

For 1 liter of media :

- Special peptone mixture.....	18,800 g
- Growth promoters	8,600 g
- Selective agents	9,700 g
- Rhamnose	7,330 g
- Colored indicator	0,097 g
- Phospholipids	1,450 g
- Bacteriological agar	14,500 g

pH of the ready-to-use media at 25°C : $7,3 \pm 0,2$.

5 INSTRUCTIONS FOR USE

- Dry the plates in an incubator (BM139), covers partially removed.
- From a single characteristic colony isolated on COMPASS® Listeria Agar inoculate a single streak on the surface of CONFIRM' L. mono Agar® (up to 6 radial streaks per plate).
- Incubate at 37 ± 1 °C for 24 ± 3 hours.

6 RESULTS

The presence of a characteristic colony is characterized by the growth on the agar surface, with a yellow discoloration and an opaque halo.

The aspect of the isolation streaks is as follows, following incubation at 37 ± 1 °C for 24 ± 3 hours :

Microorganisms	Growth	Yellow discoloration (Rhamnose fermentation)	Opaque halo (Phospholipid hydrolysis)
<i>Listeria monocytogenes</i>	good	positive	positive
<i>Listeria ivanovii</i>	partially inhibited ⁽¹⁾	negative	variable
<i>Listeria innocua</i> , <i>Listeria murrayi</i> , <i>Listeria welshimeri</i> , <i>Listeria seeligeri</i>	good	variable	negative
<i>Listeria grayi</i>	good	negative	negative
<i>Bacillus cereus</i>	partially inhibited ⁽¹⁾	negative	variable
<i>Enterococcus faecalis</i> <i>Staphylococcus aureus</i>	totally inhibited	-	-
<i>Escherichia coli</i> <i>Pseudomonas</i>	totally inhibited	-	-

⁽¹⁾ : Strains of *Listeria ivanovii* and *Bacillus cereus* are partially inhibited on **CONFIRM' L. mono Agar®**, which can be manifested in an absence of growth when struck on the plate. However, in a certain number of cases, in spite of the absence of growth at the point of inoculation, an opaque halo can appear due to the presence of the (still) active enzymes present on the cells placed on the surface of the agar.

See ANNEX 1 : PHOTO SUPPORT

7 QUALITY CONTROL

Prepared media : red, opalescent agar

Typical cultural response after (24 ± 3) hours incubation at (37 ± 1) °C:

Microorganisms		Growth	Streak characteristics	
			Discoloration (yellow)	Opaque halo
<i>Listeria monocytogenes</i>	WDCM 00109	good, score 2	positive	presence
<i>Listeria monocytogenes</i>	WDCM 00021	good, score 2	positive	presence
<i>Listeria ivanovii</i>	WDCM 00018	slowed, score 0-1	negative	presence
<i>Listeria innocua</i>	WDCM 00017	good, score 2	positive	absence
<i>Staphylococcus aureus</i>	WDCM 00034	inhibited, score 0	-	-
<i>Escherichia coli</i>	WDCM 00013	inhibited, score 0	-	-

8 STORAGE / SHELF LIFE

Pre-poured media in Petri plates : 2-8 °C.

The expiration date is indicated on the label.

9 PACKAGING

Pre-poured media in Petri plates (Ø 90 mm) :

10 plates BM13908

10 BIBLIOGRAPHY

GROVES, R. D., and WELSHIMER, H. J.. 1977. Separation of pathogenic from apathogenic *Listeria monocytogenes* by three in vitro reactions. *Journal of Clinical Microbiology*, **5**(6), 559-563.

NOTERMANS, S.H., DUFRENNE, J., LEIMESTER-WÄCHTER, M., DOMANN, E., and CHAKRABORTY, T.. 1991. Phosphatidylinositol-specific phospholipase C activity as a marker to distinguish between pathogenic and nonpathogenic *Listeria* species. *Applied and Environmental Microbiology*, **57**(9), 2666-2670.

11 ADDITIONAL INFORMATION

CONFIRM' L. mono Agar[®], **CONFIRM' L. mono broth[®]** & **COMPASS[®]** are trademarks of SOLABIA S.A.S.

The information provided on the labels take precedence over the formulations or instructions described in this document and are susceptible to modification at any time, without warning.

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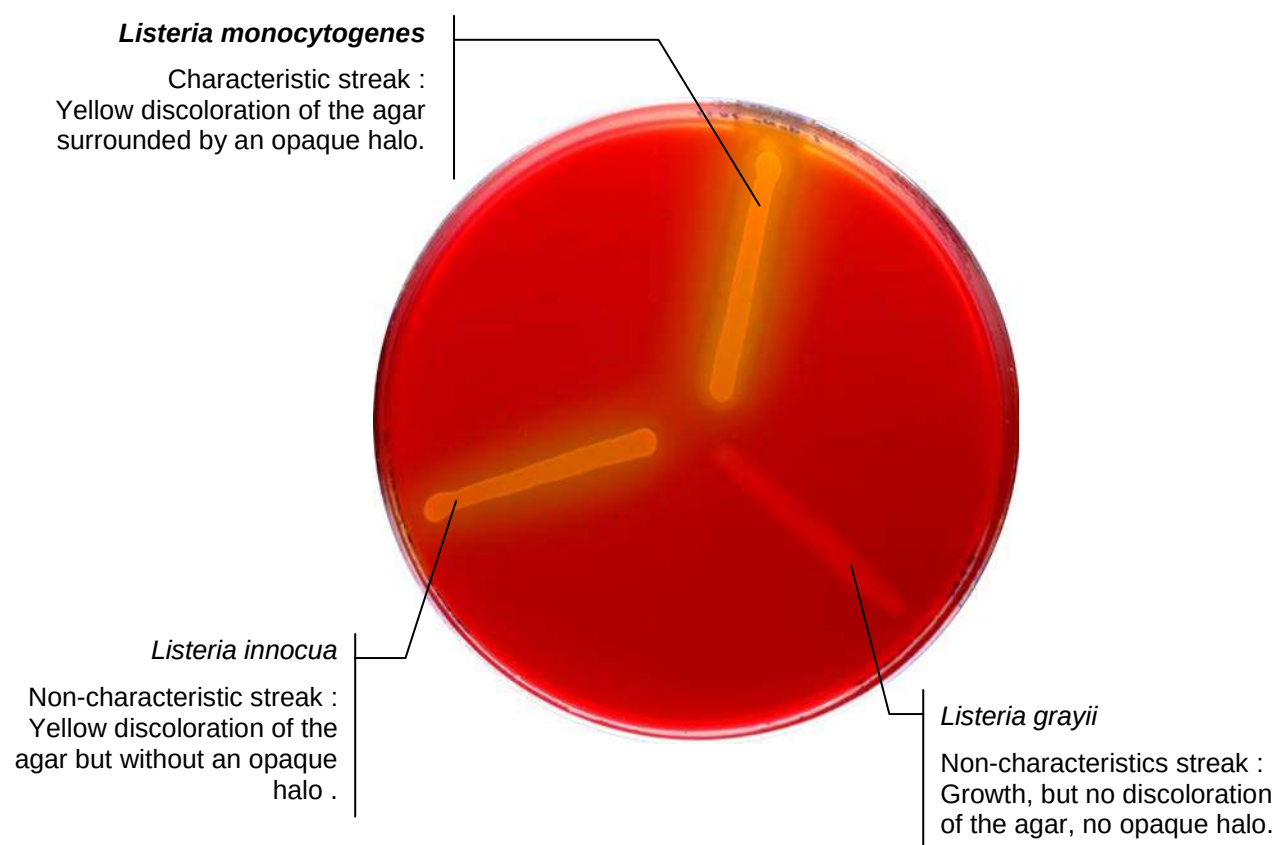
ANNEX 1 : PHOTO SUPPORT

CONFIRM' *L. mono* Agar®

Confirmation of the genus / species *Listeria monocytogenes*, from a single characteristic colony taken from COMPASS® *Listeria* Agar.

Methodology :

Incubation 24 hours at 37°C (up to 6 radial surface streaks per plate)



Product code :

BM13908 : Pre-poured media in Petri plates (Ø 90 mm) - 10 plates