

CHROMagar™ Listeria

Chromogenic medium for detection, isolation and enumeration of *L.monocytogenes*

APPLICATIONS:

• CHROMagar Listeria is suitable for use in food industry:

CHROMagar Listeria method (presence / absence):

→ Certified AFNOR VALIDATION CHR-21/1-12/01.

Valid until: 13 Dec 2013

CHROMagar Listeria enumeration method:

→ Certified AFNOR VALIDATION CHR-21/2-12/06.

Valid until: 13 Dec 2013

For use with all human food products and environmental samples.

• CHROMagar Listeria is also suitable for clinical field, allowing isolation of *L.monocytogenes* in samples of human origin eg. blood cultures, vaginal swabs, or CSF.

STORAGE Store the powder at 2/30°C and the supplement at 2/8°C until the shelflife date indicated on the label.

COMPOSITION in g/L Agar 15.0; Peptone and yeast extract 23.0; NaCl 5.0; Chromogenic mix 17.5; pH : 7.0 +/- 0.5 (Classical formula adjusted and/or supplemented as required to meet performance criteria).

PREPARATION BASE : For pre-weighed dose of medium, add CHROMagar Listeria base dry powder to the corresponding volume of purified water. Alternatively, add 51,5 g/L of the base agar to the appropriate volume of purified water. Disperse powder slowly in water by rotating for swelling of the agar. Heat to 121°C +/- 1°C for 15 minutes. Cool in a water bath to 47°C +/- 2°C. **SUPPLEMENT** : Disperse slowly CHROMagar Listeria supplement in the corresponding volume of STERILE purified water, 10ml for a 250ml dose or in 40ml per 9g per final litre. Add a magnetic bar and stir at high speed (700-1000 rpm) at least 30 minutes WITHOUT HEATING to give a creamy homogeneous suspension.

MIXING OF BASE AND SUPPLEMENT : Place the melted 47°C cooled CHROMagar Listeria base under gentle stirring. Add the homogeneous reconstituted supplement, keeping the gentle stirring during 1 or 2 minutes until complete homogenisation. Pour IMMEDIATELY into sterile Petri dishes. DO NOT STACK THE PETRI DISHES and let them cool and dry. Store in the dark. Prepared plates can be kept for one day at ambient temperature, or for up to two weeks at (2/8°C) if properly prepared and protected from light and dehydration.

INTERPRETATION

L.monocytogenes → blue, diameter less than 3mm, regular and white halo

Other microorganisms → blue, colourless, other colour, inhibited

LIMITATIONS Some strains of *L.ivanovii* may also give blue colonies with white halo are distinguished during identification. The species *L.ivanovii* is rarely found in food. Some strains of *B.cereus* can also grow as blue colonies. They can easily be distinguished from colonies of *L.monocytogenes* as they are much larger with an irregular edge to the colony and very large white halo.

DISPOSAL OF WASTE After interpretation all plates should be destroyed by autoclaving at 121°C for at least 20 minutes.

PROCEDURES of CHROMagar LISTERIA METHODS

➔ **Detection of Presence / Absence of *L.monocytogenes* in all human food products and environmental samples. (Refer to shema 1)**

USE CHROMagar Listeria Method

xg or xml in 9x Fraser 1/2 broth

→ 24h +/- 2h at 30°C +/- 1°C

spread 0,1ml on a well dried CHROMagar Listeria plate, by streaking on 1/3 of the plate until liquid is adsorbed. Isolate on the rest of the plate and incubate.

→ 24h +/- 2h at 37°C +/- 1°C

Typical colonies: **NO** → Absence of *L.monocytogenes*

YES → In the context of AFNOR Validation, all samples identified as positive by the alternative method must be confirmed in one of the following ways:

1/ using the conventional tests described in the standardized methods by CEN or ISO (including the purification step).

2/ with the CHROMagar Identification Listeria directly from the suspect colony without step purification.

In the event of discordant results (positive by the alternative method, non-confirmed by one of the means described above) the laboratory must follow the necessary steps to ensure validity of the result obtained.

Test portions weighing more than 25 g have not been tested. For environmental samples, should typical colonies be absent after 24h, incubate for additional 24h +/- 2h.

➔ **Enumeration of *L.monocytogenes* in al human food products and environmental samples. (Refer to shema 2)**

USE CHROMagar Listeria enumeration Method

xg or xml in 9x BPW or non selective half Fraser broth

→ 1h +/- 5 min at 20°C +/- 1°C

Spread 0,1 ml on one well dried CHROMagar Listeria plate (in case of estimation of small numbers spread 1ml on three plates) and incubate.

→ 48h +/- 2h at 37°C +/- 1°C

A first reading at 24h allows a more rapid detection of heavily contaminated samples. However the final result count is reached after 48 h +/- 2h. Refer to ISO 7218 standard for calculation and interpretation of results.

Typical colonies: **NO** → Absence of *L.monocytogenes*

YES → Enumerate and confirm a colony directly from CHROMagar Listeria. In the context of AFNOR Validation: If confirmation has already been done during the research phase a new confirmation is not necessary. Otherwise, all samples identified as positive by the alternative method must be confirmed in one of the following ways:

1/ using the conventional tests described in the standardized methods by CEN or ISO (including the purification step).

2/ with the CHROMagar Identification Listeria directly from the suspect colony without step purification.

In the event of discordant results (positive by the alternative method, non-confirmed by one of the means described above) the laboratory must follow the necessary steps to ensure validity of the result obtained.

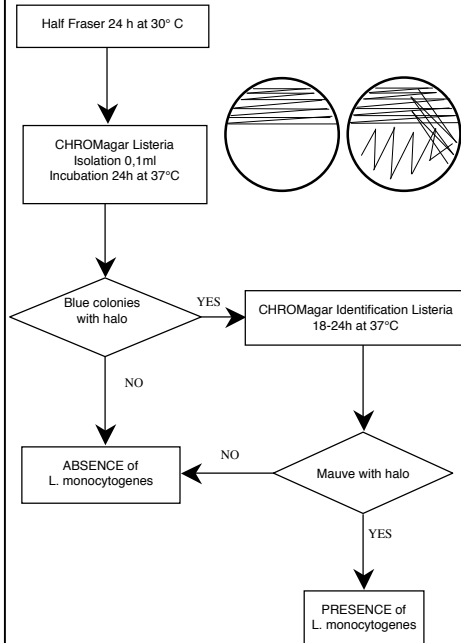
English. For *in vitro* diagnostic use. Laboratory product to be used only by trained personnel, in compliance with good laboratory practice.

NT-EXT-045

Version 2 – June 2010

Shema 1
Detection of Presence/Absence
of *L.monocytogenes*
in foodstuffs and environmental
samples.

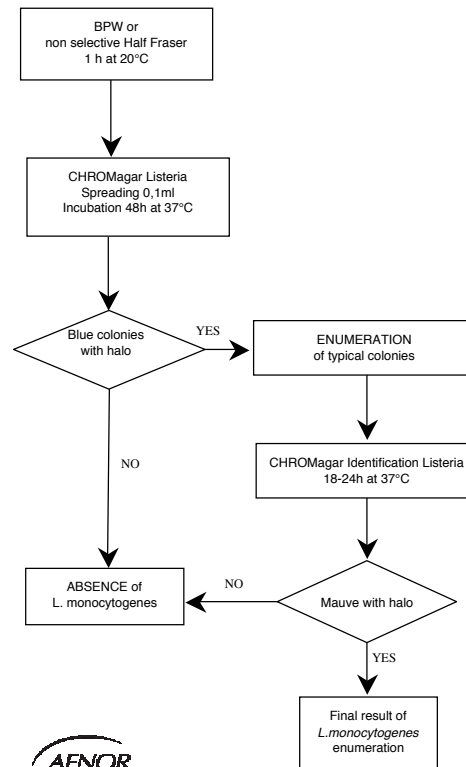
CHROMagar Listeria Method



N°CHR-21/1-12/01 : méthodes alternatives d'analyse pour
l'agro-alimentaire - certifié par AFNOR Certification
www.afnor-validation.org

Shema 2
Enumeration of *L.monocytogenes*
in foodstuffs and environmental samples.

CHROMagar Listeria enumeration Method



N° CHR-21/2-12/06: méthodes alternatives d'analyse pour
l'agro-alimentaire - certifié par AFNOR Certification
www.afnor-validation.org

REFERENCES

Ref. LM851 : 1000ml
Ref. LM852 : 5000ml bottle
Ref. LM853 : bulk

BIBLIOGRAPHY

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Available from CHROMagar :

CHROMagar™ Candida
Differentiation of major pathogenic *Candida* species

CHROMagar™ Orientation
Differentiation of urinary tract pathogens

Rambach™ Agar
Detection of *Salmonella* spp

CHROMagar™ Salmonella
Detection of *Salmonella* including *S. Typhi*

CHROMagar™ Salmonella Plus
Detection of *Salmonella* according to the ISO 6579:2002 norm

CHROMagar™ O157
Detection of *E.coli* O157

CHROMagar™ E.coli
Detection and enumeration of *E.coli*

CHROMagar™ ECC
Detection and enumeration of *E.coli* and coliforms

CHROMagar™ Liquid ECC
Broth for pad technique for *E.coli*-coliforms

CHROMagar™ Staph aureus
Detection and enumeration of *Staphylococcus aureus*

CHROMagar™ MRSA
Detection of MRSA including low level MRSA

CHROMagar™ Listeria
Detection and enumeration of *Listeria monocytogenes*

CHROMagar™ Vibrio
Detection and enumeration of *Vibrio parahaemolyticus*, *Vibrio vulnificus* and *Vibrio cholerae*

CHROMagar™ VRE
Detection of *E.faecium* VRE & *E.faecalis* VRE

CHROMagar™ StrepB
Detection of *Streptococcus agalactiae* (GBS)

CHROMagar™ ESBL
Detection of ESBL-producing bacteria

CHROMagar™ KPC
Detection of Carbapenem-resistant bacteria

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