

Harlequin® *Listeria* Chromogenic Agar (Ottaviani & Agosti) (NCM1004)

NCM1004 500G, 5KG & 10KG DCM Packs
NCM3000 90mm Pre-Poured Plates (fully supplemented)*

* Shipping restrictions may apply, enquire for regional availability

Intended Use

Listeria Chromogenic Agar (according to the formulation of Ottaviani and Agosti) is a selective medium for the isolation and presumptive identification of *Listeria monocytogenes* from foodstuffs and related materials as described in ISO 11290-1:2017. This medium is not intended for use in the diagnosis of disease or other conditions in humans.

Description

Lithium chloride in the base medium and supplementary antimicrobial compounds Ceftazidime, Polymyxin, Nalidixic acid and Cycloheximide provide the medium's selectivity. Chromogenic activity is as a result of a chromogenic substrate for the detection of the β -glucosidase enzyme, common to all *Listeria* spp. and to a few strains of Enterococci and Bacilli.

The specific differential activity of this agar is obtained with a proprietary lecithin substrate for the detection of the phospholipase enzyme present in the *L. monocytogenes* colonies growing on this media. This enzyme activity will result in a halo of precipitation surrounding the target colonies.

With the combination of both the chromogenic and phospholipase enzyme reactions, it is possible to differentiate *Listeria monocytogenes* (blue colonies surrounded by an opaque halo) from other *Listeria* spp. (blue colonies without an opaque halo).

Typical Formulation

Enzymatic Digest of Animal Tissue	18.0 g/L
Enzymatic Digest of Casein	6.0 g/L
Yeast Extract	10.0 g/L
Lithium Chloride	10.0 g/L
Sodium Chloride	5.0 g/L
Disodium Hydrogen Phosphate (anhydrous)	2.5 g/L
Sodium Pyruvate	2.0 g/L
Glucose	2.0 g/L
Magnesium Glycerophosphate	1.0 g/L
Magnesium Sulfate (anhydrous)	0.5 g/L
5-Bromo-4-Chloro-3-Indolyl- β -D-Glucopyranoside	0.05 g/L
Agar	12.5 g/L

Final pH: 7.2 \pm 0.2 at 25°C

Formula may be adjusted and/or supplemented as required to meet performance specifications.

Supplement

NCM4002 *Listeria* Chromogenic Selective Supplement
NCM4001 *Listeria* Chromogenic Diagnostic Supplement

Precaution

Refer to SDS



Technical Specification Sheet



Preparation for DCM

1. Suspend 69.5 grams of the medium in 950mL of purified water.
2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
3. Autoclave at 121°C for 15 minutes.
4. Cool to 45-50°C and add 2 vials of reconstituted NCM4002-0.5* supplement, each reconstituted using 5mL of sterile deionized/RO water. Swirl to mix.
5. Add 2 vials of NCM4001-0.5* supplement (**pre- heated to 48-50°C**). Mix well with gentle end-over-end mixing.

*Larger vials may be available. Please see appropriate supplement data sheet for availability and preparation instructions.

Test Procedure

- For the detection of *Listeria monocytogenes* and *Listeria* spp – Refer to ISO 11290-1:2017
- For the enumeration of *Listeria monocytogenes* and *Listeria* spp – Refer to ISO 11290-2:2017

Quality Control Specifications

Dehydrated Appearance: Powder is homogeneous, free flowing and beige.

Prepared Appearance: Prepared medium is an opaque, cream-yellow gel.

Expected Cultural Response: Cultural response on Listeria Chromogenic Agar (supplemented with NCM4001 and NCM4002), incubated aerobically 37 ± 1°C and examined for growth after 44 - 52 hours incubation.

MICROORGANISM	WDCM	EXPECTED RESULTS
<i>Listeria monocytogenes</i>	00021	Good growth, blue / green colonies with an opaque halo
<i>Listeria monocytogenes</i>	00109	
<i>Escherichia coli</i>	00012 or 00013	Total inhibition
<i>Enterococcus faecalis</i>	00009 or 00087	
<i>Listeria innocua</i>	00017	Blue / green colonies without an opaque halo

Expiration

Refer to expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing or appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitations of the Procedures

- Due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.
- Isolates presumptively identified as *Listeria* spp. and *Listeria monocytogenes* must be subjected to further biochemical tests to confirm their identity. Some strains of *Listeria ivanovii* may demonstrate lecithinase activity.



Technical Specification Sheet



Storage

Store dehydrated culture media (NCM1004) at 2-30 °C away from direct sunlight. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping container tightly closed.

Store pre-poured plates (NCM3000) at 2-8 °C away from direct sunlight.

References

1. ISO 11290-1:2017 Microbiology of the food chain- Horizontal method for the detection and enumeration of *Listeria monocytogenes* and *Listeria* spp.- Part 1: Detection method
2. ISO 11290-2:2017 Microbiology of the food chain- Horizontal method for the detection and enumeration of *Listeria monocytogenes* and *Listeria* spp.- Part 2: Enumeration method

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