CM 20455 - CHROMOGENIC L-MONOLISTERIA DIFFERENTIAL AGAR

INTENDED USE

For selective identification and differentiation of Listeria monocytogenes

PRODUCT SUMMARY AND EXPLANATION

Chromogenic L-Mono Listeria Differential Agarisbased on the formulation of Ottaviani and Agosti and is used for isolation and cultivation of Listeria monocytogene. It allowsselective identification and differentiation of Listeria monocytogenes.
Listeria monocytogenes is a gram-positive foodborne human pathogen responsible for serious infections in pregnant women that may ultimately result in abortion, stillbirth,birth of a child with neonatal listeriosis and meningitis or primary bacteremia in adults and juveniles. The pathogenicity of Listeriaivanovii for humans is uncertain.
This medium is based on the specific chromogenicdetection of ß-glucosidase activity, rhamnose fermentation and PIPLC activity. Listeria species hydrolyse the purified chromogenic substrate in the medium giving blue coloured colonies. Since ß-glucosidase activity is specific for Listeria species, other organisms cannot utilize the chromogenic substrate and therefore give white colonies. Differentiation between Listeria species is based on the property of rhamnose fermentation and PIPLC activity. The colonies of L.monocytogenes appear bluish green with a yellow halo (rhamnose positive) while the colonies of L.ivanovii appear bluish green without a yellow halo (Rhamnose negative).The differentiation of L.monocytogenes and L.innocua is based on PIPLC (phosphatidylinositol-specific phospholipase C) activity. Phospholipase C enzyme hydrolyses the purified substrate (TS 031) added to the medium resulting in an opaque halo around Listeria monocytogenes colonies. L.ivanovii also demonstrates PIPLC activity however since it does not ferment rhamnose it can be easily distinguished from L.monocytogenes.

COMPOSITION

Ingredients	Gms / Ltr
Peptone special	23.000
Agar	15.000
Tryptone	10.000
Rhamnose	10.000
Lithium chloride	5.000
Sodium chloride	4.000
Soya peptone	2.000
Chromogenic mixture	1.160
Phenol red	0.120

PRINCIPLE

Medium contains Peptone special,tryptone and soya peptone which provide nitrogenous substances, vitamin B complex and other essential growth nutrients. Rhamnose is the fermentable carbohydrate with phenol red as an indicator. Sodium chloride maintains the osmotic equilibrium.The added lithium chloride and Chromogenic Listeria Selective Supplement (TS 205) inhibit growth of most gram-positive bacteria, gram-negative bacteria, yeasts and moulds. Phospholipase C enzyme hydrolyses the purified substrate (TS 031) addedto the medium resulting in an opaque halo around Listeria monocytogenes colonies demonstrating PIPLC activity.

INSTRUCTION FOR USE

- Dissolve 70.28gramsin 940ml distilled water.
- Gently heat with swirling to dissolve the medium completely.
- Sterilize by autoclaving at 15 psi (121°C) for 15 minutes.

Cool to 45-50°C.
Aseptically add sterile contents of 2 vials of Chromogenic Enrichment Supplement (TS 031) and sterile rehydrated contents of 2 vial of Chromogenic Listeria Selective Supplement (TS 205).
Mix well and pour into sterile Petri plates.

Note: Lithium chloride is harmful. Avoid bodily contact and inhalation of vapours. On contact with skin wash with plenty of water immediately.

QUALITY CONTROLSPECIFICATIONS

Appearance of Powder	:	Light yellow to pink colour, homogeneous free flowing powder
Appearance of prepared medium	:	Red colour, opalescent gel
pH (at 25°C)	:	7.4 ± 0.2

INTERPRETATION

Culturalcharacteristics observed for incubation with addition of Chromogenic Listeria Selective Supplement (TS 205) and Chromogenic Enrichment Supplement (TS 031).

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Colour of colony	Recovery	Rhamnose Fermentation	PIPLC Activity	Incub.* Temp	Incub.* period
Listeria innocua	33090	50-100	Luxuriant	>= 50%	Bluish green	Positive Reaction (Yellow background)	Negative Reaction	35 ± 2°C	24 - 48 Hours
Listeria ivanovii	19119	50-100	Luxuriant	>= 50%	Bluish green	Negative Reaction	Positive Reaction#	35 ± 2°C	24 - 48 Hours
Listeria monocytogenes	19118	50-100	Luxuriant	>= 50%	Bluish green	Positive Reaction (Yellow background)	Positive Reaction#	35 ± 2°C	24 - 48 Hours
Bacillus subtilis	6633	≥ 1000	Inhibited	0%	-	-	-	35 ± 2°C	24 - 48 Hours
Candida albicans	10231	≥ 1000	Inhibited	0%	-	-	-	35 ± 2°C	24 - 48 Hours
Escherichia coli	25922	≥ 1000	Inhibited	0%	-	-	-	35 ± 2°C	24 - 48 Hours
Pseudomonas aeruginosa	27853	≥ 1000	Inhibited	0%	-	-	-	35 ± 2°C	24 - 48 Hours

#= Positive reaction, opaque halo around the colony exhibiting phosphatidyl inositol specific phospholipase activity.

PACKAGING

Inpacksizeof100gm bottles.

STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 2-8°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.
Product Deterioration: Do not use if powder show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

DISPOSAL

Afteruse,prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

REFERENCES

1. F. Ottaviani, M. Ottaviani, M. Agosti, Esperienzasuun agar selettivo e differenziale per Listeria monocytogenes, IndustrieAlimentari 36, 1-3 (1997).
2. F. Ottaviani, M. Ottaviani, M. Agosti, Differential agar medium for Listeria monocytogenes, Quinper Froid Symposium Proceedings, p.6, A.D.R.I.A. Quinper, 16-18. June (1997).
3. Schlech WF, Lavigne PM, Bortolussi RA, et al.(January 1983). "Epidemic listeriosis-evidence for transmission by food". N. Engl. J.Med. 308(4): 203–6. doi:10.1056.
4. Notermans S.H. and Dufrenne J., (1991), Applied and Environmental Microbiology, 57(09): 2666-70.
5. Mengaud J., Braun-Breton C. and Cossart P., (1991), Molecular Microbiology, 5(2): 367-372.

NOTE:PleaseconsulttheMaterialSafetyDataSheetforinformationregardinghazardsandsafehandlingPractices.

*For LabUse Only