Technical Information

L. mono Differential Agar Base

Product Code: DM 2540

Application: - L. mono Differential Agar Base has been recommended for the selective and differential isolation of *Listeria monocytogenes.*

Composition**

Ingredients	Gms / Litre
Meat peptone	18.000
Casein enzymic hydrolysate	6.000
Yeast extract	10.000
Sodium pyruvate	2.000
Glucose	2.000
Magnesium glycerophosphate	1.000
Magnesium sulphate	0.500
Sodium chloride	5.000
Lithium chloride	10.000
Disodium hydrogen phosphate anhydrous	2.500
Chromogenic substrate	0.050
Agar	15.000
Final pH (at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

L. mono Differential Agar Base is based on the formulation of Ottoviani and Agosti (1, 2) for the selective and differential isolation of *Listeria monocytogenes* from food and animal feeds which is adopted by ISO Committee (3).

Listeria monocytogenes is a gram-positive food borne 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 *Listeria ivanovii* for humans is uncertain. Since *L. monocytogenes* and *L. innocua* have similar biochemical properties, they cannot be differentiated on traditional media (PALCAM, Oxford).

Meat peptone, casein enzymic hydrolysate, yeast extract and sodium pyruvate supply essential growth nutrients and nitrogenous substances. Glucose is the fermentable carbohydrate. Sodium chloride maintains osmotic equilibrium. Phosphate buffers the medium. Lithium chloride and added selective supplements (MS 2212 and MS 2213) inhibit other microflora and allow the growth of *Listeria* species. *Listeria* species hydrolyse the chromogenic substrate which produces green coloured colonies. Differentiation of *Listeria monocytogenes* from other *Listeria* species is based on phosphatidylinositol- specific phospholipase C (PIPLC) activity. Phospholipase C enzyme hydrolyses the purified substrate (MS 2214) added to the medium resulting in an opaque halo around *Listeria monocytogenes* colonies.

Methodology

Suspend 36.02 grams of dehydrated powder media in 460 ml distilled water. Mix thoroughly & heat to boil to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C. Aseptically add sterile contents of 1 vial of L. mono Enrichment Supplement I (MS 2214) and sterile rehydrated contents of L .mono Selective Supplement I (MS 2212), L .mono Selective Supplement II (MS 2213). Mix well and pour into sterile Petri plates.

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

Quality Control

Appearance
Cream to yellow homogeneous free flowing powder

Gelling
Firm, comparable with 1.5% Agar gel

Colour and Clarity
Light amber coloured, opalescent gel forms in Petri plates

Reaction
Reaction of 7.2% w/v aqueous solution at 25°C. pH : 7.2±0.2

pH Range
7.00-7.40

Cultural Response
DM 2540: Cultural characteristics observed with added sterile L. mono Selective supplement I (MS 2212), L. mono Selective Supplement II (MS 2213) and L.mono Enrichment supplement I (MS 2214) after an incubation at 35 - 37°C for 24 - 48 hours.

Cultural Response

Organism	Inoculum (CFU)	Growth	Recovery	Colour of Colony	PIPLC activity
Cultural Response					
Candida albicans ATCC 10231	>=10^3	inhibited	0%	-	-
Enterococcus faecalis ATCC 29212	>=10^3	inhibited	0%	-	-
Escherichia coli ATCC 25922	>=10^3	inhibited	0%	-	-
Listeria innocua ATCC 33090	50-100	luxuriant	>=50%	greenish-blue	negative
Listeria grayi ATCC 19120	50-100	luxuriant	>=50%	greenish-blue	negative
Listeria ivanovii ATCC 19119	50-100	luxuriant	>=50%	greenish-blue	positive, opaque halo around the colony exhibiting phophatidylinosito specific phospholipase acivity
Listeria monocytogenes ATCC 19112	50-100	luxuriant	>=50%	greenish-blue	positive, opaque halo around the colony exhibiting phophatidylinosito specific phospholipase acivity
Listeria seeligeri ATCC 35967	50-100	luxuriant	>=50%	greenish-blue	negative
Listeria welshimeri ATCC 43549	50-100	luxuriant	>=50%	greenish-blue	negative
Pseudomonas aeruginosa ATCC 27853	>=10^3	inhibited	0%	-	-

Storage and Shelf Life

Dried Media: Store dehydrated powder and the prepared medium at 2-8 °C in tightly closed container. Use before expiry date on the label.
Prepared Media: 2-8 °C in sealable plastic bags for 2-5 days.

Further Reading

1. Ottaviani F., Ottaviani M., and Agosti M. (1997 a), Industrie Alimentari 36, 1-3.
2. Ottaviani F., Ottaviani M., and Agosti M. (1997 b), Quimper Froid Symposium Proceedings p. 6, A.D.R.I.A. Quimper, France, 16-18 June 1997.
3. Draft Amendment ISO 11290-2:1996/DAM 1.

Disclaimer :

- User must ensure suitability of the product(s) in their application prior to use.
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