# Listeria Selective Broth Base

**M889** 

Listeria Selective Broth Base with addition of selective supplement is recommended for selective isolation and cultivation of *Listeria monocytogenes* from clinical specimens.

#### Composition\*\*\*

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Ingredients	Gms / Litre
Casein enzymic hydrolysate	17.000
Papaic digest of soyabean meal	3.000
Yeast extract	6.000
Sodium chloride	5.000
Dipotassium hydrogen phosphate	2.500
Dextrose	2.500
Final pH ( at 25°C)	7.3±0.2

<sup>\*\*</sup>Formula adjusted, standardized to suit performance parameters

#### **Directions**

Suspend 36 in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to room temperature and aseptically add rehydrated contents of 1 vial of Listeria Selective Supplement II (FD063) or 2 vials of Listeria Selective Supplement II (FD063I) as desired. Mix well before dispensing.

# **Principle And Interpretation**

Listeria monocytogenes is a short, gram-positive, non spore-forming rod shaped bacterium that appears coccoidal in older cultures. Listeria multiplies over a wide range of temperatures from 3°C to 45°C with optimum temperature range of 30°C to 37°C. L. monocytogenes has been isolated from numerous environmental sources such as silage, soil, decaying vegetation, sewage, damp earth, straw and faeces (1, 2). Detection of L. monocytogenes in foods is not difficult. Low numbers of organisms are commonly isolated from raw milk, meat, vegetables, seafood and the food-processing environment. Enrichment procedures are used to isolate low numbers of L.monocytogenes. Injured L.monocytogenes are sublethally stressed as a result of exposure to heat, freezing or acidic conditions. Sublethally stressed L.monocytogenes require resuscitation in a non-selective medium at a temperature favouring repair of the sublethal injury.

Listeria Selective Broth is formulated as per Lovett et al (3) for the selective enrichment of *Listeria* species from milk and milk products and other foods. Listeria Selective Broth is recommended by ISO Committee (4) with a slight modification in the supplement (FD063I).

Casein enzymic hydrolysate, papaic digest of soyabean meal and yeast extract provide carbon and nitrogen compounds essential for bacterial metabolism. Dextrose is the energy source. The medium is rendered selective by addition of selective supplement. Cycloheximide inhibits the growth of saprophytic fungi. Nalidixic acid inhibits growth of gramnegative organisms and acriflavin suppresses gram-positive microorganisms (5, 6). Acriflavin is an acridinic derivative with bacteriostatic properties towards many gram-positive bacteria and a weak fungicidal activity.

For enrichment, 25 grams or 25 ml sample is added to 225 ml medium in a stomacher bag. Homogenize the material if required. Incubation is carried out at 30°C for upto 7 days. Ajello et al (7) showed that incubation period of 7 days allows better recovery of environmentally stressed *Listeria* from milk and milk products. The enrichment broth is further subcultured on Listeria Selective Agar (M567) after 1, 2 and 7 days.

*Listeria monocytogenes* is a highly pathogenic organism and therefore proper precautions should be taken while handling them.

## **Quality Control**

### Appearance

Cream to yellow homogeneous free flowing powder

### Colour and Clarity of prepared medium

Fluorescent yellow coloured, clear solution in tubes

#### Reaction

Reaction of 3.6% w/v aqueous solution at 25°C. pH: 7.3±0.2

#### **Cultural Response**

M889: Cultural characteristics observed with added Listeria Selective Supplement II (FD063/FD063I) after an incubation at 30-35°C for 24-48 hours.

Organism	Inoculum (CFU)	Growth		
Candida albicans ATCC 10231	>=103	inhibited		
Escherichia coli ATCC 25922	>=103	inhibited		
Listeria monocytogenes ATCC 19111	50-100	luxuriant		
Listeria monocytogenes ATCC 19112	50-100	luxuriant		
Listeria monocytogenes ATCC 19118	50-100	luxuriant		
Staphylococcus aureus ATCC 25923	50-100	none-poor		

#### Reference

- 1.Gray M. L., 1960, Science, 132:1767.
- 2. Weis J., and Seeliger H. P. R., 1975, Appl. Microbiol. 30:29.
- 3.Lovette J., Francis D. W. and Hunt J. M., 1987, J. Food Prot., 50:188.
- 4.International Organization for Standardization (ISO), 1993, 10560 Ind. Technical "Corrigendum Cor. 1:1994.
- $5. Lee\ W.\ K.\ and\ McClain\ D.,\ 1986,\ Appl.\ Environ,\ Microbiol.,\ 52:1215.$
- 6.McClain D. and Lee W. H., 1988, J. Assoc. off. Anal. Chem., 71:660.
- 7. Ajello G., Hayes P. and Fuley J., 1986, Abstracts of the Annual Meeting, ASM, Washington, D.C

### Storage and Shelf Life

Store below 30°C and prepared medium at 2-8°C. Use before expiry period on the label.