Fraser Broth Base

M1327

Intended use

Recommended, recommended as a primary as well as secondary enrichment medium, for the isolation and enumeration of *Listeria monocytogenes* from food and animal feeds.

Composition**

Ingredients	Gms / Litre
Peptone	5.000
Tryptone	5.000
Yeast extract	5.000
HM extract #	5.000
Sodium chloride	20.000
Disodium hydrogen phosphate dihydrate	12.000
Potassium dihydrogen phosphate	1.350
Esculin	1.000
Lithium chloride	3.000
Final pH (at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters
Equivalent to Meat extract

Directions

Suspend 54.92 grams of dehydrated medium in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C and aseptically add rehydrated contents of 1 vial of Fraser Selective Supplement (FD125I) and 2 vials of Fraser Supplement (FD141) to 1000 ml medium for primary enrichment or 1 vial of each to 500 ml medium for secondary enrichment. Mix well and dispense in tubes or flasks as desired.

Principle And Interpretation

L.monocytogenes primarily causes meningitis, encephalitis or septicemia in humans (1,8). In pregnant women, *L. monocytogenes* often causes influenza like bacteremic illness that, if untreated, may leaded to ammionitis and infection of the fetus, resulting in abortion, still birth or premature birth. Contaminated foods are the primary vehicles of transmission (7).

Fraser Broth Base is based on the formulation of Fraser and Sperber (9) is used for the detection of *Listeria* species in food products. Fraser Broth Base is formulated so as to provide optimum conditions for the growth of *Listeria.* Peptone, Tryptone, yeast extract, and beef extract make the media highly nutritive by providing essential nutrients including carbonaceous and nitrogenous substances. Phosphates maintain the buffering capacity of the medium. All *Listeria* species exhibit beta-glucosidase activity which is evident by the blackening of the media.

Listeria species hydrolyze esculin (substituted glucoside) to glucose and esculetin. The latter combines with ferric ions of ferric ammonium citrate (FD141), resulting in the formation of 6-7 dihydroxycoumarin, a black brown complex. Ferric ammonium citrate also enhances the growth of *L.monocytogenes* (6). The high salt tolerance (of sodium chloride) of *Listeria* is used as means to inhibit the growth of Enterococci. Lithium chloride is also used to inhibit Enterococci, which also possess the ability to hydrolyze esculin. Growth of accompanying bacteria is largely inhibited by the addition of Nalidixic acid and Acriflavin hydrochloride (FD125I).

The test sample under study is inoculated into the primary enrichment medium. After an incubation at 30°C for 18-24 hours, 0.1 ml is inoculated into Fraser Broth Base (M1327). After an incubation at 35-37°C for 24-48 hours, it is subcultured on Listeria Oxford Medium Base (M1145) or Listeria Identification Agar Base (PALCAM) (M1064).

Type of specimen

Food samples

Specimen Collection and Handling:

The test sample under study is inoculated into the primary enrichment medium. After an incubation at 30°C for 18-24 hours, 0.1 ml is inoculated into Fraser Broth Base (M1327). After an incubation at 35-37°C for 24-48 hours, it is subcultured on Listeria Oxford Medium Base (M1145) or Listeria Identification Agar Base (PALCAM) (M1064).

Warning and Precautions :

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations :

1. Further confirmation of organisms on selective media is required.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Basal medium : Yellow coloured clear solution with slight precipitate. After addition : Fluorescent yellow coloured clear solution with slight precipitate forms in tubes.

Reaction

Reaction of 5.49% w/v aqueous solution at 25°C. pH : 7.2±0.2

pH

7.00-7.40

Cultural Response

Cultural characteristics observed on addition of FD125I and FD141 after an incubation at 35-37°C for 24-48 hours.

Organism	Inoculum (CFU)	Growth	Esculin Hydrolysis
Escherichia coli ATCC 25922 (00013*)	>=10^4	inhibited	
Enterococcus faecalis ATCC 29212 (00087*)	>=10^4	inhibited	
Listeria monocytogenes subsp. *serovar 1 ATCC 19111* (00020*)	50-100	good-luxuriant	positive reaction, blackening of medium
Listeria monocytogenes ATCC 19112	50-100	good-luxuriant	positive reaction, blackening of medium
Listeria monocytogenes ATCC 19117	50-100	good-luxuriant	positive reaction, blackening of medium
Listeria monocytogenes ATCC 19118	50-100	good-luxuriant	positive reaction, blackening of medium
Staphylococcus aureus ATCC 25923 (00034*)	>=10^4	inhibited	

Key : *Corresponding WDCM numbers.

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (4,5).

Reference

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9. Schuchat A. B., Swaminathan and C. V. Broome, Clin. Microbiol., Rev. 4 : 169-1
10. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.

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