Fraser Broth Base
Fraser Broth Supplement

Intended Use
Fraser Broth Base is used with Fraser Broth Supplement in selectively enriching and detecting *Listeria*.

Summary and Explanation
First described in 1926 by Murray, Webb and Swann,[1] *Listeria monocytogenes* is a widespread problem in public health and the food industries. This organism has the ability to cause human illness and death, particularly in immunocompromised individuals and pregnant women.[2] The first reported foodborne outbreak of listeriosis was in 1985,[3] and since then, microbiological and epidemiological evidence from both sporadic and epidemic cases of listeriosis has indicated that the principle route of transmission is via the consumption of foodstuffs contaminated with *Listeria monocytogenes*.[4]

Implicated vehicles of transmission include turkey frankfurters,[5] coleslaw, pasteurized milk, Mexican-style cheese, paté and pickled pork tongue. The organism has been isolated from commercial dairy and other food processing plants and is ubiquitous in nature, being present in a wide range of unprocessed foods as well as in soil, sewage, silage and river water.[6]

Fraser Broth Base and Fraser Broth Supplement are based on the formulation of Fraser and Sperber.[7] The medium is use in the rapid detection of *Listeria* from food[8] and environmental samples. Many common food contaminants such as streptococci, enterococci, *Bacillus* species, *Escherichia coli*, *Pseudomonas aeruginosa* and *Proteus vulgaris* interfere with the isolation of *Listeria monocytogenes*.[9]

Listeria species grow on laboratory media over a pH range of 4.4-9.6, and can survive in foods of similar acidity for days or weeks.[10] *Listeria* spp. are microaerophilic, gram-positive, asporogenous, non-encapsulated, non-branching, regular, short, motile rods. Motility is most pronounced at 20°C.

Identification of *Listeria* is based on successful isolation of the organism, biochemical characterization and serological confirmation.

Principles of the Procedure
Peptones, beef extract and yeast extract provide nitrogen, vitamins and minerals. Sodium phosphate and potassium phosphate are buffering agents. Differentiation is aided by including ferric ammonium citrate in the final medium. Since all *Listeria* species hydrolyze esculin, the addition of ferric ions to the medium will detect the reaction. A blackening of the medium by cultures containing esculin-hydrolyzing bacteria is the result of the formation of 6,7-dihydroxycoumarin that reacts with the ferric ions.[7]

Selectivity is provided by the presence of lithium chloride, nalidixic acid and acriflavine in the formula. The high salt tolerance of *Listeria* is used as a means to inhibit growth of enterococci.

Formulae
Difco™ Fraser Broth Base
Approximate Formula* Per Liter

Pancreatic Digest of Casein	5.0	g
Proteose Peptone No. 3	5.0	g
Beef Extract	5.0	g
Yeast Extract	5.0	g
Sodium Chloride	20.0	g
Disodium Phosphate	9.6	g
Monopotassium Phosphate	1.35	g
Esculin	1.0	g
Nalidixic Acid	0.02	g
Acriflavine HCl	24.0	mg
Lithium Chloride	3.0	g

Difco™ Fraser Broth Supplement
Per 10 mL Vial

Ferric Ammonium Citrate	0.5	g

**Adjusted and/or supplemented as required to meet performance criteria.*

Directions for Preparation from Dehydrated Product
1. Suspend 55 g of the powder in 1 L of purified water. Mix thoroughly.
2. Heat with frequent agitation and boil for 1 minute to completely dissolve the powder.
3. Autoclave at 121°C for 15 minutes. Cool to room temperature.
4. Aseptically add 10 mL Fraser Broth Supplement. Mix well.
5. Test samples of the finished product for performance using stable, typical control cultures.

Procedure
To isolate *Listeria monocytogenes* from processed meats and poultry, the following procedure is recommended by the U.S. Department of Agriculture.[8]

1. Add 25 g of test material to 225 mL of UVM Modified Listeria Enrichment Broth and mix or blend thoroughly.
2. Incubate for 20-24 hours at 30°C.
3. Transfer 0.1 mL of the incubated broth to Fraser Broth. Incubate at 35°C for 26 ± 2 hours.
4. At 24 and 48 hours, streak the Fraser Broth culture to Modified Oxford Agar.
5. Incubate the Modified Oxford plates at 35°C for 24-48 hours.

Expected Results
1. Examine agar plates for suspect colonies. For further identification and confirmation of *Listeria* spp., consult appropriate references.[8,10,11]
2. Rapid slide and macroscopic tube tests can be used for definitive serological identification.

User Quality Control

Identity Specifications

Difco™ Fraser Broth Base

Dehydrated Appearance: Tan, free-flowing, homogeneous.

Solution:	5.5% solution, soluble in purified water upon boiling. Solution is medium amber, clear to slightly opalescent with a fine precipitate.
Prepared Appearance:	Medium amber, clear to slightly opalescent with a fine precipitate.
Reaction of 5.5% Solution at 25°C:	pH 7.2 ± 0.2

Difco™ Fraser Broth Supplement

Solution Appearance: Dark brown solution.

Cultural Response

Difco™ Fraser Broth Base and Fraser Broth Supplement

Prepare the medium per label directions. Add Fraser Broth Supplement. Inoculate and incubate at 35 ± 2°C and read for growth and blackening at 18-24 and 42-48 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY	ESCULIN REACTION
Enterococcus faecalis	29212	10^3-2×10^3	Marked to complete inhibition	–
Escherichia coli	25922	10^3-2×10^3	Marked to complete inhibition	–
Listeria monocytogenes	19114	10^2-10^3	Good	+
Listeria monocytogenes	19115	10^2-10^3	Good	+
Staphylococcus aureus	25923	10^3-2×10^3	Marked to complete inhibition	–

Uninoculated Tube

Listeria monocytogenes ATCC™ 19114

Limitations of the Procedure

1. Since *Listeria* species other than *L. monocytogenes* can grow on these media, an identification of *Listeria monocytogenes* must be confirmed by biochemical and serological testing.[11]
2. Poor growth and a weak esculin reaction may be seen after 40 hours incubation for some enterococci.

References

1. Murray, Webb and Swann. 1926. J. Pathol. Bacteriol. *29*:407.
2. Monk, Clavero, Beuchat, Doyle and Brackett. 1994. J. Food Prot. *57*:969.
3. Wehr. 1987. J. Assoc. Off. Anal. Chem. *70*:769.
4. Bremer and Osborne. 1995. J. Food Prot. *58*:604.
5. Grau and Vanderlinde. 1992. J. Food Prot. *55*:4.
6. Patel, Hwang, Beuchat, Doyle and Brackett. 1995. J. Food Prot. *58*:244.
7. Fraser and Sperber. 1988. J. Food Prot. *51*:762.
8. Lee and McClain. 1994. Laboratory Communication No. 57 (revised February 8, 1994). Food Safety and Inspection Service, Microbiology Division, USDA, Bethesda, Md.
9. Kramer and Jones. 1969. J. Appl. Bacteriol. *32*:381.
10. Ryser and Donnelly. 2001. *In* Downes and Ito (ed.), Compendium of methods for the microbiological examination of foods, 4th ed. American Public Health Association, Washington, D.C.
11. Bille, Rocourt and Swaminathan. 1999. *In* Murray, Baron, Pfaller, Tenover and Yolken (ed.), Manual of clinical microbiology, 7th ed. American Society for Microbiology, Washington, D.C.

Availability

Difco™ Fraser Broth Base

CCAM COMPF ISO USDA

Cat. No.	211767	Dehydrated – 500 g
	211766	Dehydrated – 2 kg

Difco™ Fraser Broth Supplement

CCAM COMPF ISO USDA

Cat. No.	211742	Vial – 6 × 10 mL*

**Store at 2-8°C.*