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, pate 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 used in the rapid detection of Listeria from food4 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
Esulin ................................................................. 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.

<table>
<thead>
<tr>
<th>ORGANISM</th>
<th>ATCC</th>
<th>INOCULUM CFU</th>
<th>RECOVERY</th>
<th>ESCULIN REACTION</th>
</tr>
</thead>
<tbody>
<tr>
<td>Enterococcus faecalis</td>
<td>29212</td>
<td>10^2·10^3</td>
<td>Marked to complete inhibition</td>
<td>–</td>
</tr>
<tr>
<td>Escherichia coli</td>
<td>25922</td>
<td>10^2·10^3</td>
<td>Marked to complete inhibition</td>
<td>–</td>
</tr>
<tr>
<td>Listeria monocytogenes</td>
<td>19114</td>
<td>10^3·10^3</td>
<td>Good</td>
<td>+</td>
</tr>
<tr>
<td>Listeria monocytogenes</td>
<td>19115</td>
<td>10^3·10^3</td>
<td>Good</td>
<td>+</td>
</tr>
<tr>
<td>Staphylococcus aureus</td>
<td>25923</td>
<td>10^2·10^3</td>
<td>Marked to complete inhibition</td>
<td>–</td>
</tr>
</tbody>
</table>

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
8. Lee and McClain. 1994. Laboratory Communication No. 57 (revised February 8, 1994); Food Safety and Inspection Service, Microbiology Division, USDA, Bethesda, Md.

Availability
Difco™ Fraser Broth Base
Cat. No. 211767 Dehydrated – 500 g
Cat. No. 211766 Dehydrated – 2 kg

Difco™ Fraser Broth Supplement
Cat. No. 211742 Vial – 6 x 10 mL*
*Store at 2-8°C.