

Instructions for Use

REF - BM0640

Fraser Broth

Bottled Media

1. Intended Use

BM0640 Fraser Broth is an enrichment media for the isolation of *Listeria* species from food and environmental samples.

2. Composition*

<u>Ingredient</u>	<u>g/L</u>
Meat peptone	5.0
Yeast extract	5.0
Beef extract	5.0
Tryptone	5.0
Aesculin	1.0
Disodium hydrogen phosphate (anhydrous)	9.6
Potassium dihydrogen phosphate	1.35
Sodium chloride	20.0
Lithium chloride	3.0
Nalidixic acid	0.02
Acriflavine	0.025
Ferric ammonium citrate	0.5

*Adjusted/supplemented as needed to meet performance requirements

3. Summary and Explanation

Developed in 1988 by Fraser ⁽¹⁾, Fraser Broth is the secondary enrichment medium recommended by ISO 11290 ⁽²⁾. Fraser Broth is formulated and tested to conform to ISO 11133 ⁽³⁾.

4. Principle

The peptones provide the required carbon, nitrogen, and vitamins in the medium. Sodium chloride maintains the osmotic balance and the disodium hydrogen phosphate and potassium dihydrogen phosphate are buffers. *Listeria* spp. will hydrolyse aesculin and the associated reaction with the ferric ammonium citrate gives rise to a blackening of the medium. Lithium chloride inhibits the growth of enterococci and nalidixic acid and acriflavine are further selective agents.

5. Physical Characteristics

	<u>Appearance and Colour</u>	<u>pH</u>
Medium	Yellow liquid	7.2 ± 0.2

6. Materials Provided

BM0640 Fraser Broth can be provided in pack of 50 x 10ml in Thin Wall Universals (product code: BM0640-M002-10). Each bottle is ink-jet printed with (abbreviated) product name, product code, lot number and expiry date.

7. Materials Needed but not Provided

Standard microbiological laboratory materials e.g., sterile loops or swabs, collection containers, incubators, media for isolation and quality control organisms.

8. Specimens

BM0640 Fraser Broth is suitable for the testing of the following specimens:

- Food and Environmental Specimens: products for human consumption, animal feed and environmental samples

Sampling and transport equipment must be used in accordance with the end user's suppliers' recommendations. Refer to appropriate standard method or local guidance on sample collection and subsequent processing.

9. Test Procedures and Interpretation of results

Food and Environmental Specimens:

Following the procedure in ISO 11290 ⁽²⁾:

After previous enrichment in E&O BM0647 Half Fraser Broth, transfer 0.1ml of primary culture broth to 10ml of BM0640 Fraser Broth (warmed to 37°C) and mix thoroughly. Incubate at 37 ± 1°C for 24 ± 2 hours. Sub-culture to selective plated media (e.g., E&O PP7021 COLOREX™ *Listeria* Agar) and incubate at 37 ± 1°C for 24 ± 2 hours ⁽²⁾.

After incubation, examine broths for darkening/turbidity, indicating growth; examine subcultures on agar for colonies. Perform further biochemical, serological, or mass spectroscopy testing to confirm identity of isolated colonies from subcultures if required. Refer to relevant local guidelines.

10. Quality Control

Organism	Incubation	Result (Specificity)
<i>L. monocytogenes</i> (NCTC 11994)	37 ± 1°C for 18-24 hours	Broth: Good turbidity. Subplate: Blue colonies with opaque halo
<i>L. monocytogenes</i> (NCTC 7973)	37 ± 1°C for 18-24 hours	Broth: Good turbidity. Subplate: Blue colonies with opaque halo
<i>E. coli</i> (NCTC 12241)	37 ± 1°C for 18-24 hours	Inhibited
<i>E. faecalis</i> (NCTC 12697)	37 ± 1°C for 18-24 hours	Inhibited
(N.B. Subcultures made onto E&O PP7021 COLOREX™ Listeria Agar and incubated at 37 ± 1°C for 24 ± 2 hours)		

It is the responsibility of the user to perform Quality Control testing taking into consideration the intended use of the medium and in agreement with any local relevant guidelines (e.g., frequency, strains used, atmosphere, incubation temperature).

11. Performance

To fully verify BM0640 Fraser Broth performance, samples were tested to assess colour change and turbidity/colony morphology and recovery level when incubated at 37 ± 1°C for 24 ± 2 hours, before being subcultured on E&O PP7021 COLOREX™ Listeria Agar and incubated at 37 ± 1°C for 24 ± 2 hours. All samples showed expected recovery and morphology of the required test organisms: *Listeria monocytogenes* (NCTC 11994) and *Listeria monocytogenes* (NCTC 7973), and no recovery of the non-target test organisms: *Escherichia coli* (NCTC 12241) and *Enterococcus faecalis* (NCTC 12697). Therefore, it can be concluded that BM0640 Fraser Broth, meets performance criteria when used according to the instructions outlined above. Trend analysis data available upon request.

12. Limitations of the Media

- Due to natural variation, some strains may grow poorly on this medium.

13. Precautions and Warnings

This product is considered non-hazardous under CLP regulations. Wear such PPE as recommended by laboratory COSSH assessment. During and after use, always handle all materials in a manner conforming to Good Laboratory Practices and consider that material under test should be regarded as a potential biohazard if mishandled.

Refer to E&O Bottled Media Material Safety Data Sheet.

14. Storage conditions and Shelf life

Store BM0640 in the original bottle with the lid tightly closed at between 2 and 8°C. Kept under these conditions it may be used up to date of expiry shown on product label.

Dispose of in accordance with local and national authority requirements. Do not use media if it is contaminated, discoloured or out of date. Additionally, do not use medium if it has been stored inappropriately or the packaging has been damaged.

15. References

1. Fraser, J. and Sperber, W. (1988) Rapid Detection of *Listeria* spp. in Food and Environmental Samples by Esculin Hydrolysis. *Journal of Food Protection*, 51(10), pp.762-765.
2. ISO (2017) 11290-1:2017 Microbiology of the food chain - Horizontal method for the detection and enumeration of *Listeria monocytogenes* and of *Listeria* spp. - Part 1: Detection method. Geneva: ISO.
3. ISO (2014) 11133:2014 Microbiology of food, animal feed and water — Preparation, production, storage and performance testing of culture media. Geneva: ISO.

Version History*

001 15/03/23 - New Document Created

*Note: minor typographical, grammatical, and formatting changes are not included in the revision history.



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IFU/BM0640 REV. 001

TABLE OF APPLICABLE SYMBOLS

REF Catalogue number	LOT Batch code		Manufacturer	Use by
Temperature limitation	Contents sufficient for <n> tests	Consult Instructions for Use	Keep away from direct light	Store in a dry place