BIOMÉRIEUX

REF AEB140412/AEB140413/AEB140417
REF AEB240409

140412 O - en - 2015/05 EN

Half-Fraser Broth

For microbiological control only

Selective pre-enrichment of *Listeria* from food and environmental specimens

SUMMARY AND EXPLANATION

Half-Fraser Broth is used for the selective pre-enrichment of *Listeria* from food and environmental specimens.
It complies with the standard NF EN ISO 11290-1 (1) and amendment A1 (2) for the detection of *Listeria monocytogenes* in food.

PRINCIPLE

The broth contains a rich nutritive base, consisting of a mixture of peptones and a buffer that maintains the pH close to neutral to favor the growth of the main species of *Listeria.*
The broth is made selective by the incorporation of lithium, acriflavin and nalidixic acid (3). It is therefore particularly efficient for products with a high concentration of microbes (4). The formulation of Half-Fraser (half concentration in antibiotics and acriflavin) has been developed to limit the selective capacity of the original medium (5), in order to allow better growth of stressed *Listeria*.

CONTENT OF THE KIT

Dehydrated medium	
REF AEB140412	500 g bottle
REF AEB140413	5 Kg bottle
REF AEB140417	10 Kg bottle
Ready weighed dry medium	
REF AEB240409	For 9 litres

COMPOSITION

Theoretical formula after reconstitution of the medium and addition of supplements

This medium can be adjusted and/or supplemented according to the performance criteria required :

Proteose peptone	5 g
Casein peptone	5 g
Meat extract	5 g
Yeast extract	5 g
NaCl	20 g
Disodium Hydrogen Phosphate, anhydrous	9.6 (1) g
Potassium dihydrogen Phosphate	1.35 g
Esculin	1 g
Lithium chloride	3 g
Ammonium ferric citrate	0.5 g
Acriflavin	0.0125 g
Nalidixic acid	0.01 g
Purified water	1 l

pH 7.2

(1) equivalent to 12g of Disodium Phosphate dibasic (Na_2HPO_4, $2H_2O$)

REAGENTS AND MATERIAL REQUIRED BUT NOT PROVIDED

Reagents :

- Fraser additive (Ref. AEB110422S)
- FRASER Broth (Ref. 42 072).
- PALCAM Agar (Ref. AEB522050 or Ref.AEB522049).
- Oxford Agar (Ref. AEB522000).
- ALOA® Agar (Ref.AEB520080 or Ref. AEB520079).

Material :

- Bacteriology incubator.
- Autoclave
- Bottles
- Blending Bags.

WARNINGS AND PRECAUTIONS

- **For microbiological control only.**
- **For professional use only.**
- This kit contains products of animal origin. Certified knowledge of the origin and/or sanitary state of the animals does not totally guarantee the absence of transmissible pathogenic agents. It is therefore recommended that these products be treated as potentially infectious, and handled observing the usual safety precautions (do not ingest or inhale).
- All specimens, microbial cultures and inoculated products should be considered infectious and handled appropriately. Aseptic technique and usual precautions for handling the bacterial group studied should be observed throughout this procedure. Refer to "CLSI® M29-A, *Protection of Laboratory Workers from Occupationally; Acquired Infections; Approved Guideline* – Current revision". For additional information on handling precautions, refer to "Biosafety in Microbiological and Biomedical Laboratories – CDC/NIH – Latest edition", or the current regulations in the country of use.
- *Listeria monocytogenes* may cause serious illness, particularly in certain groups at risk such as pregnant women, immunocompromised patients and the elderly.
- Culture media should not be used as manufacturing material or components.
- Do not use reagents past the expiry date.
- Do not use media which are not homogeneous (presence of lumps).
- Avoid opening bottles in a humid atmosphere (steam, condensation, etc.).
- The medium should be used according to the procedure indicated in this package insert. Any change or modification in the procedure may affect the results

STORAGE CONDITIONS

- **Store the Half-Fraser broth until the expiry date between 1 et 30 °C.**
- Store in a dry place

SPECIMENS

Follow the recommendations in the current standards to perform specimen collection and preparation.

INSTRUCTIONS FOR USE

Preparation

1. Pour 55 grams of powder into 1 litre of purified water.

Note 1: If necessary, you may bring to the boil to obtain perfect dissolution.

2. Dispatch into 225ml bottles.
3. Autoclave for 15 minutes at 121°C. DO NOT OVERHEAT.

4. Before inoculation, add aseptically to each flask 2,25ml of supplement for Fraser.

Note 2: Either liquid or dehydrated ammonium ferric citrate may be added before autoclaving, leading to a final concentration of 0,5 gram per litre of broth. A slight precipitate may appear. It is not prejudicial to the analysis.

If necessary, allow the Half-Fraser broths to come to room temperature.

Pre-enrichment :

A primary 1/10 dilution of the sample is generally performed in the blending bag.

After mixing, incubate at 30°C for 24 ± 2 hours. After pre-enrichment, subculture 0.1 ml in 10 ml of Fraser Broth. Incubate at 37°C for 48 hours. After pre-enrichment and enrichment, isolate in parallel on selective ALOA® Agar and on a medium of choice (Oxford or PALCAM).

READING AND INTERPRETATION

Refer to the indications in the corresponding reagent package insert.

QUALITY CONTROL

Half-Fraser Broth is designed and developed to meet the strictest quality requirements.
The results of the strains tested in the batch by batch quality control are given on the quality control certificate available on our website (www.biomerieux.com).

LIMITATIONS OF THE METHOD

- It has been found that Half-Fraser Broth is not suitable for the enrichment of some strains of *Listeria grayi.* This non-pathogenic species is mainly found in soil and is rarely isolated from food samples.
- The broth may show a slightly greenish precipitate that does not alter its performance.

WASTE DISPOSAL

Unused reagents may be considered as non hazardous waste and disposed of accordingly.
Dispose of all used reagents as well as any other contaminated disposable materials following procedures for infectious or potentially infectious products.
It is the responsibility of each laboratory to handle waste and effluents produced according to their nature and degree of hazardousness and to treat and dispose of them (or have them treated and disposed of) in accordance with any applicable regulations.

LITERATURE REFERENCES

1. NF EN ISO 11290-1 (1997). Microbiology of food and animal feeding stuffs - Horizontal method for the detection and enumeration of *Listeria monocytogenes* - Part 1 : detection method.
2. NF EN ISO 11290-1/A1 (February 2005) – Microbiology of food and animal feeding stuffs – Horizontal method for the detection and enumeration of *Listeria monocytogenes* - Part 1: detection method - Amendment 1: modification of the isolation media, of the haemolysis test and inclusion of precision data.
3. FRASER J.A., SPERBER W.H. - Rapid Detection of *Listeria* spp. in Food and Environmental Samples by Esculin Hydrolysis - *J. of Food protection*, 1988,vol. 51, p.762-765.
4. RODRIGUEZ L, FERNANDEZ G., GARAYZABAL J. et al. - New Methodology for the Isolation of *Listeria* Microorganisms from Heavily Contaminated Environments. - *Applied and Environmental Microbiology*, 1984, vol. 47, p. 1188-1190.
5. HOLBROOK R., ANDERSON J.M., BRIGGS T.A. et al.- Faster detection of *Listeria* in food using rapid immunoassay following culture - 3rd World Congress foodborne infections and intoxications, (16-19 June 1992), Berlin, p.1208-1210.

INDEX OF SYMBOLS

Symbol	Meaning
	Catalogue number
	Manufacturer
	Temperature limit
	Use by date
	Batch code
	Consult Instructions for Use
	Keep dry
	Date of manufacture

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