TECHNICAL DATA SHEET

COMPLETE HALF FRASER BROTH

SELECTIVE PRIMARY ENRICHMENT FOR *LISTERIA*

1 INTENDED USE

Half-Fraser Broth is used for the selective and differential enrichment (primary enrichment broth) of *Listeria monocytogenes* and *Listeria* spp. in food products, according to the standard NF EN ISO 11290-1.
The medium is also used in alternative rapid methods for the detection and enumeration of *Listeria monocytogenes* or *Listeria* spp.

2 HISTORY

The medium studied by Fraser *et al.* in 1988 is a modification of the formulation of Donnelly and Baigent. The composition of the base is identical to that of UVM Broth and was modified by the addition of lithium chloride as selective agent and of ferric ammonium citrate to visualize cultures that hydrolyze esculin, by the resultant blackening of the medium.

3 PRINCIPLES

The very good recovery of *Listeria monocytogenes* is assured by the concentration differences in nalidixic acid and acriflavin between Half-Fraser and Fraser, as well as the two enrichment steps themselves. Half-Fraser Broth allows the primary enrichment step, with secondary enrichment being performed in Fraser Broth.

Polypeptone, yeast extract and meat extract furnish the nutrients required for the growth of *Listeria*.

The high sodium chloride content increases the selectivity of the medium.

Phosphates buffer the pH of the medium.

Esculin is hydrolyzed by *Listeria* to glucose and esculetin, the latter compound forming a black complex with ferric ions supplied by ferric citrate, added just before use, which also favors the growth of *Listeria*.

Lithium chloride inhibits the growth of most enterococci which can also hydrolyze esculin.

Nalidixic acid blocks the DNA replication of bacteria sensitive to this antibacterial agent.

The growth of secondary Gram-positive microflora is inhibited by acriflavin.

4 TYPICAL COMPOSITION

The composition can be adjusted / supplemented to obtain optimal results.
For 1 liter of media :

- Enzymatic digest of animal tissues ..... 5,0 g
- Enzymatic digest of casein ..... 5,0 g
- Yeast extract ..... 5,0 g
- Meat extract ..... 5,0 g
- Sodium chloride ..... 20,0 g
- Disodium phosphate, anhydrous ..... 9,6 g
- Monopotassium phosphate ..... 1,35 g
- Esculin ..... 1,0 g
- Lithium chloride ..... 3,0 g
- Nalidixic acid ..... 10,0 mg
- Acriflavin (chlorhydrate) ..... 12,5 mg
- Ferric ammonium citrate ..... 0,50 g

pH of the ready-to-use medium at 25 °C : 7,2 ± 0,2.

5 PREPARATION

- Dissolve 55.0 g of dehydrated medium (BK215) in 1 liter of distilled or deionized water.
- Stir slowly until complete dissolution.
- Dispense in flasks at 225 mL per flask.
- Sterilize in an autoclave at 110°C for 15 minutes.
- Cool the medium to room temperature.

✓ **Reconstitution :**
55,0 g/L

✓ **Sterilization :**
15 min at 110°C

6 INSTRUCTIONS FOR USE

- To media prepared as above, or to complete media in ready-to-use vials or bags (BM016, BM133, BM134), aseptically add 25 g of the product to test.
- Mix well.
- Incubate at 30°C for (24 ± 2) hours.

✓ **Inoculation :**
25 g in 225 mL

✓ **Incubation :**
24 ± 2 h at 30°C

NOTE
The half-fraser broth may also be used as diluent for the enumeration of *Listeria monocytogenes* (NF VALIDATION, BKR 23/05-12/07).

7 RESULTS

All tubes, with or without blackening, must be transferred to selective isolation media or secondary enrichment media. A minimum period of 24 hours is necessary to permit the visualization of the black color.

Transfer 0,1 mL from each tube into Fraser broth for standardized methods.
Transfer onto COMPASS Listeria Agar in the context of rapid alternative methods of *Listeria monocytogenes* and *Listeria* spp. detection (NF VALIDATION, BKR 23/02-11/02).

NOTE :
For other uses, follow the current reference method.

8 QUALITY CONTROL

Dehydrated media : yellowish powder, free-flowing and homogeneous.
Prepared media (complete) : amber yellow solution, with bluish glints, can present a slight precipitate.

Typical culture response after 24 hours of incubation at 30°C, followed by subculture (NF EN ISO 11133) :

Microorganisms		Growth
Listeria monocytogenes 4b + *Enterococcus faecalis* + *Escherichia coli*	WDCM 00021 WDCM 00087 WDCM 00013	> 10 characteristic colonies
Enterococccus faecalis *Escherichia coli*	WDCM 00087 WDCM 00013	< 100 colonies Inhibited

9 STORAGE / SHELF LIFE

Dehydrated media : 2-30 °C.
The expiration date is indicated on the label.

Complete media prepared in vials (*) : 180 days at 2-8 °C, shielded from light.
60 days at 2-25 °C, shielded from light.
(*) Benchmark value, determined in standard conditions of preparation, following manufacturer's instructions.

10 PACKAGING

Dehydrated media
500 g bottle BK215HA
5 kg drum BK215GC

Ready-to-use media in vials :
10 x 225 mL vials BM01608

Ready-to-use media in flexible bags :
3 x 3 L bags BM13308
2 x 5 L bags BM13408

11 BIBLIOGRAPHY

Donnelly, C.W., and Baigent, G.J.. 1986. Method for flow cytometric detection of *Listeria monocytogenes* in milk. Applied and Environmental Microbiology, **52** : 689-695.

Fraser, J.A., and Sperber, W.H.. 1988. Rapid detection of *Listeria* spp. in food and environmental samples by esculin hydrolysis. Journal of Food Protection, **51** : 762-765.

NF EN ISO 11290-1. Février 1997. Microbiologie des aliments. Méthode horizontale pour la recherche et le dénombrement de *Listeria monocytogenes.* Partie 1 : Méthode de recherche.

NF EN ISO 11290-1/A1. Février 2005. Microbiologie des aliments. Méthode horizontale pour la recherche et le dénombrement de *Listeria monocytogenes.* Partie 1 : Méthode de recherche. Amendement 1 : Modification des milieux d'isolement, de la recherche de l'hémolyse et introduction de données de fidélité.

NF EN ISO 11133. Juillet 2014. Microbiologie des aliments, des aliments pour animaux et de l'eau. Préparation, production, stockage et essais de performance des milieux de culture.

12 ADDITIONAL INFORMATION

The information provided on the labels take precedence over the formulations or instructions described in this document and are susceptible to modification at any time, without warning.

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