

LISTERIA ENRICHMENT BROTH BASE FRASER ISO 11290-1

CAT N°: 1120

Enrichment medium for the detection and enumeration of *Listeria* in food and environmental samples

FORMULA IN g/l

Sodium Chloride	20.00	Beef Extract	5.00
Disodium Hydrogen Phosphate	12.00	Lithium Chloride	3.00
Tryptone	5.00	Monopotassium Phosphate	1.35
Proteose Peptone	5.00	Esculin	1.00
Yeast Extract	5.00		

Final pH 7.2 ± 0.2 at 25°C

PREPARATION

Suspend 28.7 grams of the medium in 500 ml. of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Sterilize in autoclave at 121°C for 15 minutes. Cool to 45-50°C and aseptically add one vial of Fraser Listeria Selective Supplement (Cat. 6001) for preparing Fraser or Half Fraser Listeria Selective Supplement (Cat. 6002) reconstituted in 5 ml of sterile distilled water. Homogenize gently and dispense into sterile containers. The prepared medium should be stored at 2-8°C. The color is amber.

The dehydrated medium should be homogeneous, free-flowing and beige in color. If there are any physical changes, discard the medium.

Fraser Listeria Selective Supplement (Cat. 6001)

(Composition: each vial for 500ml)

Vial A: Ferric Ammonium Citrate250 mg
Vial B: Acryflavine12.5 mg
Nalidixic Acid10 mg

Half Fraser Listeria Selective Supplement (Cat. 6002)

(Composition: each vial for 500ml)

Vial A: Ferric Ammonium Citrate 250 mg
Vial B: Acryflavine6.25 mg
Nalidixic Acid5 mg

USES

LISTERIA ENRICHMENT BROTH BASE FRASER is an appropriate medium for the selective enrichment of *Listeria* in the two-step method according to ISO 11290-2, for the preparation of Fraser or Half Fraser Broth by adding the respective supplements.

It is recommended for the detection of *Listeria spp.* in food products and in samples from the environment. All *Listeria* species hydrolyze the Esculin to esculetin, which reacts with iron ions producing a blackening of the medium. Another advantage of this medium is that the addition of Ferric ammonium citrate improves the growth of *L. monocytogenes*. Lithium chloride included in the medium, along with Nalidixic acid and Acryflavine from the supplement, inhibit the growth of the accompanying flora, which can hydrolyze the esculin. The high amount of Sodium chloride inhibits the growth of enterococci. Tryptone, Proteose Peptone and Beef extract provide nitrogen, vitamins, minerals and amino acids essential for growth. Yeast extract is the source of vitamins, particularly of the B-group. Phosphate Salts act as a buffer system.

Primary enrichment of Half Fraser medium and mix thoroughly. Incubate for 24 ± 2 hours at 30°C . Secondary enrichment: transfer 0.1 ml of incubated Half Fraser medium to 10 ml Fraser Broth. Incubate at $35\text{--}37^\circ\text{C}$ for 48 ± 2 hours. Compare each inoculated tube with a non-inoculated control tube with a white background. After incubation of the primary and secondary enrichment, inoculate the tubes in Agar Oxford Cat. 1133 (with supplement Cat.6003) and Agar Palcam Cat. 1141 (with supplement Cat.6004). Confirm the suspicious colonies.

MICROBIOLOGICAL TEST

The following results were obtained in the performance of the medium from type cultures with Listeria Enrichment Broth base Fraser with Half Fraser Listeria selective supplement (Cat. 6002) added, after incubation at a temperature of $30 \pm 1^\circ\text{C}$, in aerobic conditions and observed after 24 ± 2 hours, and with Listeria Enrichment Broth base Fraser with Fraser Listeria selective supplement (Cat. 6001) added, after incubation at temperature of $35 \pm 1^\circ\text{C}$ after 48 ± 2 hours.

According ISO 11133 24 ± 2 h/ $30 \pm 1^\circ\text{C}$ (Productivity and Selectivity) Half Fraser, 48 ± 2 h/ $37 \pm 1^\circ\text{C}$ (Productivity and Selectivity) Fraser

Microorganisms	Inoculum (cfu/ml)	Productivity Qualitative	Selectivity Qualitative	Characteristic Reaction
<i>Listeria monocytogenes</i> ATCC 13932 + <i>Escherichia coli</i> ATCC 8739 + <i>Enterococcus faecalis</i> ATCC 19433	≤ 100 $\geq 10^3$ $\geq 10^3$	>10 on Chromogenic Agar Listeria Blue green colonies with opaque halo		Blue green colonies with opaque halo
<i>Listeria monocytogenes</i> ATCC 13932 + <i>Escherichia coli</i> ATCC 8739 + <i>Enterococcus faecalis</i> ATCC 19433	≤ 100 $\geq 10^3$ $\geq 10^3$	>10 on Chromogenic Agar Listeria Blue green colonies with opaque halo		Blue green colonies with opaque halo
<i>Escherichia coli</i> ATCC 8739	$10^4/10^6$		Total Inhibition (on TSA)	
<i>Enterococcus faecalis</i> ATCC 19433	$10^4/10^6$		<100 (on TSA)	

BIBLIOGRAPHY

ISO NORMATIVE 11290-1 Microbiology of food and animal feeding stuffs -- Horizontal method for the detection and enumeration of *Listeria monocytogenes* -- Part 1: Detection method

Fraser J.A. and Sperber W.H (1988) McClain D. and Lee W.H(1988)



STORAGE

Once opened keep powdered medium closed to avoid hydration.

