## **Fraser Supplement**

## PRODUCT INFORMATION

N001-5g - Nalidixic Acid, Powder, 5g

N001-25g - Nalidixic Acid, Powder, 25g

N001-100g - Nalidixic Acid, Powder, 100g

### DESCRIPTION

Fraser Broth Base with Fraser Supplement is a secondary selective diagnostic enrichment medium for the isolation of *Listeria* spp. from food and environmental specimens.

### **BACKGROUND**

Ammonium ferric citrate is a food additive with E number E381 used as an acidity regulator.

Nalidixic acid is the first of the synthetic quinolone antibiotics. Nalidixic acid is effective against both gram-positive and gram-negative bacteria. In lower concentrations, it acts in a bacteriostatic manner; that is, it inhibits growth and reproduction. In higher concentrations, it is bactericidal, meaning that it kills bacteria instead of merely inhibiting their growth.

#### Mechanism of action

## APPLICATION IN FRASER BROTH BASE

Fraser Broth is a modification of the USDA-FSIS (United States Department of Agriculture-Food Safety Inspection Service) UVM secondary enrichment broth and is based on the formula described by Fraser and Sperber. It contains ferric ammonium citrate and lithium chloride. Blackening of the medium is presumptive evidence of the presence of *Listeria*. Contrary to early indications, cultures which do not blacken cannot be assumed to be *Listeria*-free. All Fraser Broth enrichment cultures should be subcultured to plating medium.

The medium is intended for the isolation of *Listeria* spp. from food and environmental samples when used as the secondary enrichment medium in the USDA-FSIS methodology for Listeria isolation. It is generally accepted that the USDA-FSIS two stage enrichment method employing UVM primary and secondary enrichment broths is the most suitable for

the examination of meat products. Fraser Broth has proven to be remarkably accurate in detecting *Listeria* spp. in food and environmental samples.

All *Listeria* spp. hydrolyse aesculin to aesculetin. Aesculetin reacts with ferric ions which results in blackening. Another possible advantage to the addition of ferric ammonium citrate is that it has been shown that ferric ions enhance the growth of Listeria monocytogenes3. Lithium chloride is included in the medium to inhibit the growth of enterococci which can also hydrolyse aesculin. Care must be taken when using Fraser Broth with DNA probe methodology because the high salt content of the medium may have an inhibitory effect on detection.

#### **Content concentrations**

Typical Formula*	mg/litre
Fraser Broth Base	
Proteose peptone	5
Tryptone	5
`Lab-Lemco' powder	5
Yeast extract	5
Sodium chloride	20
Di-sodium hydrogen phosphate	12
Potassium dihydrogen phosphate	1.35
Aesculin	1
Lithium chloride	3
Final pH 7.2 ± 0.2 @ 25°C	
Fraser Supplement	
Ferric ammonium citrate	500
Nalidixic acid	20
Acriflavine hydrochloride	25
* Adjusted as required to meet performance standards	

**Table 1 - Typical Formula for Fraser Broth Base and Fraser Supplement** 

### **METHOD**

## Preparation

Suspend appreciate amount of Fraser Broth Base in distilled water and mix well to dissolve completely. Sterilise by autoclaving at 121°C for 15 minutes. Cool to below 50°C, and aseptically add the contents of Fraser Selective Supplement reconstituted as directed in the product insert. Mix well and distribute into sterile containers.

Suspend appreciate mount of Listeria Enrichment

Broth Base in distilled water. Sterilise by autoclaving at 121°C for 15 minutes. Cool to 50°C.

## To Prepare Listeria Primary Selective Enrichment Medium (UVM I)

Aseptically add the contents of Listeria Primary Selective Enrichment Supplement (UVM I) reconstituted as directed. Mix well and distribute into sterile containers.

# To Prepare Listeria Secondary Selective Enrichment Medium (UVM II)

Aseptically add the contents of Listeria Secondary Selective Enrichment Supplement (UVM II) reconstituted as directed. Mix well and distribute into sterile containers.

#### **Protocol**

#### **Primary Enrichment**

- 1. Inoculate 10ml of Fraser Broth with 0.1ml of the primary enrichment broth (i.e. FDA or UVM I enrichment broth) which has been incubated for 20 to 24 hours.
- 2. Incubate at 35°C for  $26 \pm 2$  hours in air.
- 3. Compare each inoculated tube to an inoculated control against a white background. Tubes that darken or turn black should be subcultured on to Listeria Selective Medium or PALCAM Medium. Tubes that retain the original yellow colour should also be inoculated on plating media and confirmed as free from *Listeria* spp. before discarding.

Note: Fraser Medium should be incubated for  $26 \pm 2$  hours to ensure at least 24 hours incubation period to permit the development of the black colour.

## **Quality control**

Positive control:

Listeria monocytogenes ATCC® 7466: Blackening

Negative control:

Psudomonas aeruginosa ATCC® 27853: No blackening

#### REFERENCES

- 1. Fraser J.A. and Sperber W.H. (1988) J. Food Protect. 51, No.10, 762-765.
- 2. McClain D. and Lee W.H. (1988) J. Assoc. Off. Anal. Chem. 71, NO.3, 660-664
- 3. Cowart R.E. and Foster B.G. (1985) J. Infect. Dis. 151, 721-730.
- 4. Partis L., Newton K., Marby J. and Wells R.J. (1994) Appl. Env. Microbiol. 60, 1693-1694.
- 5. Microbiology of Food and Animal Feeding Stuffs Horizontal method for the detection and enumeration of Listeria monocytogenes Part. 1: Detection Method BS EN ISO 11290:1 1997.