IFU

Instructions for Use

FRASER BROTH SUPPLEMENT

Cat. no. Z210	Fraser Broth Supplement, 100ml Serum Bottle, 100ml	1 bottle

INTENDED USE

Hardy Diagnostics Fraser Broth Supplement is used with Half Fraser Broth Base for the selective enrichment and detection of *Listeria* spp. This supplement is also used in conjunction with Fraser Broth Base for the detection of *Listeria monocytogenes* from food and environmental samples.

This product is not intended to be used for the diagnosis of human disease.

SUMMARY

First described by Murray, Webb, and Swann, *Listeria monocytogenes* is a widespread problem in public health and food industry.[6,8] This microorganism can cause human illness and death, particularly in immunocompromised individuals and in the unborn fetus of pregnant women. The first reported food-borne outbreak of listeriosis was in 1985. Since then, microbiological and epidemiological evidence from both sporadic and epidemic cases of listeriosis has shown that the principal route of transmission is via the consumption of foodstuffs contaminated with *Listeria monocytogenes*.[7,10]

Potential vehicles of transmission implicated in the spread of disease include turkey frankfurters, coleslaw, pasteurized milk, Mexican-style and other soft cheeses, pate, and pickled pork tongue. Moreover, the organism has been isolated from commercial dairy and other food processing plants. It is ubiquitous in nature, being present in a wide range of unprocessed foods, in soil, sewage, silage, and river water. *Listeria* also possess the ability to grow over a pH range of 5.0-9.6 and can survive in food products with pH levels outside of these parameters.[10]

Hardy Diagnostics Fraser Broth Base is based on the formulation of Fraser and Sperber.[7] It contains pancreatic digest of casein, proteose peptone, and yeast extract as nitrogen, vitamin, and mineral sources. Hardy Diagnostics Half Fraser Broth Base is a modification of Fraser Broth Base in which the nalidixic acid and acriflavine concentrations have been reduced to 10.0mg/L and 12.5mg/L. The supplementation of Hardy Diagnostics Fraser Broth Base aids in differentiation by including ferric ammonium citrate in the final medium. Since all *Listeria* species hydrolyze esculin, the addition of ferric ions to the medium will detect the reaction. Ferric ions, combined with esculetin, produce a dark brown to black color change in the medium surrounding the colonies.

Selectivity of Fraser Broth and Half Fraser Broth is provided by the presence of lithium chloride, nalidixic acid, and acriflavine in the formula. The high salt tolerance of *Listeria* is used as a means to inhibit growth of enterococci.

FORMULA

Ingredients per liter of deionized water:*

Ferric Ammonium Citrate	5.0gm

* Adjusted and/or supplemented as required to meet performance criteria.

STORAGE AND SHELF LIFE

Storage: Upon receipt store at 2-8ºC. away from direct light. Media should not be used if there are any signs of deterioration, discoloration, contamination, or if the expiration date has passed. Product is light and temperature sensitive; protect from light, excessive heat, moisture, and freezing.

The expiration date on the product label applies to the product in its intact packaging when stored as directed. The product may be used and tested up to the expiration date on the product label and incubated for the recommended incubation times as stated below.

Refer to the document "Storage" for more information.

PRECAUTIONS

This product may contain components of animal origin. Certified knowledge of the origin and/or sanitary state of the animals does not guarantee the absence of transmissible pathogenic agents. Therefore, it is recommended that these products be treated as potentially infectious, and handle observing the usual universal blood precautions. Do not ingest, inhale, or allow to come into contact with skin.

This product is for laboratory use only. It is to be used only by adequately trained and qualified laboratory personnel. Observe approved biohazard precautions and aseptic techniques. All laboratory specimens should be considered infectious and handled according to "standard precautions." Refer to the document "Guidelines for Isolation Precautions" from the Centers for Disease Control and Prevention.

For additional information regarding specific precautions for the prevention of the transmission of all infectious agents from laboratory instruments and materials, and for recommendations for the management of exposure to infectious disease, refer to CLSI document M29: *Protection of Laboratory Workers from Occupationally Acquired Infections.*

Sterilize all biohazard waste before disposal.

Refer to the document "Precautions When Using Media" for more information.

METHOD OF PREPARATION FOR PREPARED CULTURE MEDIA

1. Suspend 55.0gm of the dehydrated culture media (Fraser Broth Base, Cat. no. C5780 or Half Fraser Broth Base, Cat. n. C8040) in 1 liter of distilled or deionized water.

2. Heat, while stirring to dissolve completely. Avoid overheating.

3. Dispense in appropriate containers and autoclave at 121ºC. for 15 minutes.

4. Cool to room temperature. Aseptically add enrichment, 10ml of a filter sterilized 5% aqueous solution of ferric ammonium citrate (Fraser Broth Supplement, Cat. no. Z210).

5. Dispense as desired into sterile containers.

PROCEDURE

Consult listed references for information regarding sample preparation and processing.[4,7,8,10]

To isolate *Listeria monocytogenes* from processed meats and poultry, the following procedure is recommended by the U.S.D.A:

1. Add 25.0gm of test material to 225ml Modified Listeria Enrichment Broth and mix or blend thoroughly.

2. Incubate for 20-24 hours at 30ºC.

3. Transfer 0.1ml of the incubated broth to the prepared Fraser Broth. Incubate at 35ºC. for 26 +/- 2.0 hours.

4. At 24 and 48 hours, streak the prepared Fraser Broth culture to Modified Oxford Agar.

5. Incubate the Modified Oxford plates at 35ºC. for 24-48 hours.

INTERPRETATION OF RESULTS

Examine plates for typical *Listeria* colonies. *Listeria monocytogenes* will appear as clear colonies surrounded by a blackening in the media.

LIMITATIONS

It is recommended that biochemical, immunological, molecular, or mass spectrometry testing be performed on colonies from pure culture for complete identification of bacteria and/or fungi.

Fraser Broth Supplement is not an isolation medium. This product is used in conjunction with Fraser Broth Base or Half Fraser Broth Base.

Refer to the document "Limitations of Procedures and Warranty" for more information.

MATERIALS REQUIRED BUT NOT PROVIDED

Standard microbiological supplies and equipment such as autoclaves, Fraser Broth Base (Cat. no. C5780), Half Fraser Broth Base (Cat. no. C8040), incinerators, and incubators, etc., as well as serological and biochemical reagents are not provided.

QUALITY CONTROL

Fraser Broth Supplement is not a growth medium. It is tested for sterility only.

USER QUALITY CONTROL

End users of commercially prepared culture media should perform QC testing in accordance with applicable government regulatory agencies, and in compliance with accreditation requirements. Hardy Diagnostics recommends end users check for signs of contamination and deterioration and, if dictated by laboratory quality control procedures or regulation, perform quality control testing to demonstrate growth or a positive reaction and to demonstrate inhibition or a negative reaction, if applicable. Hardy Diagnostics quality control testing is documented on the certificate of analysis (CofA) available from Hardy Diagnostics Certificate of Analysis website. Also refer to the document "Finished Product Quality Control Procedures," and the CLSI document M22-A3 *Quality Assurance for Commercially Prepared Microbiological Culture Media* for more information on the appropriate QC procedures. See the references below.

PHYSICAL APPEARANCE

Fraser Broth Supplement should appear clear, and dark brown in color.

REFERENCES

1. American Public Health Association. *Standard Methods for the Examination of Dairy Products*, APHA, Washington, D.C.

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3. Grau, F.H., et al. 1995. Occurrence, numbers, and growth of *Listeria monocytogenes* on some vacuum-packaged

processed meats. *J. Food Prot.* ; 55:4-4.

4. International Dairy Federation. 1990. Milk and milk products - detection of *Listeria monocytogenes* . IDF Provisional International Standard No. 143. International Dairy Federation, Brussels, 1990.

5. Kramer, P.A., et al. 1969. Media selective for *Listeria monocytogenes* . *J. Appl. Bacteriology* ; 32:381-394.

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7. Monk, J.D, et al. 1987. Irradiation inactivation of *Listeria monocytogenes* and *Staphylococcus aureus* in low and high-fat frozen and refrigerated ground beef. *J. Food Prot* ., 57:769-772.

8. Murray, E.G, et al. 1926. A disease of rabbits characterized by large mononuclear leucocytosis caused by ahitherto undescribed bacillus *Bacterium monocytogenes* . *J. Path. Bact.* ; 19:407-439.

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10. Vanderzant, C. and D.F. Splittstoesser, (ed.). 1992. *Compendium of Methods for the Microbiological Examination of Foods*, 3rd ed. APHA, Washington, D.C.

11. Wehr, H.M. 1987. *Listeria monocytogenes* - a current dilemma Special Report. *J. Assoc. Anal . Chem.* ; 80:769-7762.

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The Hardy Diagnostics manufacturing facility and quality management system is certified to ISO 13485.

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