

9880 LESS Plus

Intended Use: For the selective enrichment of *Listeria* spp.

Product Summary and Explanation

Listeria monocytogenes, described first in 1926 by Murray, Webb and Swann, is an extensive problem in public health and food industries.¹ This organism has the ability to cause human illness and death, particularly in immunocompromised individuals and pregnant women.² Epidemiological evidence from outbreaks of listeriosis indicates the principle route of transmission is via the consumption of foods contaminated with *Listeria monocytogenes*.³ Implicated vehicles of transmission include meat, eggs, chicken, vegetables, and dairy products.⁴ *Listeria* spp. are ubiquitous in nature, present in a wide range of unprocessed foods and in soil, sewage, and river water.⁵ Certain strains of *Listeria* spp. are able to survive the manufacturing and ripening processes in dairy products.

Listeria spp. grow over a pH range of 5.0 – 9.6, and survive in food products with pH level outside these parameters.¹⁰ *Listeria* spp. are microaerophilic, Gram-positive, asporogenous, non-encapsulated, non-branching, short, motile rods. Motility is pronounced at 20°C for *Listeria*.

Principles of the Procedure

The LESS Plus medium is used for the rapid recovery of *Listeria* species in foods and environmental samples, allowing detection and presumptive identification of the test organism in 24 to 26 hours.

Precautions

1. For Laboratory Use.
2. IRRITANT. Irritating to eyes, skin, and respiratory system.

Sample Preparation and Enrichment

Medium Preparation

Dissolve 44 grams of the medium into one liter of purified water. Heat the solution with frequent agitation to completely dissolve the medium if necessary. **Autoclave at 110°C for 15 minutes.**

Meat samples

1. Weigh out 125 grams of sample in a stomacher-type bag.
2. Add 375 ml of **LESS Plus** medium which has been pre-warmed to 36°C to the bag. Homogenize (Stomacher, etc.) the sample as appropriate for the sample type.
3. Incubate the sample at 36 ± 1°C for 24 to 26 hours.

Environmental samples

1. Place the sponge or swab sample in a stomacher-type bag or in a tube.
2. Add the appropriate amount of **LESS Plus** medium which has been pre-warmed to 36°C to the bag. For sponge samples, an appropriate amount is typically 100-200 ml. For swab samples, the amount is 10 ml.
3. Incubate the sample at 36 ± 1°C for 24 to 26 hours.

For all other food samples

1. Weigh out 25 grams of sample in a stomacher-type bag.
2. Add 225 ml of **LESS Plus** medium which has been pre-warmed to 36°C to the bag. Homogenize (Stomacher, etc.) the sample as appropriate for the sample type.
3. Incubate the sample at 36 ± 1°C for 24 to 26 hours.

Quality Control Specifications

Dehydrated

Appearance: Medium should be light to medium beige in color, free-flowing and homogenous.

9880 LESS Plus

Prepared

Appearance (110°C autoclave cycle): Yellow to yellow-brown, clear to slightly hazy with none to trace precipitate

pH: pH of re-hydrated medium should be 7.2 ± 0.2 .

Performance Provides for the detection of *Listeria* spp.. Strains tested: ATCC 33090, ATCC 19119, ATCC 35967, ATCC 15313, ATCC 7644 inoculated at 10 - 100 CFU.

Test Procedure

Refer to the applicable *Listeria* Test System package insert under the section for the respective test procedure.

Results

Refer to the applicable *Listeria* Test System package insert under the section Interpretation of Results for a complete discussion of the test results.

Storage

Store sealed bottle containing the dehydrated medium at 2 - 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping the container tightly closed.

Expiration

Refer to expiration date stamped on products. The dehydrated medium should be discarded if not free flowing, or if appearance has changed from the original color. Expiry applies to products in their intact package when stored as directed.

Limitations of the Procedure

1. Refer to the applicable test system package insert.
2. Identification of *Listeria monocytogenes* must be confirmed by biochemical and serological testing.¹¹
3. Use re-hydrated medium within the same day as prepared.
4. Incubation times other than those specified may lead to erroneous results.
5. Sample bags must be closed loosely to allow air exchange during incubation, which is vital for organism growth.
6. Do not use expired medium.

Packaging

LESS Plus Medium 9880A 500 grams

References

1. Murray, E. G. D., R. A. Webb, and M. B. R. Swann. 1926. A disease of rabbits characterized by large mononuclear leucocytosis caused by a hitherto undescribed bacillus *Bacterium monocytogenes*. J. Path. Bact. **29**:407-439.
2. Monk, J. D., R. S. Clavero, L. R. Beuchat, M. P. Doyle, and R. E. Brackett. 1994. Irradiation inactivation of *Listeria monocytogenes* and *Staphylococcus aureus* in low and high fat, frozen refrigerated ground beef. J. Food Prot. **57**:969-974.
3. Bremer, P. J., and C. M. Osborne. 1995. Thermal-death times of *Listeria monocytogenes* in green shell mussels prepared for hot smoking. J. Food Prot. **58**:604-608.
4. Grau, F. H., and P. B. Vanderlinde. 1992. Occurrence, numbers, and growth of *Listeria monocytogenes* on some vacuum-packaged processed meats. J. Food Prot. **55**:4-7.
5. Patel, J. R., C. A. Hwang, L. R. Beuchat, M. P. Doyle, and R. E. Brackett. 1995. Comparison of oxygen scavengers for their ability to enhance resuscitation of heat-injured *Listeria monocytogenes*. J. Food Prot. **58**:244-250.
6. Fraser, J., and W. Sperber. 1988. Rapid detection of *Listeria* in food and environmental samples by esculin hydrolysis. J. Food Prot. **51**:762-765.
7. Vanderzant, C., and D. F. Splittstoesser (eds.). Compendium of methods for the microbiological examination of foods, 3rd ed. American Public Health Association, Washington, D.C.
8. Marshall, R. T. (ed.). Standard methods for the examination of dairy products 16th ed. American Public Health Association, Washington D.C.
9. www.fda.gov/Food/ScienceResearch/LaboratoryMethods/BacteriologicalAnalyticalmanualBAM/default.htm.
10. United States Department of Agriculture, Food Safety and Inspection Service. (2011). Microbiology Laboratory Guidebook, Laboratory Quality Assurance Division, Athens, GA.

9880 LESS Plus

11. Murray, P. R., E. J. Baron, M. A. Pfaller, F. C. Tenover, and R. H. Tenover (eds.). Manual of clinical microbiology, 6th ed. American Society for Microbiology, Washington, D.C.

Technical Information

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