

PALCAM BROTH (7670)

Intended Use

PALCAM Broth is used with supplements for the selective enrichment of *Listeria* spp. from foods and environmental samples.

Product Summary and Explanation

Listeria monocytogenes, described in 1926 by Murray, Webb, and Swann, is a widespread problem in public health and food industries.¹ This organism has the ability to cause human illness and death, particularly in immunocompromised individuals and susceptible pregnant women.² Epidemiological evidence from outbreaks of listeriosis indicate the principle route of transmission is via the consumption of foodstuffs contaminated with *Listeria monocytogenes*.³ Implicated vehicles of transmission include turkey frankfurters, coleslaw, pasteurized milk, Mexican style cheese, and pate.⁴

PALCAM Broth is based on the formulation of Van Netten et al.,⁵ while he was investigating the isolation of *Listeria* spp. from food samples. PALCAM Broth has been used for testing *Listeria* from raw milk.⁶

Principles of the Procedure

Peptone and Yeast Extract provide nitrogen, vitamins, minerals, and cofactors required for growth of *Listeria* spp. The addition of Lithium Chloride, Polymixin B, Acriflavine, and Ceftazidime suppress the growth of Gram-negative bacteria and most Gram-positive bacteria. Lithium Chloride provides selectivity to PALCAM Broth due to the high salt tolerance of *Listeria* spp. Lecithin inactivates quaternary ammonium. Polysorbate 80 neutralizes phenols, hexachlorophene, and formalin. *L. monocytogenes* hydrolyses Esculin to esculetin and glucose. Esculetin form an olive-green to black complex with iron (III) ions, and PALCAM Broth turns brown-black if *Listeria* is present. Mannitol fermenting species of *Listeria* give the broth a yellow color due to the pH indicator Phenol red.

Formula / Liter

Peptone.....	23 g
Yeast Extract.....	5 g
Lithium Chloride.....	10 g
Ferric Ammonium Citrate.....	0.5 g
Esculin	0.8 g
Mannitol	5 g
Phenol Red	0.08 g
Lecithin.....	1.0 g
Polysorbate 80	2 g

Final pH: 7.4 ± 0.2 at 25°C

Formula may be adjusted and/or supplemented as required to meet performance specifications.

PALCAM Supplement / 5 mL (7987)

Polymixin B, 5 mg
Acriflavin, 2.5 mg
Ceftazidime, 10 mg
(Each vial supplements 500 mL)

Precautions

1. For Laboratory Use.
2. Harmful. Harmful if swallowed, inhaled, or absorbed through the skin. Irritating to eyes, respiratory system, and skin. Skin irritation may be severe. May cause central nervous system effects.

Directions

1. Suspend 47 g of the medium in one liter of purified water.
2. Heat with frequent agitation to completely dissolve the medium.
3. Autoclave at 121°C for 15 minutes. Cool to 45 - 50°C.
4. Aseptically add 10 mL of PALCAM Supplement (# 7987). One vial of PALCAM Supplement will supplement 500 mL of PALCAM Broth. Dispense into sterile tubes.

Quality Control Specifications

Dehydrated Appearance: Powder is homogeneous, soft lumpy, and beige to tan with a faint pink to orange tint.

Prepared Appearance: Prepared medium contains a slight haze to haze and red.

Expected Cultural Response: Cultural response in PALCAM Broth prepared with PALCAM Supplement (# 7987) incubated aerobically at $30 \pm 2^\circ\text{C}$ and examined for growth after 24 - 48 hours.

Microorganism	Approx. Inoculum (CFU)	Results
<i>Escherichia coli</i> ATCC® 25922	$\sim 10^3$	Inhibited
<i>Enterococcus faecalis</i> ATCC® 29212	$\sim 10^3$	Marked to complete inhibition
<i>Staphylococcus aureus</i> ATCC® 25923	$\sim 10^3$	Marked to complete inhibition
<i>Listeria monocytogenes</i> ATCC® 7644	10 - 300	Brown-black
<i>Listeria monocytogenes</i> ATCC® 19112	10 - 300	Brown-black
<i>Listeria monocytogenes</i> ATCC® 15313	10 - 300	Brown-black

The organisms listed are the minimum that should be used for quality control testing.

Test Procedure

To isolate *Listeria* from food and environmental samples using PALCAM Broth, the following procedure is recommended by the Canadian Health Protection Branch⁷ and FDA/BAM Method.⁸

1. On PALCAM Agar Base, *Listeria* spp form grey-green colonies with a black halo. Some *Enterococcus* and *Staphylococcus* strains form grey colonies with a brown-green halo or yellow colonies with a yellow halo.
2. Confirm the identity of each presumptive *Listeria* spp. through biochemical and / or serological testing.

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.

Expiration

Refer to expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if appearance has changed from the original color. Expiry applies to medium in its intact container.

Limitations of the Procedure

Due to nutritional variation, some strains may grow poorly or fail to grow on this medium.

Packaging

PALCAM Broth	Code No.	7670A	500 g
		7670B	2 kg
		7670C	10 kg
PALCAM Supplement, 5 mL		7987	10 vials/pkgl

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. Bacteriol. **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 and 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. Van Netten, P., I. Perales, A. Van de moosalijk, G. D. W. Curtis, and D. A. A. Mossel. 1989. Liquid and solid selective differential media for the detection and enumeration of *L. monocytogenes* and other *Listeria* spp. Int. J. of Food Microbiol. **8**:299-317.

6. **Lund, A.M.** 1991. Comparison of methods for isolation of *Listeria* from raw milk. J. Food Prot. 54:602-608.
7. **Canadian Food Directorate.** 2001. The Compendium of analytical methods. Polyscience Publications, Laval, Quebec, Canada.
8. **www.fda.gov/Food/ScienceResearch/LaboratoryMethods/BacteriologicalAnalyticalManualBAM/default.htm**

Technical Information

Contact Acumedia Manufacturers, Inc. for Technical Service or questions involving dehydrated culture media preparation or performance at (517)372-9200 or fax us at (517)372-2006.