



Listeria Enrichment Broth, Modified

M888

Intended Use:

Recommended for selective enrichment of *Listeria* species.

Composition**

Ingredients	Gms / Litre
Tryptose	10.000
Yeast extract	5.000
HM peptone B #	5.000
Sodium chloride	20.000
Disodium hydrogen phosphate	9.600
Potassium dihydrogen phosphate	1.350
Esculin	1.000
Nalidixic acid	0.020
Acriflavin hydrochloride (Trypaflavin)	0.012
Final pH (at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters

Equivalent to Beef extract

Directions

Suspend 51.98 grams in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense in tubes and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C.

Principle And Interpretation

Only *Listeria monocytogenes* among the *Listeria* species is reported to cause infection in humans. In human adults, *L.monocytogenes* primarily causes meningitis, encephalitis or septicemia. The tropism of *L.monocytogenes* for the central nervous system leads to severe disease, often with high mortality or with neurologic disorders among survivors (8). Listeria Enrichment Broth, Modified, a modification of the original formulation of Donnelly and Baigent, is used for the selective enrichment of *Listeria* species (2). In this medium, the nalidixic acid concentration has been reduced from 40 mg/l in the original composition, to 20 mg/l. Listeria Enrichment Broth, Modified is used for selective enrichment of *Listeria* species from milk, milk products and other foods.

This medium contains tryptose, yeast extract and HM peptone B which provide essential nutrients like carbon and nitrogenous compounds including vitamins, amino acids and trace ingredients. Phosphates provide buffering action to the medium while sodium chloride maintains osmotic equilibrium. Nalidixic acid and acriflavin inhibit the growth of gram-negative and gram-positive organisms respectively (5,6,7) except *Listeria* species.

For enrichment, 25 gram or 25 ml sample is added to 225 ml medium in a stomacher bag. Homogenize the material if required. Incubation is carried out at 30°C for upto 7 days and the sample is subcultured on Listeria Selective Agar (M567) after 1, 2 and 7 days.

Type of specimen

Food and dairy samples.

Specimen Collection and Handling

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (1,9,10). After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Further biochemical identification of organisms is required for confirmation.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Yellow coloured, clear to slightly opalescent solution having a bluish tinge

Reaction

Reaction of 5.2% w/v aqueous solution at 25°C. pH : 7.2±0.2

pH

7.00-7.40

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 24-48 hours.

Organism	Inoculum (CFU)	Growth
<i>Escherichia coli</i> ATCC 25922 (00013*)	≥10 ⁴	inhibited
<i>Listeria monocytogenes</i> subsp. <i>serovar 1</i> ATCC 19111 (00020*)	50-100	luxuriant
<i>Listeria monocytogenes</i> ATCC 19112	50-100	luxuriant
<i>Listeria monocytogenes</i> ATCC 19117	50-100	luxuriant
<i>Listeria monocytogenes</i> ATCC 19118	50-100	luxuriant
<i>Staphylococcus aureus</i> subsp. <i>aureus</i> ATCC 25923 (00034*)	≥10 ⁴	inhibited

Key : * - Corresponding WDCM numbers

Storage and Shelf Life

Store dehydrated and the prepared medium at 2-8° C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (3,4).

Reference

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Revision :03/2022

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