McBride Listeria HiVeg™ Agar Base

MV386

Intended Use:

Recommended for selective isolation and cultivation of Listeria species from various samples.

Composition**

Ingredients	g / L
HiVeg™ hydrolysate No. 1	10.000
HiVeg™ extract	3.000
Sodium chloride	5.000
Glycine anhydride	10.000
Lithium chloride	0.500
Phenyl ethanol	2.500
Agar	15.000
Final pH (at 25°C)	7.3±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 46.0 grams in 1000 ml purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool below 45- 50°C. Before gelling, aseptically add sterile defibrinated blood to a final concentration of 5% and add filter sterilized McBride Selective Supplement (FD070). Mix well and pour into sterile Petri plates.

Principle And Interpretation

The disease listeriosis is a frequent cause of abortions in cattle and sheep. In human, symptoms are manifested as septicemia, encephalitis and circulatory monocytosis (1). *Listeria* multiplies over a wide range of temperatures, from 3°C to 45°C, and over a pH range of 5.0 to 9.6. It also survives in food products with pH levels outside these parameters (2). Because of these properties, *Listeria* survives the various food processing techniques (3). McBride Listeria Agar (4), recommended by APHA (5) is used for isolating Listeria from clinical specimens and foodstuffs including raw milk (6). This medium helps in the detection of low numbers of L. monocytogenes present in food samples. McBride Listeria HiVeg™ Agar Base is prepared by using vegetable peptones in place of animal based peptones which make the media free of BSE/TSE risks.

HiVeg™ hydrolysate No. 1 and HiVeg™ extract in the medium supply nitrogen, carbon, sulphur and trace nutrients required for the growth of *Listeria*. Phenyl ethyl alcohol is bacteriostatic for gram-negative bacteria as it selectively inhibits DNA synthesis (7). Sodium chloride maintains the osmotic balance of the medium. Glycine inhibits certain gram-negative and gram-positive bacteria including *Escherichia coli* and *Enterococcus faecalis*, the common accompanying contaminants. Lithium chloride also has antibacterial activity. Further selectivity is achieved by the addition of McBride Listeria Supplement (FD070). The detection of L. monocytogenes is greatly improved by pre-enrichment in liquid media either by one step or two steps. In one step method (8), infected material is inoculated directly in Listeria Selective HiVeg™ Broth Base (MV889), while in two steps method (9), infected material is inoculated in Listeria Enrichment HiVeg™ Medium Base (UVM) (MV890A) and incubated at refrigeration temperature of 4°C for few weeks (cold enrichment), as the organism has the ability to grow in low temperature. It is then inoculated in Fraser Secondary Enrichment HiVeg™ Broth Base (MV1083), followed by plating onto selective agar such as McBride Listeria Agar. The presumptive *Listeria* colonies are selected under 45° transillumination. Listeria colonies are dense white to iridescent white appearing as crushed glass. Small colonies tend to be blue, while non-Listeria show yellowish orange colonies that are further purified. McBride Listeria Agar can be used as a plating medium with or without supplementation of blood

Type of specimen

Dairy samples

Specimen Collection and Handling:

For dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (6). 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 tests must be carried out 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

Gelling

Firm,comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Basal medium :Light amber coloured clear to very slightly opalescent gel. After addition of 5%v/v sterile blood : Cherry red opaque gel forms in Petri plates

Reaction

Reaction of 4.6% w/v aqueous solution at 25°C. pH : 7.3±0.2

pH

7.10-7.50

Cultural Response

Cultural characteristics observed in anaerobic atmosphere with added McBride Listeria Supplement(FD070) and 5%v/v sterile defibrinated blood, after an incubation at 35-37°C for 24-48 hours.

Organism	Inoculum (CFU)	Growth w/ FD070	Recovery w/ FD070	Growth w/ blood and FD070	Recovery w/ blood and FD070
Listeria monocytogenes ATCC 19112	50-100	good-luxuriant	>=50%	good-luxuriant	>=50%
Escherichia coli ATCC 25922 (00013*)	50-100	none-poor	<=10%	none-poor	<=10%
Pseudomonas aeruginosa ATCC 27853(00025*)	50-100	none-poor	<=10%	none-poor	<=10%
Enterococcus faecalis ATCC 29212 (00087*)	50-100	none-poor	<=10%	none-poor	<=10%

* Corresponding WDCM Numbers

Storage and Shelf Life

Store between 10-30°C in a tightly closed container 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 clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (10,11).

Reference

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