



M2032

Lowenstein Jensen Medium Base, Modified

Intended use

Lowenstein Jensen Medium (L. J. Medium) is used for the isolation of *Mycobacterium* species from mixed flora.

Composition**

Ingredients	Gms / 600 ml
L-Asparagine	3.600
Monopotassium phosphate	2.500
Magnesium sulphate	0.240
Sodium citrate	0.600
Potato Flour	30.000
Malachite green	0.400

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 37.24 grams in 600 ml distilled water containing 12 ml glycerol (for bovine bacteria or other glycerophobic organisms additions of glycerol is not desirable). Heat if necessary to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Meanwhile prepare 1000 ml of whole egg emulsion collected aseptically. Aseptically add and mix egg emulsion base and LCN Supplement (FD338) gently to obtain uniform mixture. Distribute in sterile screw capped tubes. Arrange tubes in a slanted position. Coagulate and inspissate the medium in an inspissator water bath or autoclave at 85°C for 45 minutes.

Principle And Interpretation

Solid media used for isolation and cultivation of Mycobacteria are either egg-based or agar-based. Egg-based media contain whole eggs or egg yolk, potato flour, salts and glycerol and are solidified by inspissation. Of the egg-based media, Lowenstein Jensen Medium is most commonly used (1). L.J. Medium was originally formulated by Lowenstein, containing congo red and malachite green dyes (2). Jensen (3) modified Lowensteins medium by altering the citrate and phosphate contents, eliminating the congo red dye and by increasing the malachite green concentration. This medium supports the growth of a wide variety of Mycobacteria and can also be used for niacin testing (6).

Lincomycin, Cycloheximide and Nalidixic acid along with malachite green prevents growth of the majority of contaminants surviving decontamination of the specimen while encouraging earliest possible growth of Mycobacteria. Do not add glycerol to the medium if bovine or other glycerophobic strains are to be cultured (7). Malachite green serves as an inhibitor and also as pH indicator. Formation of blue zone indicates a decrease in pH by gram-positive contaminants and yellow zones of dye destruction by gram-negative bacilli. Proteolytic contaminants cause localized or complete digestion of medium. LCN Supplement contains cycloheximide, lincomycin and nalidixic acid. Cyclohemide suppresses the growth of saprophytic organisms, Lincomycin inhibits gram positive organisms while nalidixic acid inhibits gram negative organisms in clinical samples. Refer appropriate references for standard test procedures of decontamination and isolation (1, 4,8-9).

Type of specimen

Clinical samples - Sputum sample

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (1, 4,5,8-9) Warning and Precautions :

In Vitro diagnostic Use only. 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 clincal specimens. Safety guidelines may be referred in individual safety data sheets

Limitations :

Further biochemical and serological tests must be carried out for further identification.

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

Greenish blue to peacock blue homogeneous

Gelling

free flowing powder

Colour and Clarity of prepared medium

The mixture of sterile basal medium and whole egg emulsion, when inspissated, coagulates to yield pale bluish green coloured, opaque smooth slants

Cultural Response

M2032: Cultural characteristics observed in presence of 5-10% Carbon dioxide, with added egg emulsion base, after an incubation at 35-37°C for 2-4 weeks.

Organism	Growth	Growth with Gruft Supplement (FD053)	Colony Characteristic
Cultural Response			
Mycobacterium avium ATC 25291	Cluxuriant	good-luxuriant	smooth, non- pigmented colonies
Mycobacterium gordonae ATCC 14470	luxuriant	good-luxuriant	smooth, yellow, orange colonies
Mycobacterium kansasii ATCC 12478	luxuriant	good-luxuriant	photochromogenic, smooth to rough
Mycobacterium smegmatis ATCC 14468	luxuriant	good-luxuriant	wrinkled,creamy white colonies
<i>M. tuberculosis H37RV ATCC 25618</i>	luxuriant	good-luxuriant	granular, rough, warty, dry friable colonies

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 inorder 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 (4,5).

References

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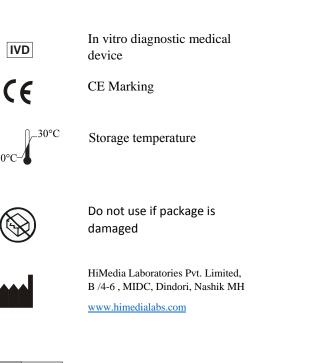
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