Lowenstein Jensen Medium Base, Modified

M2032

Intended use:

Recommended for isolation and cultivation of *Mycobacterium* species.

Composition**

Ingredients	g / 600 ml
L-Asparagine	3.600
Potassium dihydrogen 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 37.34 grams in 600 ml distilled water containing 12 ml glycerol (for other glycerophobic organism's 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 1000 ml egg emulsion to sterile medium base. Also aseptically add rehydrated contents of one vial of LCN Supplement (FD338). Mix gently to obtain uniform mixture. Distribute 8ml 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 (4).

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 (5). 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 (1,6,7,8).

Type of specimen

Clinical samples - Sputum sample

Specimen Collection and Handling:

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (1,6,7,8,9).
After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

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

Limitations :

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

2.Individual organisms differ in their growth requirement and may show variable growth patterns on the medium.

3.Each lot of the medium has been tested for the organisms specified on the COA. It is recommended to users to validate the medium for any specific microorganism other than mentioned in the COA based on the user's unique requirement.

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 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

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

Organism	Growth	Growth w/LCN Supplement (FD338)	Colony Characteristic
Mycobacterium avium ATCC 25291	luxuriant	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
M. tuberculosis H37RV ATCC 25618	luxuriant	good-luxuriant	granular, rough, warty, dry friable colonies
Staphylococcus aureus subsp. aureus ATCC 25923 (00034*)	luxuriant	Inhibition	

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 (8,9).

References

1. Murray P. R., Baron E. J., Jorgensen J. H., Pfaller M. A., Yolken R. H., (Eds.), 8th Ed., 2003, Manual of Clinical Microbiology, ASM, Washington, D.C.

2. Lowenstein E., 1931, Zentralbl. Bakteriol. Parasitenkd. Infektionskr. Hyg. Abt. 1 Orig., 120:127.

3. Jensen K. A., 1932, Zentralb. Bakteriol. Parasitenkd. Infektionskr. Hyg. Abt. I Orig., 125:222.

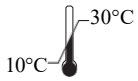
4. Boisvert H., 1960, Ann. Inst. Pasteur, 99:600.

5.MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore.
6.Cernoch P., Enns R., Saubolle M. and Wallace R., 1994, Cumitech, 16A, Laboratory Diagnosis of the Mycobacterioses coord , Ed., Weissfeld , ASM, Washington, D. C.
7.Forbes B. A., Sahm A. S. and Weissfeld D. F., Bailey & Scotts Diagnostic Microbiology, 10th Ed., 1998, Mosby, Inc.,St. Louis, Mo.
8.Isenberg, (Ed.), 1992, Clinical Microbiology Procedures Handbook, Vol. I, ASM, Washington, D. C.
9.Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Revision : 03/ 2024

HiMedia Laboratories Pvt. Limited,
Plot No.C-40, Road No.21Y,
MIDC, Wagle Industrial Area,
Thane (W) -400604, MS, India

IVD *In vitro* diagnostic medical device



Storage temperature

EC REP CEpartner4U, Esdoornlaan 13,
3951DB Maarn, NL
www.cepartner4u.eu

CE **CE Marking**

Do not use if package is damaged

Disclaimer :

User must ensure suitability of the product(s) in their application prior to use. Products conform solely to the information contained in this and other related HiMedia™ publications. The information contained in this publication is based on our research and development work and is to the best of our knowledge true and accurate. HiMedia™ Laboratories Pvt Ltd reserves the right to make changes to specifications and information related to the products at any time. Products are not intended for human or animal or therapeutic use but for laboratory,diagnostic, research or further manufacturing use only, unless otherwise specified. Statements contained herein should not be considered as a warranty of any kind, expressed or implied, and no liability is accepted for infringement of any patents.