

LOWENSTEIN JENSEN MEDIUM

Ready to use tubes

INTENDED USE

Ready to use tubes for the isolation of *Mycobacteria* spp.

TYPICAL FORMULA (g/l)

Magnesium Sulphate	0.24
Magnesium Citrate	0.60
Monopotassium Phosphate	2.50
L-Asparagine	3.60
Potato Flour	30.00
Malachite Green	0.40
Glycerol	12ml
Distilled water	600 ml
Homogenized whole eggs	1000ml

DESCRIPTION

Lowenstein Jensen Medium Base is used for the cultivation of mycobacteria, especially *Mycobacterium tuberculosis*, from both clinical specimens and pure culture. The medium is prepared according to a Jensen modification of the original Lowenstein medium, and contains malachite green as a selective agent. When heated, the egg albumin coagulates thus providing a solid surface for inoculation. Nitrogen, fatty acids and proteins are supplied by egg and asparagine. Glycerol serves as a carbon source and is favourable to growth of human type tubercle bacillus while being unfavourable to the bovine type.

TECHNIQUE

Inoculate the tubes with specimen after decontamination and neutralisation, in duplicate, streaking the material to be examined onto the surface of the medium.

Incubate in a CO₂ atmosphere at 37°C. Protect from light. Tubed media should be incubated for one week with loosened caps to allow circulation of CO₂ for the initiation of growth. Caps should be tightened after one week in order to prevent dehydration of media.

Examine the tubes within 5-7 days and weekly for up to 8 weeks. Examine tubes under light and magnifying mirror for macroscopic growth.

Mycobacterium tuberculosis grows on the medium in large dry yellowish colonies.

Examine and record each type of colony morphology, pigment and growth rate. Biochemical testing is required for definitive identification.

LIMITATIONS

L.J. Medium requires incubation in a 5-10% CO₂ atmosphere; mycobacteria are not recovered from candle extinction jar.

Because malachite green is photo-sensitive, protect L.J. tubes from all sources of light.

M.bovis will not growth on L.J. Medium because of the presence of glycerol.

The tubed medium is light green in colour with occasional areas of light yellow particles of egg yolk lipid. These particles should not be mistaken for either mycobacteria or contaminated colonies. However, any changes of colour showing bright yellow or deep blue coloured zones around yolk particles are sign of contamination.

Formation of a blue zone during incubation indicates a decrease in pH by Gram-positive contaminants (e.g. streptococci) and yellow zones of dye destruction by Gram negative bacilli. Proteolytic contaminants cause localized or complete digestion of the medium. These areas of contamination are usually progressive and medium should be discarded aseptically.

Since moisture is an essential growth requirement by mycobacteria it is important that media be kept moist prior to inoculation by keeping tubes tightly sealed.

STORAGE

Store at 2-8° away from direct light - When stored as directed the tubed media remain stable until the expiry date shown on the label. Do not use beyond stated expiry date. Media should not be used if there are any signs of deterioration (shrinking, cracking, discoloration) or contamination.

PRECAUTIONS

For *in vitro* diagnostic use only. Observe approved biohazard precautions and aseptic techniques. To be used only by adequately trained and qualified laboratory personnel. Sterilize all biohazard waste before disposal.

REFERENCES

- APHA (1963) - Diagnostic Procedure and Reagents. 4th edition.
- Canetti, G. et al (1963). *Bull. Wld. Hlth. Org.*, **29**, 565-578.
- Canetti, et al (1969) *Bull. Wld. Hlth. Org.*, **41**, 21-43.
- Sommers, M.H. and J.K. Mc Clatcky (1983) - Laboratory Diagnosis of the *Mycobacteriaceae*. Cumitech 16, ASM, Washington, D.C.

PACKAGING**551635****Lowenstein-Jensen Medium Base 20 ready to use slanted tubes**