TM 375 – LOWENSTEIN JENSEN MEDIUM BASE (L. J. MEDIUM)

INTENDED USE
For isolation and cultivation of *Mycobacterium* species.

PRODUCT SUMMARY AND EXPLANATION
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.
L.J. Medium was originally formulated by Lowenstein, containing congo red and malachite green dyes. Jensen modified Lowensteins medium by altering the citrate and phosphate contents, eliminating the congo red dye and by increasing the malachite green concentration. Gruft further modified L. J. Medium with the addition of two antimicrobics to increase selectivity. This medium supports the growth of a wide variety of Mycobacteria and can also be used for niacin testing.

COMPOSITION

Ingredients	Gms / Ltr
L-Asparagine	3.600
Potassium dihydrogen phosphate	2.400
Magnesium sulphate	0.240
Magnesium citrate	0.600
Potato starch, soluble	30.000
Malachite green	0.400

PRINCIPLE
This medium contains Penicillin and Nalidixic acid which along with malachite green prevents growth of the majority of contaminants surviving decontamination of the specimen while encouraging earliest possible growth of Mycobacteria. RNA acts as stimulant and help to increase the isolation rate of Mycobacteria. Do not add glycerol to the medium if bovine or other glycerophobic strains are to be cultured. Malachite green serves as an inhibitor and also as pH indicator. Formation of blue zone 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 medium.

INSTRUCTION FOR USE
- Dissolve 37.24 grams in 600 ml purified / 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 psi pressure (121°C) for 15 minutes. Cool to 45-50°C.
- Meanwhile prepare 1000 ml of whole egg emulsion collected aseptically. Aseptically add and mix egg emulsion base and Gruft Mycobacterial Supplement (if desired) 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.

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder : Greenish blue to peacock blue homogeneous free flowing powder.

Appearance 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.

pH (at 25°C) : 7.0 ± 0.2

INTERPRETATION

Cultural characteristics observed in presence of 5-10% CO_2, with added egg emulsion base after incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Growth with Gruft Supplement	Colony Characteristic	Incubation Temperature	Incubation Period
Mycobacterium avium	25291	50-100	Luxuriant	Good-luxuriant	Smooth, non-pigmented colonies	35-37°C	2-4 Weeks
Mycobacterium gordonae	14470	50-100	Luxuriant	Good-luxuriant	Smooth, yellow, orange colonies	35-37°C	2-4 Weeks
Mycobacterium kansasii	12478	50-100	Luxuriant	Good-luxuriant	Photochromogenic, smooth to rough	35-37°C	2-4 Weeks
Mycobacterium smegmatis	14468	50-100	Luxuriant	Good-luxuriant	Wrinkled, creamy white colonies	35-37°C	2-4 Weeks
M. tuberculosis H37RV	25618	50-100	Luxuriant	Good-luxuriant	Granular, rough, warty, dry friable colonies	35-37°C	2-4 Weeks

PACKAGING:

In pack size of 100 gm and 500 gm bottles.

STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

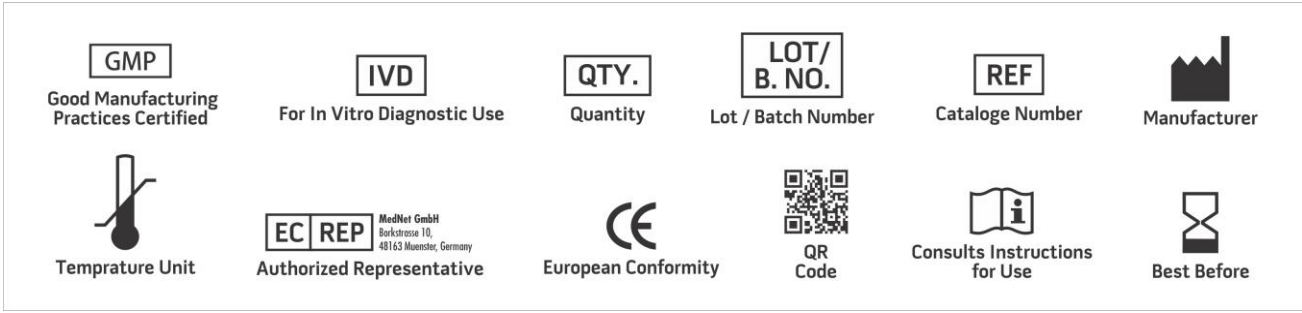
DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

REFERENCES

1. A.V. Hardy, et al, Am. J. Publ. Hlth. 48(1), 754 (1958)
2. Boisvert H., 1960, Ann. Inst. Pasteur, 99:600.

3. Gruft, 1971, Health Lab. Sci., 8:79.
4. Gruft, 1963, Am. Rev. Respir. Dis., 88:412.
5. Jensen K. A., 1932, Zentralb. Bakteriol. Parasitenkd. Infektionskr. Hyg. Abt. I Orig., 125:222.
6. Lowenstein E., 1931, Zentralbl. Bakteriol. Parasitenkd. Infektionskr. Hyg. Abt. 1 Orig., 120:127.
7. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore.



NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.

***For Lab Use Only**

Revision: 08 Nov., 2019