# TM 2168 – L.J. MEDIUM MODIFIED

## INTENDED USE
Used for the isolation of Mycobacterium species from mixed flora.

## 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. 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 |
| **Sodium citrate** | 0.600 |
| **Potato flour** | 30.000 |
| **Malachite green** | 0.400 |

## PRINCIPLE
This medium contains Malachite green which serves as an inhibitor and also as pH indicator. 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. 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.

## 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.
- Meanwhile prepare 1000 ml of whole egg emulsion collected aseptically. Aseptically add and mix egg emulsion base and LCN Supplement 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.

**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 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

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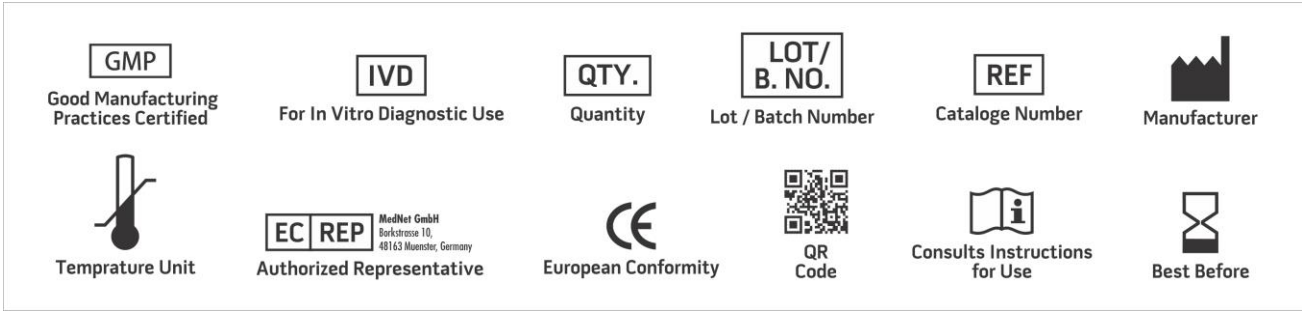


**NOTE:** Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.

***For Lab Use Only**

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