

# MAST REDIPREP® LÖWENSTEIN JENSEN MEDIA

# EM100 & EM102 Series

## Intended use

For the isolation and culture of mycobacteria.

FOR IN VITRO DIAGNOSTIC USE ONLY

## Contents

MAST REDIPREP<sup>®</sup> Löwenstein Jensen Media are supplied as packs of 50 bottles. Glycerol containing medium (EM100) has a red coloured cap label. Pyruvate containing medium (EM102) has a green coloured cap label.

## **Formulation\***

Slopes of Löwenstein Jensen (LJ) Medium containing glycerol or sodium pyruvate.

## Storage and shelf life

All containers should be kept tightly closed and stored in a dry place at 2 to 8°C until the expiry date shown on the pack label.

#### Precautions

Observe approved hazard precautions and aseptic techniques. To be used only by adequately trained and qualified laboratory personnel. Sterilise all biohazard waste before disposal. Refer to Product Safety Data sheet (available on request or via MAST<sup>®</sup> website). All specimens and suspected isolates of *Mycobacterium* species must be processed and handled in a microbiological safety cabinet in a containment level 3 room.

# Materials required but not provided

Standard microbiological supplies and equipment such as loops, MAST<sup>®</sup> selective supplements, swabs, applicator sticks, incinerators and incubators, etc., as well as serological and biochemical reagents and additives such as blood.

# Procedure

#### N-acetyl L-cysteine Method

- Prepare fresh decontamination solution by mixing equal volumes of 4% w/v sodium hydroxide (NaOH) and 0.5% w/v *N*-acetyl-L-cysteine solution (NALC). If a sputum specimen has been pre-treated with Sputagest Selectavial (SV40) or alternative liquefactant omit the NALC.
- 2. Add an equal volume of NaOH-NALC to the specimen in a sterile bottle and agitate.
- 3. Incubate at ambient temperature for between a minimum of 15 and a maximum of 30 minutes (depending on the rate of liquefaction of the specimen) agitating at least every 10 minutes.
- 4. Add 15 mL of phosphate buffer, pH6.8, to neutralise and mix.
- 5. Centrifuge at 3000g for 15 minutes.



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- Decant the supernatant into a discard jar taking care not to contaminate the exterior of the container or lose any of the deposit.
- Inoculate slopes of both MAST REDIPREP<sup>®</sup> LJ Media (EM100 - with Glycerol and EM102 - with Pyruvate) with the deposit. (The deposit can be re-suspended in 1 to 2 mL of sterile distilled water or buffer to aid even inoculation).
- Incubate slopes at 35 to 37°C for up to 8 weeks before discarding (or alternative temperatures according to the methodology followed). Slopes should be examined after 48 to 72 hours to detect gross contamination after which they should be examined weekly.
- 9. Suitable alternative decontamination protocols can be used according to current laboratory practice.

## Interpretation of results

After incubation record growth of organisms. Typical characteristics to note include: colony size, morphology and pigmentation. Typical colonies of *M.tuberculosis* are rough, crumbly, waxy, non pigmented (buff coloured) and slow growing i.e. normally appearing after a minimum of 2 weeks incubation. Colonial morphology of other *Mycobacterium* spp. depends on the species isolated. Diagnoses of Mycobacterial infection should be confirmed by, at minimum, confirmation to *Mycobacterium* genus level based on Ziehl-Nielsen smears from cultures. Acid and Alcohol Fast Bacilli (AAFB) positive isolates should be reported to the appropriate national reference laboratory.

# **Quality control**

Laboratories must be accredited to perform mycobacteriology culture and have Internal Quality Control procedures and show satisfactory performance in an External Quality Assurance proficiency scheme for microscopy, culture, identification, and susceptibility testing. The list below illustrates a performance control strain which the end user can easily obtain.

Test Organism	
<i>Mycobacterium smegmatis</i> ATCC <sup>®</sup> 14468	Growth

#### **Performance evaluation**

MAST REDIPREP<sup>®</sup> Löwenstein Jensen Media have been shown to support the growth of recent clinical isolates of *M. tuberculosis, M. kansasii* and *M. avium-intracellulare.* 

#### Limitations

MAST REDIPREP<sup>®</sup> Löwenstein Jensen Media is subject to colour variation due to photosensitivity. This does not affect performance.

#### References

Bibliography available on request.