



## Luria Bertani Agar, Miller (Miller Luria Bertani Agar)

M1151

### Intended Use:

Recommended for the cultivation and maintenance of recombinant strains of *Escherichia coli* for genetic and molecular biology studies; may be used for routine cultivation and estimation of not particularly fastidious microorganisms.

### Composition\*\*

Ingredients	Gms / Litre
Tryptone	10.000
Yeast extract	5.000
Sodium chloride	10.000
Agar	15.000
Final pH ( at 25°C)	7.5±0.2

\*\*Formula adjusted, standardized to suit performance parameters

### Directions

Suspend 40 grams in 1000 ml purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Dispense as desired. Mix well and pour into sterile Petri plates.

### Principle And Interpretation

Luria Bertani Agar is prepared as described by Lennox (4) for cultivation and maintenance of recombinant strains of *Escherichia coli*. Luria Bertani Agar, Miller (1) is slightly different with double amount of sodium chloride. The media is nutritionally rich for the growth of pure cultures of recombinant strains. Strains derived from *Escherichia coli* K12 are deficient in Vitamin B synthesis are further modified by specific mutation to create auxotrophic strains and are therefore unable to grow on nutritionally deficient media.

Tryptone provides peptides and peptones while Vitamin B complex is provided by yeast extract. Sodium chloride provides sodium ions for membrane transport and also maintains the osmotic equilibrium of the medium.

### Type of specimen

Recombinant strains of *E.coli*

### Specimen Collection and Handling

For Recombinant strain samples follow appropriate techniques for handling specimens as per established guidelines (1,4). After use, contaminated materials must be sterilized by autoclaving before discarding.

### Warning and Precautions

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 specimens. Safety guidelines may be referred in individual safety data sheets.

### Limitations

1. Some strains may show poor growth due to nutritional variations.
2. Further biochemical and serological tests must be carried out for complete identification.

### 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

Cream to yellow homogeneous free flowing powder

#### Gelling

Firm, comparable with 1.5% Agar gel

**Colour and Clarity of prepared medium**

Yellow to amber coloured, clear to slightly opalescent gel forms in Petri plates

**Reaction**

Reaction of 4.0% w/v aqueous solution at 25°C. pH : 7.5±0.2

**pH**

7.30-7.70

**Cultural Response**

Cultural characteristics observed after an incubation at 35-37°C for 18 - 24 hours.

Organism	Inoculum (CFU)	Growth	Recovery
<i>Escherichia coli</i> ATCC 23724	50-100	luxuriant	≥70%
<i>Escherichia coli</i> ATCC 25922 (00013*)	50-100	luxuriant	≥70%
<i>Escherichia coli</i> DH5 alpha MTCC 1652	50-100	luxuriant	≥70%

Key : (\*) Corresponding WDCM numbers

**Storage and Shelf Life**

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order 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 sample must be decontaminated and disposed of in accordance with current laboratory techniques (2,3).

**Reference**

1. Atlas R.M., 1993, Handbook of Microbiological Media, Ed. by Parks L., CRC Press, Inc.
2. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2<sup>nd</sup> Edition.
3. 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.
4. Lennox E.S., 1955, Transduction of Linked Genetic Characters of the host by bacteriophage P1., Virology, 1:190.

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