

# LB Agar, Miller • LB Broth, Miller

## Intended Use

LB Agar, Miller and LB Broth, Miller (Luria-Bertani) are used for maintaining and propagating *Escherichia coli* in molecular microbiology procedures.

## Summary and Explanation

LB Agar, Miller and LB Broth, Miller are based on LB Medium as described by Miller for the growth and maintenance of *E. coli* strains used in molecular microbiology procedures.<sup>1-3</sup> These are nutritionally rich media designed for growth of pure cultures of recombinant strains. *E. coli* grows more rapidly because they provide the cells with amino acids, nucleotide precursors, vitamins and other metabolites that the microorganism would otherwise have to synthesize.<sup>4</sup>

LB Broth, Miller contains twenty times the sodium chloride level of Luria Broth Base, Miller and twice the level found in LB Broth, Lennox.<sup>3,5,6</sup> This allows the researcher to select the optimal salt concentration for a specific strain in LB Agar, Miller.

## Principles of the Procedure

Peptone provides nitrogen and carbon. Vitamins (including B vitamins) and certain trace elements are provided by yeast extract. Sodium ions for transport and osmotic balance are provided by sodium chloride. Agar is the solidifying agent in LB Agar, Miller.

## User Quality Control

### Identity Specifications

#### Difco™ LB Agar, Miller

Dehydrated Appearance:	Light tan, free-flowing, homogeneous.
Solution:	4.0% solution, soluble in purified water upon boiling. Solution is light amber, very slightly to slightly opalescent.
Prepared Appearance:	Very light amber, slightly opalescent.
Reaction of 4.0% Solution at 25°C:	pH 7.0 ± 0.2

#### Difco™ LB Broth, Miller

Dehydrated Appearance:	Off-white to beige, free-flowing, homogeneous.
Solution:	2.5% solution; soluble in purified water. Solution is light amber, clear to very slightly opalescent.
Prepared Appearance:	Very light amber, clear to very slightly opalescent.
Reaction of 2.5% Solution at 25°C:	pH 7.0 ± 0.2

### Cultural Response

#### Difco™ LB Agar, Miller or LB Broth, Miller

Prepare the medium per label directions. Inoculate and incubate at 35 ± 2°C for 18-24 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY
<i>Escherichia coli</i>	33526 (K802)	10 <sup>2</sup> -10 <sup>3</sup>	Good

## Formulae

### Difco™ LB Agar, Miller

Approximate Formula* Per Liter	
Tryptone .....	10.0 g
Yeast Extract .....	5.0 g
Sodium Chloride .....	10.0 g
Agar .....	15.0 g

### Difco™ LB Broth, Miller

Consists of the same ingredients without the agar.

\*Adjusted and/or supplemented as required to meet performance criteria.

## Directions for Preparation from Dehydrated Product

- Suspend/dissolve the powder in 1 L of purified water:  
Difco™ LB Agar, Miller – 40 g;  
Difco™ LB Broth, Miller – 25 g.  
Mix thoroughly.
- Heat the agar medium with frequent agitation and boil for 1 minute to completely dissolve the powder.
- Autoclave at 121°C for 15 minutes.
- Test samples of the finished product for performance using stable, typical control cultures.

## Procedure

Consult appropriate references for recommended test procedures.<sup>3-5</sup>

## Expected Results

Growth should be evident on the agar medium by the appearance of colonies and/or a confluent lawn on the surface of the medium. In the broth medium, growth is evident by the appearance of turbidity.

## References

- Luria and Burrous. 1955. J. Bacteriol. 74:461.
- Luria, Adams and Ting. 1960. Virology 12:348.
- Miller. 1972. Experiments in molecular genetics. Cold Spring Harbor Laboratory, Cold Spring Harbor, N.Y.
- Ausubel, Brent, Kingston, Moore, Seidman, Smith and Struhl (ed.). 1994. Current protocols in molecular biology, vol. 1. Greene Publishing Associates, Inc., Brooklyn, N.Y.
- Sambrook, Fritsch and Maniatis. 1989. Molecular cloning: a laboratory manual, 2nd. ed. Cold Spring Harbor Laboratory, Cold Spring Harbor, N.Y.
- Lennox. 1955. Virology 1:190.

## Availability

### Difco™ LB Agar, Miller

Cat. No.	244520	Dehydrated – 500 g
	244510	Dehydrated – 2 kg

### Difco™ LB Broth, Miller

Cat. No.	244620	Dehydrated – 500 g
	244610	Dehydrated – 2 kg
	214906	Dehydrated – 10 kg

### Europe

Cat. No.	257269	Prepared Bottles, 450 mL – Pkg. of 10
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