

Intended Use

LB Agar, Miller is used in molecular genetic studies.

Product Summary and Explanation

LB Agar, Miller, a nutritionally rich medium designed for growth of pure cultures of recombinant strains, is based on the formulation described by Miller.¹ *E. coli* is grown to late log phase in LB Medium. Some plasmid vectors replicate to a high copy number and do not require selective amplification. Some vectors do not replicate so freely, and need to be selectively amplified. Chloramphenicol may be added to inhibit host synthesis and prevent replication of the bacterial chromosome.²

LB Agar, Miller contains 10 g/L of sodium chloride, different from the levels in Lennox formulations.¹⁻³ This allows the researcher to select the optimal salt concentration for a specific strain. The medium may be aseptically supplemented with glucose.

Principles of the Procedure

The nitrogen, amino acids, and carbon sources are provided by Enzymatic Digest of Casein. Vitamins and certain trace elements are supplied by Yeast Extract. Sodium ions for transport and osmotic balance are provided by Sodium Chloride. Agar is the solidifying agent.

Formula / Liter

Enzymatic Digest of Casein	10 g
Yeast Extract	
Sodium Chloride	
Agar	
Final pH: 7.3 ± 0.2 at 25°C	0

Formula may be adjusted and/or supplemented as required to meet performance specifications.

Precautions

- 1. For Laboratory Use.
- 2. IRRITANT. Irritating to eyes, respiratory system, and skin.

Directions

- 1. Suspend 37 g of the medium in one liter of purified water.
- 2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
- 3. Autoclave at 121°C for 15 minutes.

Quality Control Specifications

Dehydrated Appearance: Powder is homogeneous, free flowing, and beige to yellow beige.

Prepared Appearance: Prepared medium is trace to slightly hazy and yellow-beige.

Expected Cultural Response: Cultural response on LB Agar, Miller incubated aerobically at $35 \pm 2^{\circ}$ C and examined for growth after 18 - 24 hours.

Microorganism	Approx. Inocululm (CFU)	Expected Results
Bacillus subtilis ATCC® 9372	10 - 300	Growth
Escherichia coli ATCC® 25922	10 - 300	Growth

The organisms listed are the minimum that should be used for quality control testing.



Test Procedure

Consult appropriate references for recommended test procedures.^{1,2}

Results

After sufficient incubation the medium should show growth as evidenced by formation of isolate colonies and/or confluent lawn of growth.

Storage

Store sealed bottle containing the dehydrated medium at 2 - 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping container tightly closed.

Expiration

Refer to expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitation of the Procedure

Due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.

<u>Packaging</u> LB Agar, Miller	Code No.	7213A	500 g
		7213B	2 kg
		7213C	10 ka

References

1. Miller, J. H. 1972. Experiments in molecular genetics. Cold Spring Harbor Laboratory. Cold Spring Harbor, New York.

- 2. Sambrook, J., E. F. Fritsch, and T. Maniatis. 1989. Molecular cloning: a laboratory manual, 2nd ed. Cold Spring Harbor Laboratory, Cold Spring Harbor, New York.
- 3. Lennox E. S. 1955. Transduction of linked genetic characters of the host by bacteriophage P1. Virology. 1:190-206.

Technical Information

Contact Acumedia Manufacturers, Inc. for Technical Service or questions involving dehydrated culture media preparation or performance at (517)372-9200 or fax us at (517)372-2006.

