IFU

Instructions for Use

LB AGAR, LENNOX POWDER

Cat. no. C7660	CulGenex™ LB Agar, Lennox, 2L	70gm
Cat. no. C7661	CulGenex™ LB Agar, Lennox	500gm
Cat. no. C7662	CulGenex™ LB Agar, Lennox	2kg
Cat. no. C7663	CulGenex™ LB Agar, Lennox	10kg
Cat. no. C7669	CulGenex™ LB Agar, Lennox, 0.5L	6 pouches/pack

INTENDED USE

Hardy Diagnostics CulGenex™ LB Agar, Lennox is recommended for maintaining and cultivating recombinant strains of *Escherichia coli* for molecular analysis.

This dehydrated culture medium is a raw material intended to be used in the making of prepared media products, which will require further processing, additional ingredients, or supplements.

SUMMARY

LB, or "lysogeny broth", media formulations have been widely used for the cultivation of *Escherichia coli* since the 1950s, and have become an industry standard in molecular microbiology applications for the preparation of plasmid DNA and recombinant strains of *E. coli*.[3,5-8] In general, typical wild type strains of *E. coli* are capable of growth on minimal media. However, researchers have exploited auxotrophic mutations in strains used for molecular manipulations, making them dependent upon specific growth factors; one strain in particular, *E. coli* K-12, is deficient in vitamin B production and is incapable of growth on nutritionally deficient media.

The original recipe for LB medium was formulated by Giuseppe Bertani and published in 1951.[3] LB media have since been adapted by Miller, Lennox and Luria to contain differing concentrations of sodium chloride in order to provide the appropriate osmotic conditions for the strain of interest. LB Agar, Lennox contains half the sodium chloride concentration of LB Agar, Miller and ten times that found in Luria Agar, Miller.[3,5-8] Low salt formulations, such as those adapted by Lennox and Luria, are ideal for salt sensitive applications.

LB Agar, Lennox was adapted by E.S. Lennox in the mid 1950s and is a nutritionally rich medium designed to contain certain trace elements for the growth and maintenance of pure recombinant strains. CulGenex™ LB Agar, Lennox is based on this formulation and contains tryptone and yeast extract for amino acids, vitamins and essential minerals. The moderate amount of sodium chloride provides sodium ions for transport and helps maintain osmotic balance. Agar is the solidifying agent. If desired, glucose can be aseptically added during preparation to complete the medium described

in full by Lennox.

FORMULA*

Gram weight per liter:	35.0gm/L
Tryptone	10.0gm
Sodium Chloride	5.0gm
Yeast Extract	5.0gm
Agar	15.0gm

Final pH 7.0 +/- 0.2 at 25ºC.

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

STORAGE AND SHELF LIFE

Store the sealed bottle(s) containing dehydrated culture medium at 2-30ºC. Dehydrated culture medium is very hygroscopic. Keep lid tightly sealed. Protect dehydrated culture media from moisture and light. The dehydrated culture media should be discarded if it is not free-flowing or if the color has changed from its original light beige.

Store the prepared culture media at 2-8ºC.

The expiration date on the product label applies to the product in its intact packaging when stored as directed. The product may be used and tested up to the expiration date on the product label and incubated for the recommended incubation times as stated below.

Refer to the document "Storage" for more information.

PRECAUTIONS

This product may contain components of animal origin. Certified knowledge of the origin and/or sanitary state of the animals does not guarantee the absence of transmissible pathogenic agents. Therefore, it is recommended that these products be treated as potentially infectious, and handle observing the usual universal blood precautions. Do not ingest, inhale, or allow to come into contact with skin.

This product is for laboratory use only. It is to be used only by adequately trained and qualified laboratory personnel. Observe approved biohazard precautions and aseptic techniques. All laboratory specimens should be considered infectious and handled according to "standard precautions." Refer to the document "Guidelines for Isolation Precautions" from the Centers for Disease Control and Prevention.

For additional information regarding specific precautions for the prevention of the transmission of all infectious agents from laboratory instruments and materials, and for recommendations for the management of exposure to infectious disease, refer to CLSI document M29: *Protection of Laboratory Workers from Occupationally Acquired Infections.*

Sterilize all biohazard waste before disposal.

Refer to the document "Precautions When Using Media" for more information.

METHOD OF PREPARATION FOR DEHYDRATED CULTURE MEDIA

1. Suspend 35.0gm of the dehydrated culture media in one liter of distilled or deionized water. Stir to mix thoroughly.

2. Heat to boiling to dissolve completely.

3. Sterilize in the autoclave at 121ºC. for 15 minutes.

4. Cool to 45-50ºC. and aseptically dispense desired volume into sterile containers.

PROCEDURE AND INTERPRETATION OF RESULTS

For information on procedures and interpretation of results, consult listed references or refer to the prepared media Instructions for Use (IFU) for Cat. No. G77.

LIMITATIONS

Refer to the document "Limitations of Procedures and Warranty" for more information.

MATERIALS REQUIRED BUT NOT PROVIDED

Standard microbiological supplies and equipment such as autoclaves, incinerators, and incubators, etc., are not provided.

QUALITY CONTROL

The following organisms are routinely used for testing at Hardy Diagnostics:

Test Organisms	Inoculation Method*	Incubation			Results
		Time	Temperature	Atmosphere	
Escherichia coli ATCC® 25922	A	18-24hr	35°C	Aerobic	Growth

* Refer to the document "Inoculation Procedures for Media QC" for more information.

USER QUALITY CONTROL

End users of commercially prepared culture media should perform QC testing in accordance with applicable government regulatory agencies, and in compliance with accreditation requirements. Hardy Diagnostics recommends end users check for signs of contamination and deterioration and, if dictated by laboratory quality control procedures or regulation, perform quality control testing to demonstrate growth or a positive reaction and to demonstrate inhibition or a negative reaction, if applicable. Hardy Diagnostics quality control testing is documented on the certificate of analysis (CofA) available from Hardy Diagnostics Certificate of Analysis website. Also refer to the document "Finished Product Quality Control Procedures," and the CLSI document M22-A3 *Quality Assurance for Commercially Prepared Microbiological Culture Media* for more information on the appropriate QC procedures. See the references below.

PHYSICAL APPEARANCE

CulGenex™ LB Agar, Lennox powder should appear homogeneous, free-flowing, and light beige in color. The prepared media should appear slightly opalescent, and light amber in color.

REFERENCES

1. Anderson, N.L., et al. *Cumitech 3B; Quality Systems in the Clinical Microbiology Laboratory*, Coordinating ed., A.S. Weissfeld. American Society for Microbiology, Washington, D.C.

2. Ausubel, F.M., R. Brent, R.E. Kingston, D.D. Moore, J.G. Seidman, J.A. Smith, and K. Struhl. 1994. *Current Protocols in Molecular Biology* . Vol. 1. Current Protocols, New York, N.Y.

3. Bertani, G. 1951. Studies on Lysogenesis. I. The Mode of Phage Liberation by Lysogenic *Escherichia coli* . *J. Bacteriol.* ; 62:293-300.

4. Tille, P., et al. *Bailey and Scott's Diagnostic Microbiology,* C.V. Mosby Company, St. Louis, MO.

5. Lennox, E.S. 1955. Transduction of Linked Genetic Characteristics of the Host by Bacteriophage P1. *Virology* ; 1:190.

6. Luria, S.E., and J.W. Burrous. 1957. Hybridization Between *Escherichia coli* and *Shigella* . *J. Bacteriol.* ; 74:461-476.

7. Luria, S.E., J.N. Adams, and R.C. Ting. 1960. Transduction of Lactose-Utilizing Ability Among Strain of *E. coli* and *S. dysenteriae* and the Properties of the Transducing Phage Particles. *Virology* ; 12:348-390.

8. Miller, J.H. 1972. *Experiments in Molecular Genetics* . Cold Spring Harbor Laboratory, Cold Spring Harbor, N.Y.

9. Sambrook and Russell. 2001. *Molecular Cloning: A Laboratory Manual* , 3rd ed. Cold Spring Harbor Laboratory. Cold Spring Harbor, N.Y.

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Distribution Centers:
California · Washington · Utah · Arizona · Texas · Ohio · New York · Florida · North Carolina

The Hardy Diagnostics manufacturing facility and quality management system is certified to ISO 13485.

HDQA 2207C [D]